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## **PROTOZOA**:

### SPOROZQA.

#### BY

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## AUTHOR'S PREFACE.

THE present volume, dealing with Sporozoa, follows the same plan as the author's volume on Ciliophora, published two years ago. The group Sporozoa includes numerous organisms, all of which are parasitic in hosts belonging to a number of different phyla of the Animal Kingdom. Several of these organisms are responsible for producing disease in man and domestic animals, and consequently come within the ken of the medical and veterinary workers : but there are numerous others which parasitize animals such as earthworms, polychætes, crustaceans, insects, myriopods, arachnids, molluses, fishes, amphibians, reptiles, birds, etc., and, though causing damage to various tissues of the host, are on the whole tolerated by them. To the student of Protozoa all are equally interesting. and it is his province to study them as animals, and name and classify them. The medical and veterinary workers will find in this volume the correct zoological position and nomenelature of the pathogenic organisms they study. With regard to the nomenclature, the synonyms recorded under each species will serve to indicate the extent to which opinion has in the past varied with regard to the systematic position of the particular organisms. In a few cases the reader may feel disposed to differ from me, but in such cases he will find the argument or the authorities on which I have relied clearly stated, and if, after considering them, the reader still differs, he is welcome to do so.

The most approved and up-to-date system of classification has been followed, and in the Identification Tables of Families I have included those families which are at present not known

from India. The 320 species described in this volume are but a small fraction of the total known from other parts of the world. All the species that are as yet known from India, Burma, and Cevion have been brought together, but a large number still await discovery. The geographical distribution of the parasites usually follows that of their hosts. Lists of Parasites and their Hosts, and of Hosts and their Parasites have been given in the Introduction and the Appendix, and it is hoped that these will be of use to those looking for the parasites in particular hosts. It may be pointed out, however, that the discovery of a parasite in a host not recorded in the list does not mean that the organism is new to science. It may already have been recorded from the same host or in some other host in other parts of the world. Keeping this in view, the discoverer should not rely too much on this volume, but should earefully explore the monographs and original papers to which the Bibliography, given at the end of the volume, will furnish a guide. The literature on the subject of Sporozoa is very vast, and it has been possible to give a selection only. For the convenience of the reader the Bibliography has been divided into a number of sections corresponding to the subclasses or orders, and if a reference is not found under "Textbooks and Sporozoa in General" it should be looked for in the section of the Bibliography relating to the particular order.

The species of each genus have been arranged in alphabetical order, except in a few cases where the species occurring in man or some other group of hosts have been brought together. In the synonymies, given under each species, references to all the records from India, Burma, and Ceylon have been included, and a  $\dagger$  mark prefixed to all such references. A selection of other references which are considered important or useful is also given.

In the Introduction I have given a short account of the general organization and structure, and discussed the phylogeny and classification of the group. It seems obvious that Sporozoa should preferably be divided into two classes, viz., Sporozoa s. str. and Amœbosporidia, but the two classes

cannot be arranged in a subphylum, as the former are evolved from the Flagellata and the latter from the Sarcodina : so in deference to the usage among English authors, the group is described as a single class and is subdivided into a number of independent subclasses. A short account of the principal methods employed in the study of Sporozoa has also been given.

A volume such as this is bound to incorporate very largely the work of others, and my grateful acknowledgements are due to all those whose works have been drawn upon, and especially to Minchin, Wenyon, Hartmann, Reichenow, Calkins, Hegner, and Kudo. Where available, figures have been given for the species dealt with. A certain number of these are taken from my own work, but a large number have been borrowed with the kind permission of the authors or publishers concerned. My thanks are due to the editors and publishers of journals and text-books who have given permission to reproduce the figures, and due acknowledgement is made in every case by giving the name of the author from whom the figure has been copied.

My special thanks are due to Dr. B. Prashad, Director of the Zoological Survey of India, for special facilities given to me on the occasion of several visits to Calcutta to consult the literature in the splendid library maintained by the Zoological Survey of India; and also to Dr. S. L. Hora for his help in getting some figures copied under his supervision by the artists working under him. Finally, I have to offer my most grateful thanks to the Editor, Lieut.-Colonel R. B. Seymour Sewell, C.I.E., F.R.S., for a thorough and critical revision of the text, and for generous help and guidance during the production of this work.

B. L. BHATIA.

Hotu Singh Road, Lahore, July, 1938.

### GLOSSARY OF TECHNICAL TERMS.

- Acephaline.—A gregarine not possessing an epimerite at any stage of its life-history.
- Acnidosporidia.—Term sometimes used to include Sarcosporidia and Haplosporidia.
- Actinomyxidia.—Cnidosporidia with large spores, trivalved membrane, and three distinctly visible polar capsules.
- Adeleidea.--Coccidia in which the gametocytes are dissimilar in size and are associated with each other during the later part of trophic life.
- Agamete.---An agamic spore or product of asexual reproduction.
- Agamogony.—Asexual or agamic reproduction by equal, unequal or multiple division.
- Agamont.--An asexual individual reproducing without conjugation or fertilization.
- Amitotic division.—Direct division of the nucleus unaccompanied by the formation of a spindle of threads.
- Amæbosporidia.—Term used by Hartmann to denote a separate class of Protozoa, in which are included Cnidosporidia, Sarcosporidia, and Haplosporidia.
- Amæbula stage.-The sporoplasm which, by amæboid movements, has left the spore membrane, a stage leading up to the schizont.
- Anisogamy .--- Conjugation between dissimilar gametes.
- Anterior end.—The end of an organism which is habitually forward in locomotion. In Cnidosporidia the end of the spore from which the polar filament becomes extruded through the foramen. If the two extremities are dissimilar in form, the anterior end is usually more or less attenuate.
- Association.--A group formed by the attachment of two or more sporonts.
- Autogamy.-Fusion of the two daughter nuclei to form a zygote or sporont.
- Biassociative.—Referring to an association of two sporonts attached by unlike ends.
- Binary fusion.—A mode of reproduction in which the division of the nucleus into two is followed by the division of the cell.
- Budding.—The process of unequal fission, resulting in the formation of daughter organisms, which show a simplified structure when first formed.

- Cephaline.—A gregarine possessing an epimerite at some stage in its life-history.
- Cephalont.--Young gregarine with an epimerite.
- Chitinous.—Corresponding in appearance and character to chitin, the horny material which forms the protective covering of insects and other Arthropoda.
- Cnidosporidia.---Sporozoa in which the spore is provided with one or more thread-capsules.
- Coccidia.—Telosporidia in which the mature trophozoite is intracellular and small, the zygote is non-motile, and the sporozoites are developed within a spore.
- Coccidiomorpha.--Term used by Doflein to include Coccidia and Hæmosporidia.
- Calozoic.—Parasites that live in the lumen of the alimentary canal or other cavities in the body of the host.
- Conjugation.—Union of two organisms leading to reproduction by germs or spores.
- Cortical.-Relating to the external layer of an organism.
- *Cyst.*—Impervious membrane surrounding an organism or a pair of associated sporonts at the beginning of reproduction. It is an adaptation to a change of hosts in parasitic forms.
- *Cytoplasm.*—The protoplasm of the cell-body as contrasted with that of the nucleus.
- Cytozoic.--Parasites that are lodged inside a cell.
- Deutomerite.--The portion of a cephaline gregarine behind the septum, by which it is separated from the protomerite.
- Dicystid.---A gregarine in which the trophozoite is divided into two parts only, that is, an epimerite is present, but the rest of the body is not divided into a protomerite and a deutomerite.
- Disporoblastic .- Producing two sporoblasts.

Disporous.-Producing two spores.

Ectoparasitic.---Having the nature of an external parasite.

- Ectoplasm.—The outer zone of the body of unicellular organisms, comprising the epicyte, sarcocyte, and myocyte.
- *Eimeridea.*—Coccidia in which the gametocytes are similar in size and develop independently of one another.
- Encystment.—The phenomenon of becoming motionless and excreting a membranous cyst.
- Endogenous or internal budding.—Formation of buds in the interior of the cytoplasm of the parent.
- Endoparasitic.-Having the nature of an internal parasite.
- Endoplasm.—The inner or granular zone of the body of unicellular organisms, lying within the ectoplasm.
- Endospore.—The inner covering of a spore.

Epicyte.-The thin, fragile, external layer of the ectoplasm.

- *Epimerite.*—The temporary, or rarely permanent, structure at the anterior end of a gregarine by which the young gregarine is attached to the host-cell. It is epicytal in origin.
- Epispore.-The outer covering of a spore.

Eugregarinaria.---Gregarinida not showing schizogony.

- Exogenous or external budding.--Formation of buds from the external surface of the body.
- Fission.—Division of the nucleus followed by a division of the cell-body.
- Gamete.--Specialized cells destined to meet and fuse in conjugation.
- Gametocyst.-The cyst formed round two associated sporonts or gametocytes.
- Gametocyte.-The mother-cell which gives rise to a number of gametes.
- Gamogony.—The process of production of gametocytes or gametes by a gamont.
- *Hamont.*—An individual destined to form gametes; also known as sporont.
- Globidia.—Sarcosporidia in which the spores are fusiform and cysts occur exclusively in the intestinal submucosa.
- Golgi apparatus.—A cytoplasmic inclusion which shows a tendency to clump together in masses or to form a network in the neighbourhood of the nucleus.
- Gregarinida.—Telosporidia in which the mature trophozoite is extracellular and large, the zygote is non-motile, and sporozoites are developed within a spore.
- *Gymnospore.*—Naked germ or protoplasmic hody, formed by sporulation, which is not enclosed in a protective envelope.
- Hamosporidia.—Telosporidia in which the mature trophozoite is intracellularand small, the zygote motile, and sporozoites are without an envelope.
- Hæmosporidiidea.—Term used by Wenyon to denote Hæmosporidia which form pigment in the red blood-cells.
- Haplocyta.—Gregarines in which the trophozoite is not divided by an ectoplasmic septum.
- Haplosporidia.--Sporozoa characterized by possession of large spores, and a simple type of development.
- Helicoeporidia.—Cnidosporidia with small barrel-shaped spores, containing a thick filament coiled beneath the spore-membrane and three sporoplasms.
- Heteropolaridea.-Haplocite, producing sporocysts with dissimilar poles.
- Histozoic.--Parasites that occur in the spaces between groups of cells.
- Holozoic.—Animals which are entirely dependent for food on other organisms, which they capture, devour and digest.
- Homopolaridea.---Haplocyta producing sporozysts with similar poles.

Isogametes.-Gametes which are similar in shape and size.

Isogamy.--Conjugation between similar gametes.

Karyogamy.—Union of two gamotes whose nuclei undergo intermingling. Karyosome.—A chromatic mass surrounded by plastin and contained within the nucleus.

Longitudinal striations.—The very delicate ridges on the outside of the epicyte.

- Macrogamete.--The larger or inactive gamete in anisogamous conjugation.
- Macrogametocyte.-The mother-cell of the macrogamete.
- Meront .--- A schizont of a microsporidian.
- Merozoite .-- A product of asexual reproduction or schizogony.
- Metabolic .--- Changeable in form ; polymorphic.
- Metagamogony.—The process of zygotic or post-conjugation reproduction. Metamorphic.—Changeable in form.
- Microgamete.--The smaller or active gamete in anisogamous conjugation.
- Microgametocytes.-The mother-cell of microgametes.
- Microsporidia.—Cnidosporidia with small spores, membrane in one piece, and one or, rarely, two polar filaments that are invisible in vivo.
- Microsporoblastic.--Producing a variable number of sporoblasts.
- Microsporous .--- Producing a variable number of spores.
- Mitochondria.---Minute cytoplasmic inclusions, of a lipoidal nature, occurring in the form of spherical granules or rod-shaped or crescentic bodies.
- *Mitotic.*—Indirect division of the nucleus, which is accompanied by the formation of a spindle of threads.
- Monocystid.—Gregarines in which the body of the trophozoite consists of a single part, *i. e.*, is not divided by a septum.
- Monosporoblastic .-- Developing into a single sporoblast.
- Monosporous.-Developing into a single spore.
- Multinucleate .- Possessing many nuclei.
- Multiple fission.—A mode of reproduction in which the division of the nucleus is not immediately followed by the division of the cell, but, after repeated nuclear division, the cell divides into as many parts as there are nuclei.
- Myocyte.—The ectoplasmic layer consisting of the myonemes.
- Myoneme.—Specialized muscle-like fibrils which cause the contraction of the whole or a part of the body. They are embedded in the periphery of the endocyte and form a network around the organism.
- Myxosporidia.—Cnidesporidia with large spores, bivalved membrane, with two or four polar capsules visible in vivo.
- Neosporidia.—Term used by Schaudinn to include Chidosporidia, Sarcosporidia, and Haplosporidia. The common features are that the life of an individual does not come to an end when reproduction takes place, but that reproduction continues throughout the trophic phase, the sporoblasts being carried about by the more or less active parent organism, which may ultimately become a large mass of spores.
- Nucleolus.—An exceedingly minute, more solid particle developed singly or in varying number within the nucleus of an animal or vegetable cell. Its homologue among the Protozoa is generally referred to as Endoplastule or Karyosome.
- Nucleus.—More densely granular body within the substance of an animal or vegetable cell.

Octosporoblastic .- Producing eight sporoblasts.

Octosporous.-Producing eight spores.

Octozoic spore.- A spore containing eight sporezoites.

Oöcyst .-- A cyst containing the conjugated gametes.

Oökinete.-The motile zygote in Hæmosporidia.

- Pansporoblast.—An enclosed area in a myxosporidian trophozoite in which two sporoblasts become differentiated. The term is also used to designate in general a grown-up sporont of the polysporous genera in which two or many sporoblasts are formed.
- Parasite.—An organism living in or upon the body of another organism and dependent for its existence on that organism or a limited group of organisms.
- Piroplasmidea.—Hæmosporidia which do not form pigment in the red blood-cells.
- Planont.—The stage between free amœbula and schizont stages, which are found in the alimentary canal or hody-cavity of the host soon after the spore gorminates.
- Plasmodium.--Multinucleated cell formed by the repeated divisions of the nucleus.
- Plasmotomy.—Cleavage of a multinucleated body into two or more multinucleate parts.
- Polar capsule.—A sac in which the polar filament is coiled, a structure characteristic of a enidosporidian spore.
- Polar filament.—A fine and long filamont coiled in the polar capsule, which is extruded under suitable stimulation.
- Polycystid.—Referring to gregarines possessing a septum which divides the trophozoite into regions; also known as "septate."

Polymorphic .--- Exhibiting a diversity of form.

Polysporoblastic .-- Producing numerous sporoblasts.

Polysporous .- Producing many spores.

- Posterior end.—The cnd of an organism which is habitually behind in locomotion. In Cnidosporidia the end of the spore opposite to the anterior; it is usually more or less rounded if the two ends are dissimilar.
- Primite.-The first individual in an association of two or more sporonts.
- Protomerite.-The portion of a septate gregarine which precedes the septum.
- Protoplasm.—The physical basis of life, or elementary formative matter of all living organisms.

Protozoa .--- Animals in which the body is not divided into cells.

Pseudocyst.—The residual protoplasm which, after the spores are separated, acquires a membranous wall, swells until the true cystwall bursts, and allows the extrusion of the ripe spores.

Sarcocyte .-- The middle layer of the ectoplasm.

Sarcosporidia.—Sporozoa in which cysts form long rod-like masses, and the spores are crescentic.

- Satellite.—Any sporont in an association which is attached behind the primite. Generally there is one, but sometimes several are attached linearly, one behind the other, or in a cluster to the posterior end of the primite.
- Schizogony.—Asexual or agamic reproduction by equal, unequal or multiple division.
- Schizogregarinaria .--- Gregarinida showing schizogony.
- Schizont.—That stage which is about to divide into a number of parts called merozoites.

- Schizontocyte.—A special form of agamont which breaks up into a number of agamete-forming centres by multiple division.
- Septata.---Gregarines in which the trophozoite is divided by an ectoplasmic septum.
- Septum.—The thin layer of sarcocyte which separates the protomerite from the deutomerite.
- Siliceous.--Partaking of the nature and qualities of silica; composed of flint.
- Spore.—The body into which the zygote develops after the acquisition of a resistant outer coating.
- Spore-duct.—A tubular growth from the cyst, through which the spores are extruded when ripe.
- Sporoblast.—A product of the initial reproduction of the zygote, including both capsule and contents; a cell which develops directly into a spore.
- Sporocyst.—The covering or coverings of the spore. Sometimes used in the same sense as a spore.

Sporogony.-The development of spores from the sporont.

- Sporont.—An adult sporozoan which is destined to form gametes or to give rise to sporoblasts.
- Sporoplasm.—The protoplasmic mass inside the spore; the sporozoite of a Cnidosporidian spore.
- Sporozoa.—A class of the Protozoa in which spore-formation is the prevailing mode of reproduction.
- Sporozoite.—Each of the small falciform bodies which are released when the spore-wall is absorbed.
- Sporulation or multiple fission.—Mode of reproduction in which the repeated division of the nucleus is followed by the splitting of the organism into as many parts as the nuclei.
- Syngamy.—Sexual union or conjugation involving a complete fusion of two gametes.
- Synkargon.—The combination nucleus resulting from the fusion of two nuclei derived from two individuals.
- Syzygy.—Linear chains formed by two or more organisms attached end to end.
- Telosporidia.—-Sporozoa with trophic and reproductive phases typically distinct. Term used by Schaudinn to include Gregarinida, Coccidia, and Hæmosporidia.

Tetrasporoblastic .-- Producing four sporoblasts.

Tetrasporous.-Producing four spores.

Tetrazoic spore.--- A spore containing four sporozoites.

Trophozoite .--- The young feeding and growing parasite.

Vacuolate.-Having a number of clear spaces or vacuoles.

Vermicular .-- Resembling a worm in shape.

Zygote.-The cell resulting from the complete fusion of two gametes.

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### SPOROZOA.

#### INTRODUCTION.

#### Position of Sporozoa in the Animal Kingdom.

SPOROZOA are a class of parasitic organisms belonging to the phylum PROTOZOA. In the PROTOZOA the body usually consists of a single undivided mass of protoplasm, and these organisms are consequently described as unicellular or noncellular. In other classes of the PROTOZOA the animals, which may be free-living or parasitic, move by characteristic organs of locomotion such as pseudopodia, flagella or cilia: but the class SPOROZOA includes organisms which are entirely parasitic, and as a rule are incapable of free locomotion. Some of them, when immature, move about by means of pseudopodia, but they never possess cilia or flagella. All are characterized by producing SPORES.

Our knowledge of the SPOROZOA commenced just over a hundred years ago. In 1826 Dufour gave the first detailed account of an organism, which he afterwards, in 1828, named Gregarina. He found it in the alimentary canal of several species of COLEOPTERA, and later in the gut of the earwig. Both he and Siebold, who investigated the genus in 1837 and 1839, failed to recognize it as a Protozoan. Schleiden, Henle, and others recognized the unicellular nature of Gregarina, but regarded it as an overgrown plant-cell, and it was only as late as 1848 that Kölliker demonstrated the Protozoan nature of the genus, described the formation of pseudonavicellæ, and inferred that they represented one method of propagation, a view that was confirmed the same year by Stein. The complete life-cycle of a Gregarine was worked out much later by Siedlecki (1899).

In 1839 Hake published the earliest account of a Coccidian. He described and figured the occysts of Eimeria stiedæ of the rabbit's liver, but did not interpret them as of parasitic origin. Important observations on a Coccidian were published by Kloss (1855), who described the organism now known as Klossia helicina in the renal organ of the snail. Eimer (1870) described parasites of the gut and the liver of various animals, including man, which have since been referred to the genera SPOR. R

*Eimeria* and *Isospora*. Pfeiffer (1892) was the first to describe the theory of alternate generations, and Schaudinn and Siedlecki (1897) showed that there are two kinds of gametes in *Coccidium*, and that the macro- and microgametes conjugate to form zygotes. Later (1898) Siedlecki published an account of the sexual cycle of the Coccidian *Aggregata*, and in 1899 gave a detailed account of the entire life-history of *Adelea*. This series of classical researches led up to the publication of the description of the complete life-cycle of *Coccidium schubergi* by Schaudinn (1900).

The HEMOSPORIDIA, or blood-inhabiting SPOROZOA, were discovered as late as the eighties of the last century. Laveran observed the malarial parasites in the human blood in 1880, and published a description in the following year. He observed the amœboid, rosette, sphere, crescent and the flagellate stages, but did not determine the relationship of the various stages, nor did he recognize the animal nature of the parasites. The genus Plasmodium was instituted by Marchiafava and Celli (1895). Further discoveries leading to the elucidation of the complete life-cycle of the malarial parasites were made by Metchnikoff (1887), Golgi (1889), Danilewsky (1891), Manson (1894, 1896), MacCallum (1897, 1898), Ross (1898, 1899), Grassi (1898), and others, and will be referred to elsewhere in this volume. Smith and Kilborne (1893) demonstrated that Babesia, the cause of Texas fever of cattle, was transmitted from host to host by ticks, and were thus the first to show the important part played by insects in the transmission of parasitic PROTOZOA.

We owe the term SPOROZOA to Leuckart (1879), who introduced it to include the GREGARINIDA and the COCCIDIA. The HEMOSPORIDIA were included later, when the study of the complete life-cycle showed the close resemblance of various stages with those of the COCCIDIA. Besides these three well recognized groups, there are other groups of parasites, found in Fishes, Arthropods, etc., which, on account of their forming spores, came to be included in the SPOROZOA. In a large majority of these, known as the CNIDOSPORIDIA, the spore contains a coiled filament inside a polar capsule. Schaudinn (1900) divided the SPOROZOA into two subclasses-(i) the TELOSPORIDIA, to include the GREGARINIDA, COCCIDIA, and HAEMOSPOBIDIA; and (ii) the NEOSPORIDIA, to include the CNIDOSPOBIDIA, SARCOSPORIDIA, and HAPLOSPORIDIA. The members of both these subclasses produce resistant spores, but according to many authors the affinities between the two are not sufficiently close to justify their inclusion in the same Hartmann (1907) established two classes, for which class. he employed Schaudinn's names-TELOSPORIDIA and NEO-SPOBIDIA. Later, Prowazek and Jollos (1922) and Hartmann

(1923-5) designated the two classes as Sporozoa and AMCEBOSPOBIDIA, and something can be said in favour of this system of classification. Wenyon (1926) also divided the entire group into two classes, and named them as SPOROZOA and CNIDOSPORIDIA, leaving the SAROOSPORIDIA and HAPLOSPORIDIA as parasites of undetermined position. Theoretically speaking, there is no difficulty in restricting the term SPOROZOA to the original connotation which Leuckart gave it, but in practice it would seem impossible so to restrict it, unless a new term like SPOROGENEA is coined for a subphylum to include the two classes. In view of the term SPOROZOA having become fixed in its extended usage, and the phylogeny of the two groups being altogether hypothetical. Reichenow (1929, 1935) divided the class SPOROZOA into four distinct subclasses, viz., TELOSPORIDIA, CNIDOSPORIDIA, SARCOSPORIDIA, and HAPLOSPORIDIA. Similarly Kudo (1931) and Calkins (1933) divide the class SPOROZOA into three subclasses, TELOSPORIDIA, CNIDOSPORIDIA, and ACNIDO-SPORIDIA, combining in the last the SARCOSPORIDIA and HAPLO-SPORIDIA. The balance of opinion at the moment does not seem to favour the recognition of only two classes, but, if this were done, they would be named SPOROZOA (sensu stricto) and AMEBOSPORIDIA. For practical convenience, and in deference to general practice, I shall follow Reichenow (1929, 1935) in dividing the class SPOROZOA (sensu lato) into four subclasses.

The phylum PROTOZOA may thus be divided as follows :---

A. Subphylum PLASMODROMA Doflein, 1901, emended.

Movement effected by pseudopodia or flagella, and syngamy takes place, in all known cases, by the complete fusion of gametes.

- I. Class MASTICOPHOBA Diesing, 1865.
  - The predominating phase flagellate, locomotion being effected by filamentous whip-like structures called flagella. The body may be corticate or non-corticate.
- II. Class RHIZOPODA von Siebold, 1845 (=SARCODINA Hertwig & Lesser, 1874).

The predominating phase amœboid, locomotion being effected by temporary extensions of the body called pseudopodia. The body is non-corticate, *i. e.*, has no tough limiting membrane or cuticle.

III. Class SPOROZOA Louckart, 1879.

Exclusively parasitic forms which lack definite organs of locomotion. Reproduction takes place by spore-formation.

(a) 1. Subclass TELOSPORIDIA Schaudinn, 1900 (= SPOROZOA sensu stricto).

Trophozoite becomes full grown before reproduction begins; spore aimple, with one to several sporozoites or without resistant envelope; the sporozoite a gregarinula; asexual and sexual reproduction alternate regularly. (b) NEOSPORIDIA Schaudinn, 1900 (=AMŒBOSPORIDIA Hartmann).

Trophozoites may begin to form spores when still growing or even quite young; the sporoblasts are formed by a process of internal gemmation; the sporozoite is an amœbula; and the life-cyole is passed in as ingle host. This group includes the remaining three subclasses.

- 2. Subclass CNIDOSPORIDIA Doflein, 1901. Spore with polar filament which is typically coiled within a polar capsule.
- 3. Subclass SARCOSPORIDIA Balbiani, 1882.

Spores crescentic, without thread capsules. Cysts forming long rod-like masses. Parasites of striped muscles of Vertebrates.

4. Subclass HAPLOSPORIDIA Caullery & Mesnil, 1905.

Spores large, containing a single voluminous nucleus; no thread-capsules. Type of development simple.

B. Subphylum CILIOPHORA Doflein, 1901.

Movement effected by cilia.

IV. Class CILIATA Perty, 1852. Organisms bear cilia throughout life.

- V. Class Suctoria Claparède & Lachmann (= Tentaculifera Huxley, Acinetaria Lankester).
  - Ciliated in the young stages, but later usually attach themselves to other objects, lose their cilia, and develop knobbed tentacles which serve as sucking tubes.

#### General Organization and Structure.

This volume deals with the class SPOROZOA as defined above, and I give below a brief survey of the general organization and structure of the organisms included in this group, so as to give the reader a general idea of the group and to introduce him to the principal technical terms employed in the description of the forms.

Modes of Life.--All SPOROZOA are obligatory parasites, and there are no free-living forms among them. Parasitic forms are also found among other classes of the PROTOZOA, but these may be regarded as free-living forms which have been introduced casually into the body of their host and have become adapted to a parasitic mode of life. In SPOROZOA the dependance has reached an extreme limit, and the parasites have no existence apart from the hosts in which they are found to occur. The transference of the parasite from one host to another is effected by means of SPORES, which may be defined as resistant seed-like bodies, containing one or more sporozoites or germs and protected by a firm envelope or capsule. The spore is a contrivance to enable the parasite to withstand the vicissitudes of the outside world until they pass again into the body of a suitable host. Once inside the body of the new host, the spore germinates and a fresh infection is

started. The mode of transfer of the spores is usually contaminative, that is to say, the spores are ingested with the food that has been contaminated with fæces in which the spores have been passed. In some forms, however, where the parasite passes part of its life-cycle in the blood of one host and part in the body of a widely different host, the sporozoite is not enclosed within a definite membrane or cyst, and the method of transfer is inoculative.

The method of nutrition is by osmosis only. The organism does not possess pseudopodia, flagella or cilia for the purpose of food-capture. Where the organism is amæboid, its pseudopodia are for the purpose of increasing the extent of the bodysurface for absorption, rather than for the ingestion of solid particles of food. The parasite may invade the cells (cutozoic), or spaces between the cells (histozoic), or may live in the lumen of the alimentary canal or other eavities in the body of the host (cælozoic). The food material absorbed from the host will thus be dissolved cytoplasm, tissue fluid, body fluid, or digested food material from the alimentary canal of the host. The SPOROZOA are parasitic in animals of almost every phylum from PROTOZOA to CHORDATA, and while many of them have come to be tolerated, others are responsible for causing deadly disease and heavy mortality among the hosts. Ĩn accordance with their widely varied habitat, they show manifold adaptations suiting them to the highly specialized conditions of their existence.

Form and Structure.-The SPOROZOA show a more or less complicated life-history, consisting of various stages. The starting point is the minute germ or sporozoite, which may have one of two forms. In AMŒBOSPORIDIA it is a minute amœboid organism termed an amœbula; in the TELOSPORIDIA it is more definite in form, being a rod-like or sickle-shaped body ("falciform body") which is capable of twisting or bending movements and progresses by gliding, and is described as a gregarinula. The sporozoite, after being set free in the body of the new host, nourishes itself and grows, often to a relatively large size, at the expense of the host. This is the trophic phase, and the organism during this phase is described as the trophozoite. The trophozoite absorbs nourishment in the fluid state, and does not exhibit any organs of locomotion, ingestion or digestion; neither food-vacuoles nor contractile vacuoles are present. The parasite has usually a fixed form with definite contours, being limited externally by a cuticle of greater or less thickness, and may either grow inside a host cell or be attached to a cell of the host by a special organ of fixation known as an epimerite. The ectoplasm is in certain groups differentiated into three layers, known as epicyte, sarcocyte, and myocyte, the latter being composed of myonemes

#### SPOROZOA.

or contractile filaments which bring about contortions of the body. The sarcocyte may run inward as one or more septa and thus divide the body into a number of apparent segments : in the cephaline Gregarines (fig. 30) the body is thus seen to consist of three segments, known respectively as the *epimerite*, *protomerite*, and *deutomerite*. Sometimes the organisms may attach themselves to each other and form clusters or linear chains, termed syzygies. The *endoplasm* is usually granular, and contains sorted-up food material in various forms. The *nucleus* is usually very large, spherical, and vesicular in type, with one or more distinct karyosomes.

Reproduction.-Like other parasites, SPOBOZOA possess the power of prolific multiplication as a necessary adaptation for the maintenance of the species. Both asexual and sexual methods are known in all groups. Asexual reproduction may be by binary fission (as in Babesia), by multiple fission (as in COCCIDIA and HÆMOSPORIDIA), or by budding, which may be exogenous (as in MYXOSPORIDIA) or endogenous (as in certain Schizogregarines). The trophozoite when it is about to enter on asexual reproduction is known as a schizont (or agamont), the process being termed schizogony (or agamogony) and the resulting products of division being termed merozoites. Sexual reproduction (syngamy) is by isogamous or anisogamous conjugation or sometimes by the fusion of sister individuals derived by fission of the same parent cell or nucleus (autogamy). Trophozoites which associate with one another for the purpose of sexual reproduction are designated as sporonts or gametocytes, which may differ from one another and are then known as microgametocytes and macrogametocytes, according as they give rise to males or *microgametes* and females or *macrogametes*.

The fusion of two gametes results in the formation of a zygote which, when actively motile and vermiform (as in HÆMOSPORIDIA), is generally known as an oökinete. The zygote may secrete a distinct membrane round itself and become a passive spherical body known as an oöcyst. Cellular division of the protoplasm within the opeyst gives rise to the formation of a number of sporoblasts, each of which becomes surrounded by a wall, the sporocyst, and is then known as a spore. Inside a spore are developed a smaller or larger number of minute germs or sporozoites which are the infective bodies, and start the cycle again. These various stages, starting from the zygote and leading to the formation of the spores and sporozoites, constitute sporogony. Asexual and sexual methods of reproduction regularly alternate in the life-cycle of the majority of forms, and complicated life-histories result. In such cases schizogony takes place usually in the body of one host and gametogony and sporogony in the body of an animal belonging to a widely different group. The former

is described as an *intermediate host*, and the latter, in which the sexual process is perfected, as the *final* or *definitive host*.

Life-history.—The life-histories of different groups vary a good deal, but there is always some resemblance to a common fundamental type. The life-history of the Coccidium Eimeria stiedæ (Lindem.) (fig. 1), occurring in the liver and intestine



Fig. 1.— Life-cycle of a typical coccidium, *Eimeria stiedæ* (Lindem.). 1-5, sporogony; 6-15, schizogony; 17-20, gametogenesis; 21-23, fertilization. (After Reich).

of the rabbit, may be described as typical. The sporozoite (6) escapes from the spore in the intestine of the host, and, gliding like a minute Gregarine, bores its way into the epithelial cell (7) and becomes a trophozoite (8). The latter feeds at the expense of the cytoplasm of the host-cell and grows into a large spherical schizont with a vesicular nucleus containing a large karyosome. The nucleus of the schizont divides repeatedly until sixteen or thirty-two daughter nuclei are produced (9, 10), and the schizont breaks up into an equal number of merozoites (11). The merozoite may attack another epithelial cell and undergo schizogony again, or, on entering a fresh epithelial cell, may produce a set of only four merozoites (13-15), which differ from the others in possessing flagellum (16). The flagellated merozoites penetrate other epithelial cells and become either macrogametocytes (17 a) or microgametocytes (17 b). The macrogametocyte grows into a large oval body containing many chromatinoid and plastinoid granules, which gather round the periphery to form a membrane (17 a-20 a), and thus gives rise to a single macrogamete (21). The microgametocyte becomes large and spherical, and within it are formed a large number of biflagellated microgametes (17 b-20 b). Fertilization (21) takes place either in an epithelial cell or in the lumen of the intestine. The resulting zygote (22, 23) passes out of the intestine as an occyst in the faces (1). The protoplasm within the occyst contracts into a spherical mass (2), and then divides into four spherical sporoblasts (3), which become ovoidal (4), secrete a sporocyst, and become spores, part of the cytoplasm not being used up in the process. Within each spore two sporozoites develop (5), again leaving a small amount of residual protoplasm. Fresh infection takes place through the food becoming contaminated with faces containing these spores.

While the above life-history may be taken as typical, considerable variations are met with in different groups. In the majority of SPOBOZOA the hosts are of the same species, and infection is brought about by eating contaminated food; but in many forms there is an alternation of hosts, schizogony taking place in an animal of one group and gametogenesis and sporogony in a host belonging to an entirely different group. Thus in the case of *Aggregata* asexual reproduction takes place in a Crustacean (crab) and sexual reproduction in a Cephalopod (cuttle-fish). Among the HÆMOSPORIDIA schizogony takes place in the blood of a Vertebrate (man, other mammals, or various birds), while sexual reproduction, followed by sporogony, occurs in an Insect (mosquito). In such cases spore-capsules are not formed, as the sporozoites are introduced directly into the blood by the mosquito.

In the Eugregarines the asexual cycle is entirely wanting, the sporozoite developing directly into a gametocyte. Two individuals lie side by side, and an envelope or gametocyst is secreted enclosing both individuals. Each individual then forms a large number of gametes, those from one individual fusing with those from the other, and a corresponding number of zygotes are formed. Within the membrane the zygotes develop into spores, each dividing to form a definite number of sporozoites.

Among the AMŒBOSPORIDIA (=NEOSPORIDIA) the life-cycle is less complicated, and sexual dimorphism and change of hosts are not met with. Reproduction takes place more or less continuously throughout the trophic phase, the organism ultimately becoming a huge mass of spores. Spore-formation is also on an entirely different plan from that in the TELOSPORIDIA, and does not usually result from the divisions of a zygote. The life-histories among the subclasses CNIDOSPORIDIA. SARCOSPORIDIA, and HAPLOSPORIDIA differ considerably, and it is impossible to attempt a generalized account : reference must therefore be made to the detailed accounts given under these different subclasses. Among the CNIDOSPORIDIA both schizogony and sporogony are met with. Schizogony is carried out by binary or multiple fission, by budding, or by the cleavage of the multinucleated plasmodium into two or more multinucleate parts (plasmotomy). Isogamous, anisogamous, and autogamous reproduction have been reported in a number of forms, and the zygote becomes the sporont, in which one to many spores become differentiated. The spores are of a unique structure. Each spore possesses one to four polar capsules, each containing a coiled polar filament, and one to many sporoplasms. The mode of development of these spores differs in different cases. In the orders MYXOSPORIDIA and ACTINOMYXIDIA several cells appear during the process. These cells give rise to one or more sporoplasms or generative cells, capsulogenous cells, and the spore-membrane. In the order MICROSPORIDIA the amæboid sporoplasm undergoes schizogonic multiplication, and the schizonts later become sporonts, each producing the characteristic number of spores.

The SARCOSPORIDIA produce long rod-like masses of spores among the muscle-fibres, which they parasitize. The spores are crescentic in outline and do not contain polar capsules. Only portions of the full life-cycle are known.

The HAPLOSPORIDIA show a comparatively simple life-cycle. The spores are spherical or ellipsoidal, with a single large nucleus and no polar capsule. The amœbula grows, and by the repeated division of the nucleus forms a plasmodium. The plasmodium may divide (*plasmotomy*) or may produce merozoites (*schizogony*) or form spores. The spores arise either from sporoblasts, each of which gives rise to a single spore, or from pansporoblasts, which give rise to a number of spores.

#### Phylogeny and Classification.

All parasitie forms are assumed to have been evolved from free-living ancestors. The problem naturally presents itself as to what must have been the nature of the ancestral forms of the SPOROZOA and what is their relationship to the remaining classes of the PROTOZOA. The CILIOPHORA are at once excluded, as they are far too specialized, and the SPOROZOA do not show any special relationship with them. Two rival theories of Sporozoan ancestry have been put forward in the past, one claiming their descent from the RHIZOPODA, the other from the MASTIGOPHORA. The RHIZOPODA and the MASTIGO-PHORA are themselves linked together. Many RHIZOPODA show flagellated stages in their life-cycle, and there are many MASTIGOPHORA which are ameeboid. Most authorities agree with Awerinzew (1910) that an amœboflagellate type represents the primitive stock of the PROTOZOA, which gave rise to all existing groups, becoming differentiated into RHIZOPODA on the one hand and MASTIGOPHORA on the other. Bütschli (1882) was the first to advance the theory of the Euglenoid ancestry of SPOROZOA, according to which a typical Flagellate would become adapted first to a saprophytic and then to a parasitic mode of life, and thus lose the special organs of locomotion, nutrition, etc. As remarked by Minchin (1903), " an Euglena or Astasia deprived in this way of flagellum, mouth, chromatophores, stigma, and vacuoles, nutritive or contractile, would be practically indistinguishable from a simple Gregarine." The euglenoid movements of the Gregarines, and of the motile stages of other SPOROZOA, such as the sporozoites and merozoites of COCCIDIA, the free stages of the HÆMOGREGARINIDA, and the oökinetes of the HÆMO-SPORIDIA, lend support to this view, and additional support has been furnished by the discovery of flagellated stages in the life-cycle in many Sporozoan forms.

The life-cycles of the AMCEBOSPORIDIA (==NEOSPORIDIA), however, do not lend support to the above-mentioned view. They have no Euglenoid phases, do not possess flagella at any stage of their life, and are amceboid throughout their trophic phase. Thus the AMCEBOSPORIDIA give support to the rival theory of Rhizopod ancestry. It was doubtless this fundamental difference that led Schaudinn (1900) to divide the SPOROZOA into two subclasses, viz., TELOSPORIDIA and NEOSPORIDIA; and led Hartmann (1907) to regard the SPOROZOA (sensu stricto) and the AMCEBOSPORIDIA as distinct classes. Minchin (1912) expressed the opinion that from such forms as Cercomonas arose on the one hand the RHIZOPODA and their derivatives (NEOSPORIDIA) by loss of flagella and specialization of the amceboid form in the adult, and on the other the MASTIGOPHORA and their derivatives (TELOSPORIDIA, INFUSORIA) by specialization of the flagellar apparatus, combined with the acquisition of a cortex and loss of amæboid movement. Hartmann (1923-5) and Fantham (1936) have also discussed the philogeny of the SPOROZOA.

Taking the TELOSPORIDIA (=SPOROZOA sensu stricto) first, we are struck by the obvious points of difference between the GREGARINIDA and the COCCIDIOMORPHA. In the GREGARINIDA two sporonts become enveloped in a common cyst before they give rise to gametes, and the union of gametes takes place within the cyst. The cyst contains many zygotes, and each zygote gives rise to a single spore. In the Coccidiomorpha, on the other hand, the gametoeytes are more or less widely separated from one another when producing the gametes; the female gametocyte remains undivided to form a single macrogamete, which is very much larger than the microgametes ; and the zygote undergoes a process of division, giving rise to a large number of sporoblasts and sporozoites. The common ancestral type may be assumed to have been one in which the trophozoites that grew into gametocytes were separated from one another and produced their gametes separately, as in COCCIDIA; but each gametoeyte produced a number of gametes which were more or less alike, as in the GREGARINIDA.

As the GREGARINIDA came to acquire an intereellular trophic phase, the sporonts became free and motile, and it was thus possible for the gametocytes to associate and encyst together, producing their gametes in close proximity. There would be neither any difficulty for the male gametes to find the female, nor any need for increased specialization of the gametes. They would lose even the slight degree of specialization inherited from the ancestral form, with the result that they would be similar and would be produced in equal numbers bythe two gametocytes.

The COCCIDIA, on the other hand, retained an intracellular habitat for their trophozoites, and the gametocytes were widely separated when producing the gametes. The gametes thus had to seek each other, and this led to greater specialization. The male gametes became very small and very motile, and were produced in large numbers; the female gametocyte no longer divided into a number of gametes, but became a single maerogamete. After fertilization the suppressed divisions of the female gametocyte would take place in the zygote, leading to the production of a number of sporoblasts, spores, and sporozoites. The spore may be regarded as comparable to the encysted zygote of the FLAGELLATA. In the suborder ADELEIDEA, however, the gametocytes acquired the habit of association prior to gamete formation, but this led merely to a reduction in the number of male gametes produced.

The HÆMOSPORIDIA resemble the COCCIDIA very closely. Their life-cycle can be described in identical terms and the points of difference attributed to their becoming adapted to parasitism upon a special kind of cell, viz., the blood-corpuscles: the two orders are thus so closely allied that Doflein (1901) was led to consider the two as suborders of a single order, the COCCIDIOMORPHA. The HÆMOSPORIDIA exhibit an alternation of generations, the asexual process or schizogony alternating with the sexual process leading to sporogony; but an essential difference from the COCCIDIA is that there is an alteration of hosts, schizogony taking place in the blood or internal organs of a Vertebrate, and sporogony in the digestive tract or other organs of an Invertebrate. Fertilization takes place in the stomach of the Invertebrate host, which has sucked up the gametocytes from the blood of the Vertebrate. The zygote, instead of being a motionless body, is a motile vermicule (oökinete), which penetrates the wall of the stomach and forms its occvst, which increases in size with the growth of the zygote, and only persists while the zygote is producing sporozoites. The absence of spores with resistant cysts is due entirely to the fact that the parasite is always sheltered within the body of one or the other of its two hosts.

So far as the AMŒBOSPORIDIA (==NEOSPORIDIA) are concerned, their affinities are entirely with the RHIZOPODA. The bodyform of the sporozoite and the adult is that of an amœba, and no flagellated stages are known to occur. As remarked by Minchin, "the union of the TELOSPORIDIA and NEOSPORIDIA in one class—the SPOROZOA—is a quite artificial arrangement." Practical convenience and common practice alone justify their inclusion in one class.

The AMCEBOSPORIDIA comprise the three subclasses CNIDO-SPORIDIA, SARCOSPORIDIA, and HAPLOSPORIDIA. The CNIDO-SPORIDIA are a well-defined group and are characterized by the spores possessing the polar capsules. Laveran and Mesnil (1899) described in the spores of *Sarcocystis tenella* a striated structure representing a polar capsule, and Minchin (1912) was led to include SARCOSPORIDIA among the CNIDOSPORIDIA. Later authorities have shown that among the SARCOSPORIDIA there is nothing corresponding to the polar capsule, so the CNIDOSPORIDIA, SARCOSPORIDIA, and HAPLOSPORIDIA are best regarded as distinct subclasses, having no affinities with one another or with the TELOSPORIDIA.

The CNIDOSPORIDIA comprise the orders MYXOSPORIDIA, ACTINOMYXIDIA, and MICROSPORIDIA, to which a fourth order has been added by Kudo (1931), under the name of HELICOSPORIDIA, to include a single species described by Keilin. Dunkerly (1925), discussing the development and relationship of the MYXOSPORIDIA, pointed out that these, like the VOLVOCACEE among the Flagellates, represent an

unsuccessful line of advance from the typical Protozoan, not reaching, however, the Metazoan type of structure. He discussed the origin and relationship of the spore-forming nuclei and cells in the pansporoblast, and suggested that physiologically the spore of a Myxosporidian is a multicellular unit analogous to the Infusoriform embryo of the Mesozoan Dicyema, although the MYXOSPORIDIA exhibit Rhizopodan relationships, while the MESOZOA are probably derived from ciliated ancestors. Although the MYXOSPORIDIA do not represent a direct link between PROTOZOA and METAZOA, they seem to indicate a physiological reason for the origin of a soma, as a protective accessory to germ-cells. There is a well-marked alternation of generations among the CœLEN-TERATA, as there is among the MYXOSPORIDIA, and the occurrence of nematocysts in the former and the polar-capsules in the latter is not without significance.

The SPOROZOA are divided in this work into the following subclasses, orders, and suborders :---

- I. Subclass TELOSPORIDIA Schandinn.
  - I. Order GREGARINIDA A. Schneider em. Doflein.
    - 1. Suborder EUGREGARINARIA Doflein.
      - 1. Legion HAPLOCYTA Lankester.
      - 2. Legion SEPTATA Lankester.
    - 2. Suborder Schizogregarinaria Léger.
  - II. Order Coccidia Leuckart.
    - 1. Suborder ADELEIDEA Léger.
    - 2. Suborder EIMERIDEA Léger.
- III. Order HÆMOSPOBIDIA Danilewsky em. Doflein.
  - 1. Snborder HÆMOSPORIDIIDEA Wenyon,
  - 2. Snborder PIROPLASMIDEA Wenyon.
- II. Subclass CNIDOSPORIDIA Dofiein.
  - I. Order MYXOSPORIDIA Bütschli.
  - II. Order ACTINOMYXIDIA Stole.
  - III. Order MICBOSPORIDIA Balbiani.
  - IV. Order HELICOSPORIDIA Kudo.
- III. Subclass SARCOSPORIDIA Balbiani.
  - I. Order SARCOSPORIDIA Babudieri.
  - II. Order GLOBIDIA Babudieri.
- IV. Subclass HAPLOSPORIDIA Caullery & Mesnil.

The further classification into families and the genera and species dealt with will be seen from the Systematic Index.

#### Study of the Group in India.

Vandyke Carter (1888), Evans (1888), Hehir (1893), and Crombie (1894) were the earliest to observe the malarial parasites in India. It was, however, Ronald Ross (1895, 1897) who observed certain stages of development of the malarial parasites in the stomach of the mosquitoes fed on the blood of malarial patients, and later (1898) elucidated the life-cycle of the malarial parasites of birds and their transmission by *Culex* mosquitoes. In the course of his investigations between 1895 and 1899 he also noted a number of other parasites of mosquitoes. Ross had not received any special protozoological or entomological training, and had to coin a new terminology for the various phases of the parasites that he observed. Since that time India has been the field of the labours of many workers on different groups of SPOROZOA, and it will be convenient briefly to review their work by dealing with the various orders one by one.

GREGARINIDA.—Ross (1895) was the first to describe a Gregarine parasite from a mosquito, which he referred to as Gregarina culicis. Guenther (1914) described a parasite from another mosquito in Ceylon, and Mackie (1915) and Swaminath (1923) recorded certain organisms from sandflies in Assam and Bengal, which were later fully described by Short and Swaminath (1927). All these organisms are now known to belong to the genus Lankesteria. Cornwall (1915), while studying the anatomy and life-history of Lepisma saccharina (?), described some of its Gregarine parasites, which have been considered as new species of Gregarina in the present work. Ghosh (1923) described some monocystids from the earthworms of Calcutta.

About the same time I took up the study of the monocystids of earthworms in the Punjab, and Bhatia (1924), Bhatia and Chatteriee (1925), and Bhatia and Setna (1926) described quite a number of new species, belonging to the genera Monocystis, Apolocystis, Nematocystis, Stomatophora, Rhynchocystis, and Dirhynchocystis, from the earthworms from the Punjab and Bombay. Setna (1927) described a remarkable new organism under the name Gravallia quadrispina from Pheretima heterochæta from Bombay. Bhatia and Setna (1924) described several cephaline Gregarines from certain Insects, including Caulocephalus crenata from a beetle and Leidyana xylocopæ from the carpenter-bee, the latter being the first Gregarine to he recorded from a hymenopteran host. Later Setna (1931) described three new Gregarines, Bhatiella morphysz, Ferraria cornucephali, and Extremocystis dendrostomi from certain Polychætes, etc., taken at Port Blair in the Andaman Islands. Setna and Bhatia (1934) also described some new Gregarines from a prawn from Bombay.

Gates (1926, 1933) gave a description of two very curious parasites from the coolom of certain Burmese earthworms, *Aikinetocystis singularis* and *Nellocystis birmanica*, which have had to be placed in a family by themselves.

Working at Calcutta, Ray initiated a series of studies on

SPOROZOA from Indian Millipedes, and Ray (1933), Ray and Chakravarti (1933), and Chakravarti (1933, 1935, 1936) have described Stenophora khagendræ, Stenophora ellipsoidi, Monoductus lunatus, Hyalosporina cambolopsisæ, and Hyalosporina rayi. Three of these forms are placed in new families by their authors.

CocciditA.—Simond in 1901 was the first to publish a series of papers on Hæmogregarines from Indian tortoises and the gavial, and Laveran and Mesnil (1902) and Laveran and Nattan-Larrier (1912) observed Hæmogregarines in certain other tortoises. The pioneer work on the HæMATOZOA in Ceylon was carried out by Castellani and Willey (1904, 1905), Robertson (1908, 1910), and Dobell (1910). James (1905), Bentley (1905), Christophers (1905, 1906), and Patton (1908, 1909) described species of Hæmogregarina and Hepatozoon from various mammals, and the sexual cycle of Hepatozoon canis was fully worked out by Christophers (1907, 1912).

Froilano de Mello and his colleagues, working in Portuguese India, have published a number of papers (1915–1937) dealing with the Hæmogregarines of lizards and Toxoplasmids of various birds. De Mello also described (1921) Adelea pachelebræ in a mollusc.

In the family Eimeriidæ many species are now known from hosts belonging to all classes of Vertebrates. Cooper and Gulati (1926), Cooper (1926, 1927), Sen (1932), and Ware (1936) recorded cases of bovine Coccidiosis. Knowles and Das-Gupta (1931, 1934) studied the CocciDIA of the mongoose, cat, and lizard, and Das-Gupta (1934) recorded a case of coccidial infection in man. Ray (1935 *a*, *b*) described *Wenyonella hoarei* front the squirrel and *Eimeria laminata* from the common toad; and Ray and M. Das-Gupta (1935) described *Isospora wenyoni* from the toad, and also (1937 *a*, *b*) described CocciDIA from a lizard and the cobra. Setna and Bana (1935 *a*, *b*), working in Bombay, described *Eimeria* harpodoni from a fish and *Eimeria flaviviridis* from a lizard. They have also recorded the occurrence of CocciDIA in a number of species of marine fish.

*H.ÆMOSPORIDIA.*—As mentioned already, Vandyke Carter (1888), Evans (1888), Hehir (1893), and Crombie (1894) were the earliest to observe the malarial parasites in India. But it was the memorable work of Ronald Ross (1895, 1897, 1898) that laid the foundation of the correct understanding of the life-cycle of the malarial parasites in man and birds. He succeeded in establishing the fact that further development of the human malarial parasites takes place in the body of the dapple-winged mosquitoes that have fed on the blood of malarial patients; and he elucidated the complete life-cycle of the malarial
parasites of birds and the part played in their transmission by Culex mosquitoes. His discoveries were confirmed by Daniells (1899, 1900), Stephens and Christophers (1903), and Christophers (1904). Cornwall (1901), Donovan (1909), and Cragg and Naidu (1918) recorded observations on the morphology of the malarial parasites. In more recent years important contributions have been made to our knowledge of the morphology of these parasites by Acton, Curjel, and Dewey (1921), Knowles (1923), Lal (1925), Knowles and Senior White (1927), Clark (1927), Hehir (1927), Row (1928, 1930), Knowles, Acton, and Das-Gupta (1929), and Knowles (1931). Sinton (1929) has published a complete bibliography of the literature dealing with malaria in India. Papers dealing with transmission by various species of Anophelines and the seasonal incidence of malaria include (among others) those of Stephens and Christophers (1902), James (1903), Bentley (1911), James and Liston (1912), J. R. & H. Adie (1913), Gill (1925), James, Nicol, and Shute (1927), Carter and Jacocks (1929), Carter (1930), Knowles, Senior White, and Das-Gupta (1930). King (1931), and Ivengar (1931-4). The distribution of the various species of malarial parasites has been dealt with by Christophers and Sinton (1926), and the distribution of different species of Anopheline carriers in important monographs by Covell (1927, 1931).

The malarial parasites of monkeys have been studied by Knowles (1919), Donovan (1920), Knowles and Das-Gupta (1932, 1934), Sinton and Mulligan (1933), Sinton (1934), and Mulligan (1935). Malarial parasites have also been described in various other mammals, viz., by Mackie (1914) in a bat, Sheather (1919) in the buffalo, de Mello and Paes (1923) in the horse, de Mello and colleagues (1928) in the ant-eater, and de Mello (1936) in the otter, and in various birds by de Mello (1935).

Parasites belonging to the family Hæmoproteidæ have been described by Castellani and Willey (1904, 1905), Acton and Knowles (1914), Alcock (1914), Adie (1915), de Mello and colleagues (1917), de Mello and Raimundo (1934), de Mello (1935), and de Mello and Afonso (1935).

The Piroplasmids have also been studied by a number of workers. Lingard and Jennings (1904) were the earliest to observe these parasites in various animals. Webb (1906) observed them in fox-hounds and Axe (1906) in horses. Christophers (1907) described the developmental stages of *Piroplasma canis* in the tick, and Patton (1910) described *P. gibsoni* from the dog and the jackal. Baldrey (1911), Gaiger (1911), and Symons and Patton (1912) also studied the Piroplasmids. Sinton (1921) described *Nuttallia ninense* from the hedgehog, Rau (1926) and Symons (1926) studied the Piroplasmids from hounds, Cooper (1926) from cattle, Krishna Iyer (1933) from goats, and Achar and Shrikantiah from sheep. Sarwar (1935) described *Piroplasma taylori* from a goat, but I (1936) have shown that this form appears to be identical with *Theileria hirci*. Shortt (1936) has re-studied the life-history of *Babesia canis* in the dogtick.

CNIDOSPORIDIA.—Very little work has been done on the CNIDOSPORIDIA. The MYXOSPORIDIA have been studied by Bosanquet (1910), Southwell (1915), Southwell and Prashad (1918), and Ray (1933). As regards the MICROSPORIDIA, Korke (1916) described a species of Nosema parasitic in the dog-flea. Mrs. H. A. Adie (1922) observed certain bodies in Cimex rotundatus in a Kala-azar infected area in Assam, and believed them to be Leishman-Donovan bodies, but Christophers (1922) and Short and Swaminath (1922) regarded the organism as a species of Nosema, and described it as N. adiei. Iyengar (1929) and Kudo (1929) have studied the inicrosporidian parasites of Anopheline larvæ.

SARCOSPORIDIA.—Shipley (1904) and Willey, Chalmers, and Phillip (1904) described sarcosporidian infection in buffaloes in Ceylon; Chatterjee (1907) described what was probably the same species from the heart-muscle of a cow in Calcutta. Vasudevan (1927) described a case of sarcosporidian infection of man in Madras. Hassan (1935) has described a new species of *Globidium* from a cow.

 $H_{APLOSPORIDIA}$ .—Minchin and Fantham (1905) described an organism, which they named *Rhinosporidium kinealyi*, from a nasal polyp, and regarded it as belonging to the HAPLO-SPORIDIA, but Ashworth (1923) has shown that the organism is a fungus. Vasudevan (1932) described a case of rhinosporial infection of the fore-arm of a man in Ceylon; but no true HAPLOSPORIDIA have so far been described from India.

#### DISTRIBUTION.

The geographical distribution of parasites usually follows that of their hosts. The names of the hosts, and the localities in which any parasite has been found, are noted after the description of the species, and, generally speaking, the same parasite may be expected to occur in the same species of hosts in other localities also. The following lists of (i) parasites and their hosts, and (ii) the hosts and their parasites, will, it is hoped, be found useful, and indicate at a glance which of our commoner animals still remain to be examined for their parasites.

SPOR.

### (i) List of Parasites and their Hosts \*.

Parasito.	Host.	Seat.
GREGARINIDA.		
Fam, MONOCYSTIDÆ.		
Monocystis beddardi	Eutyphœus nicholsoni . Eutyphœus sp	Seminal vesicles.
Monocustis bengalensis	Pheretima posthuma	Seminal vesicles.
Monocustis Uoidi	Pheretima posthuma .	Seminal vesicles.
Monocystis pheretimi	Pheretima posthuma .	Seminal vesicles.
Apolocystis matthaii	Megascolex trilobatus .	Seminal vesicles.
Nematocystis hessei	Pheretima heterochæta .	Seminal vesicles.
Nematocystis lumbricoides	Pheretima heterochæta.	Seminal vesicles.
Nematocystis plurikaryo- somata.	Allolobophora (Eisenia) fætida.	Seminal vesicles.
Nematocystis stephensoni . Nematocystis vermicularis .	Eutyphœus incommodus Pheretima barbadensis .	Seminal vesicles. Seminal vesicles.
Fam. RHYNCHOCYSTIDÆ.		
Rhynchocystis awatii	Pheretima elongata	Seminal vesicles.
Rhynchocystis cognetti	Allolobophora caliginosa	Seminal vesicles.
Rhynchocystis mamillata .	Pheretima elongata	Seminal vesicles.
Fam. STOMATOPHORIDÆ.		
Stomatophora bulbifera	Pheretima elongata	Seminal vesicles.
Stomatophora coronata	Pheretima barbadensis.	Seminal vesicles.
Stomatophora diadema	Pheretima barbadensis.	Seminal vosicles.
	Pheretima posthuma .	Seminal vesicles.
Fam. Zygocystidæ.		
Extremocystis dendrostomi.	$Dendrostoma\ signifier$ .	Cœlomic cavity.
Fam. AIKINETOCYSTIDÆ.		
Aikinetocystis singularis .	Eutyphœus foveatus	Cœlomic cavity.
• •	Eutyphœus spinulosus .	Cœlomic cavity.
	Eutyphœus rarus	Cœlomic cavity.
	Eutyphœus peguanus	Cœlomic cavity.
Nellocystis birmanica Fam. DIPLOCYSTIDÆ.	Pheretima compta	Cœlomic cavity.
Lankesteria culicis	Aëdes (Stegomyia)	Stomach and Mal-
	ægypti.	pighian tubes.
	Aëdes (Stegomyia) albo-	Stomach and Mal-
Lankesteria mackiei	pictus. Phlebotomus argentipes.	pighian tubes. Alimentary canal
	Phlebotomus papatasi .	and hæmocœle. Alimentary canal
		and hæmooæle.
Lankesteria tripteroidesi	Tripteroides doffeini	Hæmocœle and respiratory sys- tem.
Fam. LECUDINIDÆ.		
Lecudina brasili	Lumbriconereis sp	Intestine.
Bhatiella morphyse	Morphysa sanguinea .	Mid-gut.
Ferraria cornucephali	Morphysa sanguinea .	Mid-gut.
INCERT & SEDIS.		
Dirhynchocystis globosa	Eutyphœus sp Pheretima nosthuma	Seminal vesicles.
Grayallia quadrispina	Pheretima heterochæta.	Cœlomic cavity.

\* For Supplementary List of Parasites and their Hosts recorded since the above list was in type, see p. 374.

Parasite.	Host.	Seat.
Fam. STENOPHORIDÆ.		
Stenonhora ellipsoidi	Diplopola sp	Mid-out
Stenophora khagendræ	Zikadesmus (?) sp.	Intestine.
		1100000000
Fam. Hyalosporinidæ.		
Hyalosporina cambolop- sisæ.	Cambolopsis sp	Alimentary canal.
Hyalosporina rayi	Polydesmus sp Strongylosoma contor- tipes.	Alimentary canal. Alimentary canal.
Fam. GREGARINIDÆ.	1	
Leidyana gryllorum	Gryllus sp	Gizzard and mid-
Leiduana xulocopæ	Xulocopa æstuans	Alimentary canal.
Gregarina aciculata	Lepisma saccharina	Mid-gut.
Gregarina cornwalli	Lepisma saccharina	Mid-gut.
Gregarina oviceps	Gryllus sp	Gizzard and mid- gut.
Caulocephalus crenata	Aulacophora foveicollis.	Alimentary canal.
Protomagalhænsia (?)	Parapeneopsis sculptilis	Intestine.
attenuata. Hirmocystis (?) parapene- opsisi.	Parapeneopsis sculptilis	Intestine.
Fam ACTINGTERNATID		
Staining materiani	Matanlan dontines	Intesting
Steinina metapiaxi	Metaplax aentipes	intestine.
Fam. DACTYLOPHORIDÆ.		
Grebneckiella navillæ	Scolopendra sp	Intestine.
Fam. MONODUCTIDÆ.		
Monoductus lunatus	Strongylosoma contor-	Alimentary canal.
COCCIDIA.	upco.	
Fam. ADELEIDA		
Adelea pachelebræ	Pachelabra mæstra	Intestine and di-
Adelina schellacki	Cormocephalus dentipes	Intestine.
Fam HEMOGREGARINDE		
Lamonanying bound	Pana homodostulo	Blood
namoyregarina veresinejji.	Rana limnocharis	Blood
	Rana tiarina.	Blood.
Hæmogregaring cantliei	Eryx conicus	Blood.
Hæmogregarina hankini .	Crocodilus porosus	Blood.
	Gravialis gangeticus	Blood.
Hæmogregarina laverani .	Lissemys punctata granosa.	Blood.
Hæmogregarina leschen- aultii.	Hemidactylus leschen- aulti.	Blood.
Hæmogregarina magna	Rana limnocharis	Blood, Blood
Hæmooreoarina mala-	Lissemus punctata	Blood, liver, bone-
barica.	granosa.	marrow, etc.
Hæmogregarina mcsnili	Kachuga teetum	Blood.
Hæmogregarina mirabilis .	Tropidonotus asperri-	Blood.
	mus. Tropidonetus niscator	Blood
	1 TOWNSONOLUS PROCEEDT .	C 2
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Host.

Parasite.	Host.	Seat.
Fam. HEMOGREGARINIDE		
(cont.)		
Ham a maganing main	Naia naia	Blood
Hamogregaring micorig	Generanda triinga	Blood
Lizemogregarina moorize	Liesenus nunctata	Blood
	aranora	Dioou.
	Orchranchus shinleyi	Body
Homogragaring nucleo.	Bufo melanostictus	Blood
hisecons	Dayo mounconcoras	Dioou.
Hæmogregaring nuthonis.	Puthon molurus	Blood.
Hæmooregaring rara	Chinemus reevesii	Blood.
Hæmogregaring rodriguesi.	Hemidactulus brookei.	Blood.
Hæmogregaring stepanowi.	Chinemus reevesii	Blood.
ana.	•	
Hæmogregarina testudinis.	Testudo emys	Blood.
Hæmogregarina thomsoni.	Agama tuberculata	Blood.
Hæmogregarina thyrsoidea.	Thyrsoidea macrurus .	Blood.
Hæmogregarina tiedri	Hemidactylus triedrus.	Blood.
Hæmogregarina vittatæ	Lissemys punctata	Blood.
	granosa.	
Hæmogergarina xavieri	Lissemys punctata	Blood, lungs,
	granosa.	spleen, liver.
Hæmogregarina sp	Trichogaster fasciatus.	Blood.
Hæmogregarina sp	Varanus monitor	Blood.
Hæmogregarina sp	Bungarus cæruleus	Blood.
Hæmogregarina sp	Chrysopelea ornata	Blood.
Hæmogregarina sp	Zaocys mucosus	Blood.
Hæmogregarina sp	Porocephalus pationi	Body.
Hæmogregarina sp	Coluber nelena	B1000.
Hæmogregarina sp	Dimensional Services	Dlood.
Hæmogregarina sp	stenii.	blood.
Hæmogregorina sp	Dipsadomorphus ceylon- ensis.	Blood.
Hæmogregarina sp	Dendrophys pictus	Blood.
Hæmogregarina sp	Dryophis mycterizans .	Blood.
Hæmogregarina sp	Eryx johnii	Blood.
Hæmogregarina sp	Naja bungarus	Blood.
Hæmogregarina sp	Naja naja	Blood.
Hæmogregarina sp	Naja naja var. atra	Blood.
Hæmogregarina sp	Naja sp	Blood.
Hæmogregarina sp	Python sp	Blood.
Hæmogregarina sp	Tropidonotus stolatus.	Blood.
Hæmogregarina sp	Vipera russellii	Blood.
Hepatozoon adiei	Eagle (not identified) .	cytes.
Hepatozoon canis	Canis aureus	Internal organs and leucocytes.
	Canis familiaris	Internal organs
	Cyon dukhunensis	Internal organs and leucocvtes.
	Vulpes bengalensis	Internal organs and leucocytes
	Rhipicephalus san-	Body.
	guineus.	v
Hepatozoon felis domestici.	Felis sp	Leucocytes.
Hepatozoon funambuli	Funambulus pennantii.	Leucocytes.

# Funambulus pennantii. Leucocytes. Hæmatopinus sp. .... Body.

Parasite.	Host.	Seat.
Hepatozoon gerbilli	Tatera indica Hæmatopinus stephensi	Erythrocytes. Body.
Hepatozoon leporis Hepatozoon muris	Lepus nigricollis Rattus norvegicus	Blood. Liver-cells and
	Rattus rufescens	Liver-cells and leucocytes.
<b>TT</b>	Lælaps echidninus	Body.
Karyolysus jorgei	Calotes versicolor major	Blood, and endo- thelial cells of
Fam. EIMERIIDÆ.		nvoi and iungs.
Lankesterella minima	Rana tigrina	Blood.
Lankesterella monilis	Rana limnocharis	Blood.
	Rana tigrina	Blood.
Isospora belli Isospora bigemina	Homo sapiens Canis sp	Alimentary canal. Sub-epithelial tissues of the
Isospora caloti	Calotes versicolor	intestine. Intestinal epithe- lium.
Isospora felis	Felis domesticus	Intestinal epithe- lium.
Isospora knowlesi	Hemidactylus flaviviri- dis.	Alimentary canal.
Isospora minuta	Naja naja	Alimentary canal.
Isospora rivolta	Felis domesticus	Intestinal epithe- lium.
T	Herpestes auropunctatus	Cæcum.
Isospora wenyoni	Bay en	Alimentary canal
Eimeria clupearum	Homo sapiens	Intestine.
Eimeria columbæ	Columba livia inter- media.	Intestine.
Eimeria cylindrica	Natrix piscator	Roctum.
Eimeria faurei	Uvis sp	Intestine.
Eimeria jusoioiriuis	dis.	intestine.
Eimeria hemidactyli	Hemidactylus flaviviri- dis.	Intestine.
Eimeria kermorganti	Gavialis gangeticus	Spleen.
Limeria knowlesi	Hemidaciyius flaviviri- dis.	Intestine.
Eimeria laminala	Lissemus nunctata	Gall-bladder and
Lonier as regers	granosa.	bile-ducts.
Eimeria mitraria	Chinemys reevesii	Intestine.
Limeria najee	Naja naja	Rectum.
Eimeria precatori	Homo services	Alimentary canal
Eimeria southwelli	Atobatis narinari	Intestine.
Eimeria yakimovi	Boselaphus tragoca- melus.	Alimentary canal.
Eimeria sp	Bos sp	Alimentary canal.
Eimeria sp.	Trichiurus savala	Intestine.
L'imeria sp.	Batrachus grunniens	Intestine.
wimeria sp	pinepneius tauvina	LIIVOSCIIIO,

Parasite.	Host.	Seat.
Fam. EIMERIIDÆ (cont.).		
Fam. EIMERIDÆ (cont.). Eimeria sp Eimeria sp Eimeria sp Eimeria sp Eimeria sp Eimeria sp Eimeria (1) sp Wenyonella hoarei Pythonella bengalensis Aggregata sp INCERTÆ SEDIS.	Engraulis mystax Otolithus ruber Sillago sihama Coilia dussumieri Plotossus canius Epinephelus diacanthus Culex sp Sciurus sp Parapeneopsis sculp- tilis.	Intestine. Intestine. Intestine. Intestine. Intestine. Stomach cells. Intestine. Intestine. Intestine.
Toxoplasma columbæ Toxoplasma columbæ Toxoplasma cuniculi	Columba livia Lepus sp	Liver and spieen. Leucocytes. Liver, spieen, bone- marrow, and heart-blood.
Toxoplasma fulicæ	Fulica atra	Lung.
Toxoplasma sp	Passer sp	Blood.
Toxoplasma sp Toxoplasma (?) merogenes	Saxicola caprala Homo saniens	Blood and anleen
I coopidania (1) pgrogenes 1		Dioou una spicem
HAMOSPORIDIA.		
Hamomatau auticau	Authoridan wings	Pland
Hæmoproteus asturis	Anthropotaes virgo Astur badius dussu- mieri.	Blood.
Hæmoproteus cerchneisi	Cerchneis tinnunculo- sus objurgatus.	Blood.
Hæmoproteus columbæ	Columba livia	Blood.
Hæmoproteus coraciæ	Lynchia maura Coracias benghalensis benghalensis	Stomach. Blood.
Hæmoproteus corvusi	Corvus levaillanti macrorhynchus.	Blood.
77	Corvus splendens	Blood.
Hæmoproteus danilewskyt	Anas (Fuligula) Daeri . Consuchus saularis	Blood.
	Coracias benghalensis indica.	Blood.
	Garrulax albigularis	Blood.
	Garrulus lanceolatus	Blood.
	Kittagingla magnoung	Blood.
	Melonhus melanicterus	Blood.
	Mesia argentauris	Blood.
	Nettapus coromandeli- anus.	Blood.
	Otus barkamæna var. malabarica.	Blood.
	Propasser rhodochrous.	Blood.
Homometry dayoidi	doni.	Blood
Hæmoptoteus gumorhidis.	Gumnoris xonthocollis	Blood.
Hæmoproteus herodiadis .	Egretta intermedia intermedia.	Blood.
Hæmoproteus kopki	Hemidactylus brooki	Blood.

Parasite.	Host.	Seat.
Hæmoproteus machlolophi.	Machlolophus xantho- genus.	Blood.
Hæmoproteus metchnikovi.	Chitra indica	Blood.
Hæmoproteus orioli	Oriolus oriolus kundoo.	Blood.
Hæmoproteus raymundi .	Leptocoma zeylonica	Blood, lungs, spleen.
Hæmoproteus, rileyi	Pavo cristatus	Blood.
Hæmoproteus simondi	Hemidactylus leschen- aulti.	Blood.
Hæmoproteus_sturni	Sturnia malabarica	Blood.
Hæmoprcteus upupæ	Upupa epops orientalis.	Blood, lungs, liver.
Hæmoproteus wenyoni	Orthotomus sutorius	Blood.
Hæmoproteus sp.	Antigone antigone	Blood.
Hæmoproteus sp.	Calænas nicobarica	Blood.
Hæmoproteus sp	Centropus sinensis	Blood.
Hæmoproteus sp	Chloropsis aurijrons	Blood.
<i>Hæmoproleus</i> sp	striatus.	Blood.
Hæmoproteus sp	Uyanops flavifrons	Blood.
Hæmoproteus sp	Denarocitta ruja vaga- bunda.	Blood.
Hæmoproteus sp	Haliaeëtus leucoryphus.	Blood.
Hæmoproteus sp	Hemidactylus sp	Blood.
Hæmoproteus sp	Coryllis beryllinus	Blood.
Hæmoproteus sp	fasciala.	Blood.
Hæmoproteus sp	Otus bakkamæna	Blood.
Hæmoproteus sp.	Strix ocellata	Blood.
Hæmoproteus (?) ægithinæ.	Ægithina liphia	Blood.
El æmoproteus (?) anthi	Anthus richardi rufulus	Blood.
Hæmoproteus (?) branæ .	Athena orama	Blood. Blood
Hæmoproteus (1) centropi	parroti.	Blood.
Hæmoproteus (!) dicrurs	macrocercus.	Blood.
Hæmoproteus (?) elani	Elanus cæruleus voci- ferus.	Blood.
Hæmoproteus (?) gallinulæ	Gallinula chloropus	Blood.
Hæmoproteus (?) halcyonis	Halcyon smyrnensis	Blood.
Hæmoproteus (?) otocompsæ	Elathea jocosa	Blood.
Hæmoproteus (?) pastoris.	Pastor roseus	Blood.
Hæmoproteus (?) plataleæ.	Platalea leucorodia major.	Blood.
Hæmoproteus (?) tephro- dornis.	Tephrodornis pondiceri- anus pondicerianus.	Blood.
Hæmoproteus (?) therei- cerycis.	Thereiceryx zeylanicus inornata.	Blood.
Hæmoproteus (?) therei-	Thereiceryx zeylanicus	Blood.
cerycis var. zeylonica.	zeylanjcus.	~
Leucocytozoon chloropsidis.	Chloropsis aurifrons davidsoni.	Blood.
Leucocytozoon coracite	Coracias benghalensis benghalensis.	Blood and smears from lungs.
Leucocytozoon melloi	Coracias benghalensis benghalensis.	Blood and smears from lungs.
Leucocytozoon sp	Athene brama	Blood.
Leucocytozoon sp	Falco sp	Blood.
Leucocytozoon sp	Ianthocincla rufogularis	Blood.
Leucocytozoon sp.	Leiothrix lutea	Blood.

Parasite.	Host.	Seat.
Fam. Hæmophoteidæ (cor	ut.).	
Leucocytozoon sp	Oreicola ferrea	Blood.
Leucocytozoon sp	Propasser rhodochrous.	Blood.
Leucocytozoon sp	Peking robins	Blood.
Fam. PLASMODIDÆ.		
Proteosoma chloropsidis	Chloropsis aurifrons davidsoni.	Blood.
Proteosoma columbæ	Columba sp	Blood.
Proteosoma gallinulæ	Gallinula chloropus	Blood.
Proteosoma herodiadis	Egretta intermedia in- termedia.	Blood.
Proteosoma moruony	Copsychus saularis	Blood.
Proteosoma passeris	Passer domesticus	Blood.
Proteosoma præcox	Aidemosyne malabarica	Blood.
	Sturnus vulgarus pol- taratskyi	Blood.
	Tragopan satyra	Blood.
	Anopheles subpictus	Body.
Bustassen a sa	Culex jangans	Blood
Proteosoma sp.	Antigone antigone	Blood
Proteosoma ap	Corollin borullinus	Blood
Proteosoma sp	Cuanons flavifrons	Blood.
Proteosoma sp	Elathea jocosa	Blood.
Proteosoma sp.	Emberiza fucata	Blood.
Proteosoma sp.	Gracula indica	Blood.
Proteosoma sp.	Garrulax leucolophus .	Blood.
Proteosoma sp	Leiothrix lutea	Blood.
Proteosoma sp	Melophus melanicterus	Blood.
Proteosoma sp	Saxicola caprata	Blood.
Proteosoma sp	Turdus boulboul	Blood.
Laverania malariæ	Homo sapiens	Blood.
	Anopheles annularis	Body.
	Anopheles culterfactes.	Body.
	Anopheles fluvratilis .	Body.
	Anopheles junestus	Body.
	Anophetes jegportensis.	Body.
	Anopheles miscululus	Body.
	Anonheles nhilinninen.	Body.
	sis.	Doug.
	Anopheles stephensi	Body.
Diana di una minara	Anopheies sundaicus	Body.
Plasmoarum vivax	Amonheles annularis	Blood. Body
	Anopheles culicitacies	Body.
	Anopheles fluviatilis	Body.
	Anopheles funestus	Body.
	Anopheles ieuporiensis.	Body.
	Anopheles maculatus .	Body.
	Anopheles minimus	Body.
	Anopheles philippinen- sis.	Body.
	Anopheles stephensi	Body.
	Anopheles sundaicus	Body.
Plasmodium molarise	Homo sapiens	Blood.
	Anopheles annularis	Body.
	Anopheles culicifacies.	Body.

Parasite.	Host.	Seat.
Plasmodium malariæ	Anopheles fluviatilis	Body.
	Anopheles funestus	Body.
	Anopheles jeyporiensis.	Body.
	Anopheles maculatus .	Body.
	Anopheles minimus	Body.
	Anopheles philippinen-	Body.
	sis.	
	Anopheles stephensi	Body.
	Anopheles sundaicus	Body.
Plasmodium cynomolgi	Silenus irus	Blood.
	Silenus rhesus	Blood.
	Silenus sinicus	Blood.
	Anopheles annuaris	Salivary glands.
	Anopheles curicijacies .	Salivary glanda.
	Anomholes enlendidus	Salivary glanda
Plasmodium inui	Silenus irus	Blood.
	Silenus rhesus	Blood.
Plasmodium knowlesi	Silenus irus	Blood.
	Silenus rhesus	Blood.
Plasmodium kochi	Cercopithecus spp	Blood.
Plasmodium pitheci	Simia satyrus	Blood.
Plasmodium semnopithecus	Presbytes pileatus	Blood.
	Pygathrix entellus	Blood.
Diama d'un hatali	Silenus rhesus	Blood.
rasmoarum ouoans	Dos maicus	Blood
Plasmodium canis	Canis familiaris	Blood
Plasmodium equi	Equus caballus	Blood.
Plasmodium mackiei	Myotis muricola	Blood.
Plasmodium narayani	Lutra lutra	Blood.
Plasmodium pteropi	Pteropus medius	Blood.
Plasmodium ratufæ	Ratufa indica	Blood.
Plasmodium tyrio	Manes pentadactyla	Blood.
Fam. THEILERIIDÆ.		
Theileria celli	Macacus pileatus	Blood.
Theileria hirci	Capra hircus	Blood.
Theileria mutans	Bos indicus	Blood.
Theileria parva	Bos indicus	Blood.
Fam. BABESHDÆ.		
Babesia bigemina	Bos indicus	Blood.
Babesia caballi	Equus caballus	Blood.
Babesia canis	Canis aureus	Blood.
	Canis familiaris	Blood.
	Rhipicephalus sangui-	Blood.
Raberia emi	neus. Eaune caballue	Blood
Bahesia felis	Felis en	Blood.
Babesia gibsoni	Canis aureus	Blood.
	Canis familiaris	Blood.
Babesia motasi	Ovis sp	Blood.
Babesia ninense	Erinaceus sp	Blood.
Babesia sergenti	Capra hircus	Blood.
Babesia soricis	Crocidura cærulea	Blood.
Babesia tropicus	Elephant, camel, gui-	B1000
	nea-pigs, iowis, and lizards.	

#### Sect

Parasite.	Host.	Seat.
Fam. BABESHDÆ (cont.).		
Babesia sp Babesia sp Babesia sp	Axis axis Cyon dukhunensis Herpestes edwardii	Blood. Blood. Blood.
INCERTÆ SEDIS.		
Grahamella muris Grahamella sp Paraplasma sp Anaplasma sp	Rat	Blood. Blood. Blood. Blood. Blood. Blood. Blood. Blood. Blood. Blood. Blood. Blood. Blood. Blood.
CNIDOSPORIDIA.		
MYXOSPORIDIA.		
Ceratomyxa gobioidesi	Gobioides rubicundus .	Liver, gall-bladder kidney, ovary.
	Macrones gulio	Liver, gall-bladder, kidney, ovary.
	Trichogaster fasciatus .	Liver,gall-bladder, kidney, ovary
Chloromyxum amphinovi .	Amphipnous kuchia	Gall-bladder.
Fam. SPHÆROSPORIDÆ.		** 3 /1 1
Sphærospora sp	Barilius barna	Under the scales.
Fam. Myx1D11DÆ. Myxidium danilewskyi Myxidium mackiei Myxidium sp	Chinemys reevesii Trionyx gangeticus Clarias batrachus Geoemyda trijuga Kachuga smithi Lissemys punctata granosa.	Kidney. Kidney. Gall-bladder. Gall-bladder. Gall-bladder. Gall-bladder.
Zechokkella prashadi	Ophiocephalus punctatus Saccobranchus fossilis. Bufo melanostictus Lissemys punctata granosa. Rana tiorina	Gall-bladder. Gall-bladder. Gall-bladder. Gall-bladder. Gall-bladder.
Fam. MYXOBOLIDÆ.		
Myxobolus calbasui	Cirrhina mrigala	Liver and gall- bladder.
Myzobolus nodularis Myzobolus sp	Clarias batrachus Katla katla Rasbora daniconius Rasbora daniconius	Ovary and liver. Gills. Muscles. Subcutaneous tis- sue.
Thelohanellus rohitse Thelohanellus seni	Labeo rohita Labeo rohita	Gills. Median and caudal
Henneguya ophiocephali	Ophiocephalus puncta- tus.	Gills and muscles.

Parasite.	Host.	Seat
Henneguya otolithus	Otolithus maculatus Otolithus ruber	Bulbus arteriosus. Bulbus arteriosus.
MICROSPORIDIA.		
Fam. Nosematidæ.		
Nosema adiei	Cimex rotundatus	Gut, salivary glands, and ovaries.
Nosema bombycis	Bombyx mori	All tissues.
Nosema ctenocephali	Ctenocephalus canis	Digestive tract of larvæ.
Thelohania indica	Anopheles hyracanus .	Adipose tissue of larvæ.
Thelohania legeri	Anopheles barbirostris .	Adipose tissue of larvæ.
	Anopheles fuliginosus .	Adipose tissue of larvæ.
	Anopheles funestus	Adipose tissue of larvæ.
	Anopheles hyracanus .	Adipose tissue of larvæ.
	$An opheles\ pseudojamesi$	Adipose tissue of larvæ.
	Anopheles ramsayi	Adipose tissue of larvæ.
	Anopheles subpictus	Adipose tissue of larvæ.
Thelohania obscura	Anopheles funestus	Adipose tissue of
Microsporidian (indet.)	Stegomyia sp	Nerve-cord of
Microsporidian (indet.)	Culex fatigans	Nerve-cord of imago.
SARCOSPORIDIA.		
Fam. SARCOCYSTIDÆ.		
Sarcocystis blanchardi	Bos bubalus	Muscles.
~	Bos indicus	Heart-muscle.
Sarcocystis lindemanni	Homo sapiens	Muscles.
Fam. GLOBIDIDÆ.		•
Globidium fusiformis Globidium sp	Bos indicus Wallaby	Alimentary canal. Alimentary canal.
HAPLOSPORIDIA		
INCEPT & SEDIS		
Dhimooporidium eachani	Home equipme	Need polymi
Rhinosporidium sp	Homo sapiens	Abscess cavities.
(ii) List of	Hosts and their Parasit	es *.
Host.	Parasite.	Seat.
MAMMALIA.		
Axis axis	Babesia sp.	Blood.
Bet	Anaplasma sp.	Blood.
Bos bubalus	Plasmodium bubalis Sarcocystis blanchardi.	Blood. Muscles.

\* For Supplementary List of Hosts and their Parasites recorded since the above list was in type, see p. 375.

Host.	Parasite.	Seat.
MAMMALIA (cont.).		
Bos indicus	Plasmodium bubalis	Blood.
	Theileria mutans	Blood.
	Theileria parva	Blood.
	Babesia bigemina	Blood.
	Sarcocystis blanchardi .	Heart-muscle.
	Globidium fusiformis .	Alimentary canal.
Bos sp	Isospora sp.	Alimentary canal.
•	Eimeria sp	Alimentary canal.
Camelus sp	Babesia tropicus	Blood.
Canis aureus	Hepatozoon canis	Internal organs
		and leucocytes.
	Babesia canis	Blood.
~	Babesia gibsoni	Blood
Canis familiaris	Hepatozoon canis	Internal organs
	<i>m</i> , ;	and leucocytes.
	Toxoplasma canis	Blood.
	Plasmodium canis	Blood.
	Babesia canis	Diood.
Constante en	Baoesia giosoni	Subouitbolial tic.
Canis sp	1808pora begemina	anes of the in-
		testine.
Carra birrous	Theileria hirci	Blood.
	Babesia sergenti	Blood
Cavia porcellus (?)	Babesia tropicus	Blood.
Cerconithecus sp.	Plasmodium kochi	Blood.
Crocidura cærulea	Babesia soricis	Blood.
Cyon dukhensis	Hepatozoon canis	Internal organs
- <b>9</b>	-	and leucocytes.
	Babesia sp	Blood.
Elephas sp	Babesia tropicus	Blood.
Equus caballus	Plasmodium equi	Blood.
-	Babesia caballi	Blood.
	Babesia equi	Blood.
Erinaceus sp.	Babesia ninense	Blood.
Felis domesticus	Isospora felis	Intestinal epithe-
	<b>T</b> • <b>Y</b>	hum.
	Isospora rivolta	Intestinal epitne-
Walie an	Hanataraan falia	lium.
<i>r</i> ens sp	domentici	Leucocytes.
	Bahasia felio	Blood
Funambular non nantis	Henatoroon funambuli	Lencoevtee.
Herpestes auronunctatus.	Isospora rivolta	Cæcum.
Hernestes edwardii	Babesia sp.	Blood.
Homo sapiens	Isospora belli	Alimentary canal.
	Eimeria clupearum	Intestine.
	Eimeria sardinæ	Alimentary canal.
	Toxoplasma (?) pyro- genes.	Blood and spleen.
	Laverania malariæ	Blood.
	Plasmodium malariæ	Blood.
	Plasmodium vivax	Blood.
	Paraplasma sp	Blood.
	Anaplasma sp	Blood.
	Sarcocystis lindemanni.	Blood.
	Rhinosporidium seeberi	Nasal polypi.
	Kninosporidium sp	ADCOSS CAVITIOS.

Host.	Parasite.	Seat.
<i>Lepus</i> sp	Toxoplasma cuniculi	Liver, spleen, bone-marrow,
T	77 days 1	and neart-blood.
Lepus nigricollis	Hepatozoon leporis	Blood.
Lutra lutra	Plasmodium narayani	Blood.
Macacus pileatus	Ineuteria cetti	Blood.
Manes pentadactyla	Plasmodium tyrio	Blood.
	Grahamella sp.	Blood.
Myotis muricola	Plasmodium mackiei	Blood.
$Ovis sp. \ldots$	Eimeria faurei	Intestine.
	Babesia motasi	Blood.
Presbytes pileatus	Plasmodium semnopi- thecus.	Blood.
Pteromys petaurista	Hepatozoon sp	Blood.
Pteropus medius	Plasmodium pteropi	Blood.
Pygathrix entellus	Plasmodium semnopi- thecus.	Blood.
Rattus norvegicus	Hepatozoon muris	Liver-cells and erythrocytes.
Rattus rufescens	Hepatozoon muris	Liver-cells and erythrocytes.
Rattus sp	Grahamella muris	Blood.
Ratufa indica	Plasmodium ratufæ	Blood.
Sciurus sp	Wenyonella hoarei	Intestine.
Silenus irus	Plasmodium cunomoloi	Blood.
	Plasmodium inui	Blood.
	Plasmodium knowlesi .	Blood.
Silenus rhesus	Plasmodium cunomolai.	Blood.
	Plasmodium inui	Blood.
	Plasmodium knowlesi .	Blood.
	Plasmodium semnopi- thecus.	Blood.
Silenus sinicus	Plasmodium cynomolgi.	Blood.
Simia saturus	Plasmodium pitheci	Blood.
Tateria indica	Hepatozoon gerbilli	Ervthrocytes.
Vulpes bengalensis	Hepatozoon canis	Internal organs and leucocytes.
Wallaby	<i>Globidium</i> sp	Alimentary canal.
Aves.		
Ægithina tiphia	Hæmoproteus (?) ægi- thinæ.	Blood.
Aidemosyne malabarica	Proteosoma præcox	Blood.
Anas (Fuligula) baeri	Hæmoproteus danilew- skyi.	Blood.
Anthropoides virgo	Hæmoproteus antigone.	Blood.
Anthus richardi rufulus	Hæmoproteus (?) anthi.	Blood.
Antigone antigone	Hæmoproteus sp	Blood.
	Proteosoma sp.	Blood.
Astur badius dussumieri .	Hæmoproteus asturi	Blood.
Athene brama	Hæmoproteus (?) bramæ	Blood.
	Leucocytozoon sp	Blood.
Calænas nicobarica	Hæmoproteus sp	Blood.
Centropus sinensis parroti	Hæmoproteus (?) sp	Blood.
Cerchneis tinnunculosus objurgatus.	Hæmoproteus cerchneisi	Blood.
Chloropsis aurifrons	Hæmoproteus sp	Blood.

Host.	Parasite.	Seat.
Aves (cont.).		
Chloropsis aurifrons david-	Leucocytozoon chlorop-	Blood.
80mi.	Proteosoma chloronada	Blood
Columba linia	Toronlama columba	Langantes
	Homoproteus columbo	Blood
Columba linia intermedia	Eimeria columba	Integtine
Columba sp.	Proteosoma columba	Blood.
Copsychus saularis	Hæmoproteus danilew- skui.	Blood.
	Proteosoma moruony	Blood.
Coracias benghalensis ben-	Hæmoproteus coraciæ .	Blood.
ghalensis.	Leucocytozoon coraciæ.	Blood and smears from lungs.
	Leucocytozoon melloi	Blood and smears from lungs.
Coracias benghalensis in- dica.	Hæmoproteus sp	Blood.
Corvus levaillanti macro- rhunchus.	Hæmoproteus corvusi	Blood.
Corvus splendens	Hæmoproteus corvusi .	Blood.
-	Proteosoma sp	Blood.
Coryllis beryllinus	Hæmoproteus sp	Blood.
	Proteosoma sp	Blood.
Cyanops flavifrons	Hæmoproteus sp	Blood.
	Proteosoma sp	Blood.
Dendrocitta rufa vagabunda Dicrurus macrooercus macro- cercus.	Hæmoproteus sp Hæmoproteus (?) dicruri	Blood. Blood.
Eagle	Hepatozoon adiei	Lungs and leuco- cytes.
Egretta intermedia inter-	Hæmoproteus herodiadis	Blood.
media.	Proteosoma herodiadis .	Blood.
Elanus cæruleus vooiferus.	Hæmoproteus (?) elani.	Blood.
Elathea jocosa	Hæmoproteus (?) oto- compsæ.	Blood.
	Proteosoma sp	Blood.
Emberiza fucata	Proteosoma sp	Blood.
Falco sp	Leucocytozoon sp	Blood.
Fulica atra	Toxoplasma fulicæ	Lung.
Gallinula chloropus	nulæ.	Blood.
ан н :	Proteosoma gallinulæ	Blood.
Gallus gallus	Babesia tropicus	Blood.
Garrulax alongularis	næmoproteus aanuew- skyi.	Blood,
Garruax leucolophus	Homoprotosia danilar	Dlood.
Garruius lanceolatus	skyi.	Blood,
Glareola pratincola	Hæmoproteus danuew- skyi.	Blood.
Glaucidium radiatum	Hæmoproteus glaucidii.	Blood.
Gracula indica	rroteosoma sp.	Blood.
Gymnoris xanthocollis	riæmoproteus gymno- rhidis.	B1000.
Halcyon smyrnensis	Hæmoproteus (?) halcy- onis.	Blood.
Haliaeëtus leucoryphus	næmoproteus sp	Blood.

Host.	Parasite.	Seat.
Ianthocincla rufogularis .	Leucocytozoon sp	Blood.
Kittacincla macroura	Hæmoproteus danilew- skyi.	Blood
Leptocoma zeylonica	Hæmoproteus raymundi	Blood, lungs, spleen.
Leiothrix luteus	Leucocytozoon sp	Blood.
Maclolophus xanthogenys.	Hæmoproteus machlo- lophi.	Blood.
Melophus melanicterus	Hæmoproteus danilew- skyi.	Blood.
Mesia argentauris	Proteosonia sp Hæmoproteus danilew-	Blood. Blood.
Nettapus coromandelianus.	Hæmoproteus danilew- skyi.	Blood.
Oreicola ferrea	Leucocytozoon sp	Blood.
Oriolus oriolus kundoo	Hæmoproteus orioli	Blood.
Orthotomus sutorius	Hæmoproteus wenyoni.	Blood, lungs, liver.
Otus bakkamæna	Hæmoproteus sp	Blood.
Otus bakkamæna var. malabarica.	Hæmoproteus danilew- skyi.	Blood.
Passer domesticus	Proteosoma passeris	Blood.
Passer sp	Toxoplasma sp	Blood.
Pastor roseus	Hæmoproteus (?) pas- toris.	Blood.
Pavo cristatus	Hæmoproteus rileyi	Blood.
Poking robins	Leucocytozoon sp.	Blood.
Platalea leucorodia major.	Hæmoproteus (1) plata- leæ.	Blood.
Propasser rhodochrous	Hæmoproteus danilew- skyi.	Blood.
Prunella strophiata jerdoni	Hæmoproteus danilew- skyi.	Blood.
Psittacula alexandri fasci- ata.	Hæmoproteus sp	Blood.
~ • •	Leucocytozoon sp	Blood.
Saxicola caprata	Toxoplasma sp	Blood.
<b>A</b> 1 <b>B</b> 1	Proteosoma sp	Blood.
Strix ocellata	Hæmoproteus sp	Blood.
Sturnia malabarica	Hæmoproteus sturni	Blood.
Sturnus vulgarus poltarat- skyi. Tenbrodornis pondicerianus	Proteosoma præcox	Blood.
pondicerianus.	dornis.	Diood.
ornata.	Hæmoproteus (!) therei- cerycis.	Blood.
Inereiceryx zeyianicus zey- lanicus.	erycis var. zeylonica.	Blood.
Tragopan satyra	Proteosoma præcosx	Blood.
Turdoides striatus striatus.	Hæmoproteus sp	Blood.
Turaus boulboul	Proteosoma sp.	Blood.
Upupa epops orientalis REPTILIA.	Hæmoproteus upupæ .	Rlood.
Agama tuberculata	Hæmogregarina thom-	Blood.
<b>n n</b>	soni.	101.1
Bungarus cœruleus	n æmogregarina sp	Blood.

Host.	Parasite	Seat.
REPTILIA (cont.).		
Calotes versicolor	Isospora caloti	Intestinal epithe-
Calotes versicolor major	Karyolysus jorgei	Blood, and endo- thelial cells of
Chinemys reevesii	Hæmogregarina rara Hæmogregarina stepa- nowiana.	Blood. Blood.
Chitra indica	Eimeria mitraria Myxidium danilewskyi. Hæmoproteus metchni- kovi.	Intestine. Kidney. Blood.
Chrysopelea ornata Coluber blumenbachii Coluber helena Coluber sp Crocodilus porosus Dendrophys pictus	Anaplasma sp Hæmogregarina sp Hæmogregarina sp Hæmogregarina sp Hæmogregarina hankini Hæmogregarina sp	Blood. Blood. Blood. Blood. Blood. Blood. Blood.
Dipsadomorphus ceylonen- sis. Dipsadomorphus forstenii. Dryophis mycterizans	Hæmogregarina sp Hæmogregarina sp Hæmogregarina sp	Blood. Blood. Blood.
Eryx conicus Eryx johnii Gavialis gangeticus	Hæmogregarina canutei Hæmogreyarina sp Hæmogreyarina han- kini.	Blood. Blood.
Geoemyda trijuga	Eimeria kermorganti Hæmogregarina nicoriæ Myxidium sp	Spieen. Blood. Gall-bladder.
Hemidactylus brookt	fizmogregarna roari- guesi. Hæmoproteus kopki Anaplasma sp Isospora knowlesi Eimeria flaviviridis	Blood. Blood. Alimentary canal. Gall-bladder and intestine.
Hemidactylus leschenaulti.	Eimeria hemidactyli Eimeria knowlesi Hæmogregarina leschen- aultii.	Intestine. Intestine. Blood.
Hemidactylus triedrus Hemidactylus sp Kachuga smithi Kachuga tectum Lissemys punctata granosa.	Hæmoproteus simondi . Hæmogregarina tiedri . Hæmoproteus sp Isospora knowlesi Hæmogregarina mesnili Hæmogregarina laver-	Blood. Blood. Blood. Gall-bladder. Blood. Blood.
	Hæmogregarina mala- barica. Hæmogregarina nicoriæ Hæmogregarina vittatæ. Hæmogregarina zavieri.	Blood, liver, bone- marrow, etc. Blood. Blood, lungs, spleen, liver. Call bledder and
	Myxidium sp Zschokkella prashadi	Gall-bladder. Gall-bladder.

Host.	Parasite.	Seat.
Lizards	Babesia tropicus	Blood.
Naja bungarus	Hæmogregarina sp	Blood.
Naja naja	Hæmogregarina najæ	Blood.
	Hæmogregarina sp	Blood.
	Isopsora minuta	Alimentary canal.
	Finneria najæ	Rectum.
Main wain soon atom	Anaplasma sp.	Blood.
Naja naja var. aira	Hamogregaring sp	Blood
Nation minator	Eimeria sulindring	Biolog.
Marria processi	Eimeria niecatori	Rectum
Python molurus	Hæmogregarina pytho-	Blood.
Putton an	Homouragering or	Blood
<i>1 gaton</i> sp	Puthonella bengalensie	Intesting
Testudo emus	Hæmonrenaring testu.	Blood
1. 680000 Cings	dinis.	Dioou.
Trionux gangeticus	Muxidium mackiei	Kidney.
Tropidonotus asperrimus .	Hæmogregarina mira-	Blood.
- · · · · · · · · · · · · · · · · · · ·	bilis.	
Tropidonotus piscator	Hæmoyregarina mira- bilis.	Blood.
Tropidonotus stolatus	Hæmogreyarina sp	Blood.
Varanus monitor	Hæmogregarina sp	Blood.
Vipera russellii	Hæmoyergarina sp	Blood.
Zaocys mucosus	Hæmogregarina sp	Blood.
Amphibia.		
Bufo melanostictus	Hæmogregarina nucleo- bisecans.	Blood.
	Isospora wenyoni	Small intestine.
	Eimeria laminata	Small intestine.
	Zschokkella prashadi .	Gall-bladder.
Rana esculenta	Anaplasma sp.	Blood.
Rana hexadactyla	Hæmogregarina berest- neffi.	Blood,
Rana limnocharis	Hæmogregarina berest- neffi.	Blood.
	Hæmogreg <b>a</b> rina magna.	Blood.
	Lankesterella monilata.	Blood.
Rana tigrina	Hæmogregarina berest- neffi.	Blood.
	Hæmogregarina magna.	Blood.
	Lankesterella minima .	Blood.
	Lankesterella monilata.	Blood.
	Anaplasma sp.	Blood.
Dragma	zsспоккеца prasnaai .	Gall-Diadder.
FISCES.	<b>T</b>	<b>T</b> /
Atobatis narinari	Ermeria southwell	Intestine.
Amphipnous kuchia	Unioromyxum amphi-	Gall-bladder.
Barilius barna	Spherospora sp.	Under the scales
Batrachus arunniens	Eimeria sp.	Intestine.
Cirrhing mrigala	Myxobolus calbasur	Liver.
Clarias batrachus	Myxobolus calbasui	Ovary and liver.
	Myxidium sp	Gall-bladder.
Coilia dussumieri	Eimeria sp.	Intestine.
SPOR.	-	D

#### Parasite.

Host.	Parasite.	Seat.	
PISCES (cont.).			
Engraulis mystax Epinephelus diacanthus Epinephelus tauvina Gobioides rubicundus	Eimeria sp Eimeria sp Eimeria sp Ceratomyxa gobioidesi.	Intestine. Intestine. Intestine. Liver, gall-bladder	
Harpodon nehereus Kaila katla Labeo rohita	Eimeria harpodoni Myxobolus sp Thelohanellus rohitæ Thellohanellus seni	Alimentary canal. Gills. Median and caudal	
Macrones gulio	Ceratomyxa gobioidesi .	Liver, gall-bladder	
Ophiocephalus punctatus .	Henneguya sp Myxidium sp	Gills and muscles. Gall-bladder.	
Otolithus ruber	Eimeria sp Henneguya otolithus	Intestine. Bulbus arteriosus.	
Plotossus canius Rasbora daniconius	Eimeria sp Myxobolus nodularis . Myxobolus sp	Intestine. Muscles. Subcutanens tis-	
Saccobranchus fossilis Sillago sikama Thyrsoidea macrurus	Myxidium sp Eimeria sp Hæmogregarina thyrsoi- deæ.	Gall-bladder. Intestine. Blood.	
Trichiurus savala Trichogaster fasciatus	Eimeria sp Hæmogregarina sp Ceratomyxa gobioidesi .	Intestine Blood. Liver, gall-bladder kidney, ovary.	
MOLLUSCA.			
Pachelabra mæstra	Adelea pachelebræ	Intestine and di- gestive glands.	
ARACHNIDA.			
Lælaps echidninus Porocephalus pattoni Rhipicephalus sanguineus.	Hepatozoon muris Hæmogregarina sp Hepatozoon canis	Body. Body. Body.	
INSECTA.			
Aëdes (Stegomyia) ægypti.	Lankesteria culicis	Stomach and Mal- pighian tubes.	
	Laverania malariæ	Body.	
Aëdes (Stegomyia) albopic- tus.	Lankesteria culicis	Stomach and Mal- pighian tubes.	
Anopheles annularis	Plasmodium cynomolgi. Plasmodium malariæ Plasmodium vivaz	Salivary glands. Body. Body.	
Anopheles barbirostris	Thelohania legeri	Adipose tissue of larvæ.	
Anopheles culicifacies	Laverania malariæ Plasmodium vivax Plasmodium malariæ Plasmodium malariæ	Body. Body. Body.	
Anopheles fluviatilis	Laverania malariæ Plasmodium vivax	Body. Body. Body. Body.	
Anopheles fuliginosus	Thelohania legeri	Adipose tissue of larvæ.	

Host.	Parasite.	Seat.
Anopheles funestus	Laverania malariæ	Body. Body
	Plannodium maar	Body.
	Thelohania legeri	Adipose tissue of
	Thelohania obscura	Adipose tissue of
Anopheles hyracanus	Thelohania indica	Adipose tissue of
	Thelohania legeri	Adipose tissue of larva.
Anopheles jeyporiensis	Laverania malariæ	Body.
	Plasmodium malariæ.	Body.
Anonheles maculatus	Lawrania malariz	Body.
Anopheces machines	Plasmodium malarix.	Body.
	Plasmodium vivax	Body.
	Plasmodium cynomolgi	Salivary glands.
Anopheles minimus	Laverania malariæ	Body.
	Plasmodium malarix.	Body.
Amembolas mbilingingania	Plasmoaium vivax	Body. Body
Anophetes phurppinensis.	Plasmodium malariz	Body.
	Plasmodium vivax	Body.
$An opheles\ pseudojamesi$ .	Thelohania legeri	Adipose tissue of
Anopheles ramsayi	Thelohania legeri	Adiposo tissue of larvæ.
Anopheles splendidus	Plasmodium cynomolgi.	Salivary glands.
Anopheles stephensi	Laverania malariæ	Body.
	Plasmodium malariæ	Body.
4 7 7 7 1	Plasmodium vivax	Body.
Anopheles sundarcus	Laverania malariæ	Body. Body
	Plasmodium vivar	Body
Anopheles subpictus	Proteosoma præcox	Body.
	Thelohania legeri	Adipose tissue of larvæ.
Aulacophora foveicollis	Caulocephalus crenata.	Alimentary canal.
Bombyx mori	Nosema bombycis	All tissues.
Cimex rotundatus	Novema aurer	Gut, salivary glands, and ovaries.
Ctenocephalus canis	Nosema ctenocephali	Digestive tract of larvæ.
Culex fatigans	Proteosoma præcox	Body.
	Microsporidian (indet.)	Nerve cord of imago.
Сиleх вр	<i>Eimeria</i> (?) sp	Stomach cells.
Gryllus sp	Leidyana gryllorum	Gizzard and mid- gut.
	Gregarina oviceps	Gizzard and mid- gut.
Hæmatopinus stephensi	Hepatozoon gerbilli	Body.
Hæmatopinus sp	Hepatozoon funambuli.	Body.
Lepisma saccharina	Gregarina aciculata	Mid-gut.
Phishotomus anasytics	Gregarina cornwalli	Alimentary concl
r nievotomus argentipes	лапкезиети таские	and hæmocœle.
		D 2

Host.	Parasito.	Seat.
INSECTA (cont.).		
Phlebotomus papatasi	Lankesteria mackiei	Alimentary canal
Stegomyia sp	Microsporidian (indet.)	Nerve cord of
Triptroidesi dofleini	Lankesteria tripterodesi.	Hæmocœle and respiratory sys- tem
Xylocopa æstuans	Liedyana xylocopx	Gizzard and mid- gut.
MYRIOPODA.		
Combolopsis sp	Hyalosporina combo- lopяів,	Alimentary canal.
Cormocephalus dentipes	Adelina schellacki	Intestine.
Diplopoda sp	Stenophora ellipsoidi .	Mid-gut.
Polydesmus sp	Hyalosporina rayi	Alunentary canal.
Scolopendra sp.	Grebneckiella navilla	Alimentary and
Strongytosoma contortipes.	Hydomoticus (unatus	Alimentary canal.
Zikadesmus (?) sp	Stenophora khagendræ.	Mid-gut.
CRUSTACEA.		
Metaplax dentipes	Steinina metaplari	Intestine.
Parapeneopsis sculptilis .	Protomagalhænsia (?) attenuata.	Intestine.
	Hirmocystis (?) para- peneopsisi. Agaregata so	Intestine.
	2199/09000 Sp	LittoStino.
Der store sont		
Lumbriconereis en	Lecudina brasili	Intestino
Morphysa sanguinea	Bhatiella morphysie Ferraria cornucephali .	Mid-gut. Mid-gut.
OT TOOCH #TA	-	0
Allolohomborg galiginosg	Pharmalumustic com atti	Saminal mariala
Allolobophora fætida	Nematocystis cogneui . Nematocystis pluri- karyosomata.	Seminal vesicles.
Eutyphæus foveatus	Aikinetocystis singu- laris.	Cœlomic cavity.
Eutyphœus incommodus	Nematocystis stephen- soni.	Seminal vesicles.
Eutyphœus nicholsoni	Monocystis beddardi	Seminal vesicles.
Eutyphæus peguanus	Aikinetocystis singu- laris	Cælomic cavity.
Eutyphœus rarus	Aikinetocystis singu- laris.	Cœlomic cavity.
Eutyphœus spinulosus	Aikinetocystis singu- laris.	Cœlomic cavity.
Eutyphœus sp	Monocystis beddardi Dirhynchocystis alobosa.	Seminal vesicles. Seminal vesicles.
Megascolex trilobatus Pheretima barbadensis	A polocystis matthai Nematocystis vermicu- laris.	Seminal vesicles. Seminal vesicles.
Pheretima compta	Stomatophora coronata . Stomatophora diadema . Nellocystis birmanica .	Seminal vesicles. Seminal vesicles. Cœlomic cavity.

Host.	Parasite.	Seat.
Pheretima elongata	Rhynchocystis awatii Rhynchocystis mamil- lata.	Seminal vesicles. Seminal vesicles.
	Stomatophora bulbifera.	Seminal vesicles.
Pheretima heterochæta	Nematocystis hessei	Seminal vesicles.
	Nematocystis lumbri- coides.	Seminal vesicles.
	Gravallia avadrispina	Colomic cavity.
Pheretima posthuma	Monocystis bengalensis. Monocystis lloidi Monocystis pheretimi Stomatophora diadema. Dirhunchocystis alohosa	Seminal vesicles. Seminal vesicles. Seminal vesicles. Seminal vesicles. Seminal vesicles.
HIRUNDINEA.		
Ozobranchus shipleyi	Hæmogregarina nicoriæ	Body.
GEPHYREA.		
Dendrostoma signifer	Extremocystis dendro- stomi.	Cœlomic cavity.

#### Technique.

Methods for the examination and study of PROTOZOA are adequately dealt with in such works as Prowazek and Jollos (1921), Wenyon (1926), Hartmann (1928), Bělař (1928), Gatenby and Cowdry (1928), Knowles (1929) and Hegner and Andrews (1930). The principal methods followed in the study of the SPOROZOA are given here for the benefit of those taking up the study of the group.

Examination in the Living Condition.-It is always desirable to make observations on the living organisms in the first instance. The specimens are mostly studied in a drop of the natural medium or body-fluid of the host-animal in which they are found. Any pressure of the cover-glass might cause deformities, and this should be guarded against by including in the preparation small bits of detritus or a hair which will support the cover-glass. If it is intended to continue the observations for some length of time, "cavity slides" may be used. The drop of fluid containing the organisms should be put on a circular cover-glass, the margin of which is smeared with melted paraffin or Czokor's wax, and the cover-glass inverted over the cavity. This will fix the cover-glass to the slide and prevent evaporation. The wax is made by heating together and mixing in a shallow tin provided with a lid equal weights of bees-wax and Venetian turpentine. The wax becomes a solid mass when cool, and can be applied by placing the heated portion of a wire on the wax and then passing it round the cover-glass or the slide. To study the developmental stages and natural movements the slide should be kept at the body temperature of the host when this is a warm-blooded animal.

Occursts of CocciDia escaping in the faces in an incompletely developed condition may be observed to complete their development, especially when mixed with a 5 per cent. solution of bichromate of potash. Malarial parasites, when observed as described above, will continue to develop for several hours, and the production of merozoites can sometimes be observed. Occasionally the removal of the blood from the Vertebrate host may induce the development of stages which normally take place inside the body of an Invertebrate host : thus the production of gametes from the gametocytes, fertilization, and the development of oökinetes may be followed in a drop of blood enclosed between a slide and a cover-glass. During the observations the cover-glass may be removed and permanent preparations made showing particular stages of development.

When the body-fluid containing the parasites is too small in amount to make a suitable preparation, physiological salt solution (0.7 per cent. solution of sodium chloride in distilled water for cold-blooded animals and 0.85 or 0.9 per cent. solution for warm-blooded animals) or Ringer's solution may be added. Ringer's solution consists of sodium chloride 0.8 grm., potassium chloride 0.02 grm., calcium chloride 0.02 grm., sodium bicarbonate 0.02 grm., distilled water 100 c.c.

Intra-vitam Staining.—Examination of the living organisms is facilitated by intra-vitam staining, different parts of the organism or its contents being coloured without killing the animal or affecting its movements. For this purpose neutral red or methylene blue are used in very dilute solutions (1 in 10,000). A drop of the stain may be allowed to dry on the slide and the fluid containing the organisms be placed on the area and covered with a cover-glass. The stains are slowly absorbed by the parasites. Neutral red assumes a bright cherry-red colour in acid and a brown colour in alkaline media, and thus serves to indicate the reaction of the substance which it stains.

Eosin is used as an indicator of the life or death of an organism. It will not stain living cytoplasm, and by running a dilute solution under the cover-glass the living organisms can be distinguished from those that are dead.

Cultivation.—Various methods have been adopted to provide favourable conditions of growth and multiplication, so as to afford large numbers of a particular organism outside the body of the host for extensive study or for the purposes of a class. No artificial medium has so far been discovered in which GREGARINIDA or COCCIDIA will grow: but various methods have proved successful for the cultivation of blood-inhabitivir parasites, as the malarial parasites and the Piroplasmits. These cultures are incubated at  $37^{\circ}$  to  $40^{\circ}$  C., and the forms which appear are those which normally occur in the Vertebrate host. Bass and Johns (1912) were the first to succeed in cultivating malarial parasites. Their methods, with slight modifications, were successfully followed by Ziemann (1913, 1914), J. G. & D. Thomson, (1913), Row (1917), Sinton (1922), and others. Knuth and Richters (1913), Ziemann (1913, 1914), and Thomson and Fantham (1914) were the first to apply these methods to the Piroplasmids. The technique of some of these methods is given below :—

Bass and Johns's Method. -- Withdraw 10 c.c. of malarial blood from a vein with a syringe, and transfer immediately to a defribinating flask containing 0.1 c.c. of 50 per cent. solution of dextrose. Defibrinate by stirring with a glass rod. Transfer the defibrinated dextrose blood to culture-tubes not less than 1.25 cm. in diameter and 12.5 cm. in depth. The quantity of blood in each tube may vary in depth from 2.5 to 5 cm., which will give a column of serum 1.25 to 2.5 cm. deep above the cells, when they have settled to the bottom of the tube. Incubate the tubes in the vertical position at a temperature of 40° C. The parasites live and develop in the red bloodcorpuscles just below the surface of the deposit. Carefully withdraw the cells from this layer by means of a fine pipette and examine at intervals. The young trophozoites will be seen to grow into schizonts and break up into merozoites. Some of these may enter other corpuscles and grow into schizonts, but as a rule those escaping from the corpuscles are devoured by the leucocytes. To remove the leucocytes centrifuge the defibrinated glucose blood at a speed sufficient to cause the leucocytes to occupy the upper layer of the deposit. Transfer the supernatant serum to flat-bottomed culture-tubes, filling to a depth of 1.25 to 2.5 cm. Pass a pipette into the middle of the deposit in the centrifuge-tube, draw off the red corpuscles with the parasites, and transfer them to the bottom of the culture-tubes. In this way, in the absence of the leucocytes, the parasites may complete two or three cycles; but it has not been found possible to cultivate them for more than three generations. For making subcultures, take normal blood, treat it in the same manner, and inoculate it by means of a pipette with infected blood from the previous culture.

Row (1917) and Sinton (1922) have devised modifications by which the growth of a single generation can be followed with a few drops of blood.

Row's Method.—Draw the blood from the finger into a small tube and defibrinate it in the same. Transfer the small quantity of defibrinated blood by means of a pipette to a small, flat-bottomed tube containing serum to which the requisite quantity of dextrose has been added. Place the small tube in a larger tube (the ordinary bacteriological potato-tube) provided with a constriction on which the smaller tube may rest. In the portion of the outer tube below the constriction put some solution of pyrogallic acid and add 2 or 3 c.c. of a 10 per cent. solution of sodium hydrate immediately before introducing the smaller tube. Cork the larger tube tightly with a rubber cork. The pyrogallic acid and sodium hydrate absorb the oxygen, and the culture thus takes place in an oxygen-free atmosphere.

Sinton's Method.-A specially constructed tube about 20 cm. in length is employed (fig. 2). To prepare this apparatus, take a tube of 0.4 to 0.5 cm. bore. Draw out one end as in an ordinary pipette and, slipping a narrow metal tube over the thin drawn-out portion while it is still soft, press upwards to produce a dilated bulb with its lower surface flattened (B) and with the thin drawn-out tube arising from it (A). Now heat the tube a short distance, about 1 cm., above the flattened surface and draw out till it forms a tube about 6 to 8 cm. long and 0.2 cm, wide (C). At the upper end of this narrow tube make a slight constriction (D), and about 0.4 to 0.5 cm. above it make another (E). Drop three glass beads (F) into the upper part of the tube (G), which is allowed to remain wider in diameter, and then draw out and bend as in a Wright's capsule (H). Keep the upper and lower drawn-out ends sealed and sterilize the whole tube.

To make the culture, open the tube at both ends, and bend the upper capillary portion at right angles to the rest, so that the apparatus may lie on the table with the open upper end pointing upwards. Insert the upper end into an already prepared Wright's capsule containing ascitic or hydroccele fluid, to which the requisite quantity of dextrose has been added, and allow the fluid to enter till the upper section is about a third or half full. Prick the carefully sterilized finger of a malarial patient, and allow five to ten drops of blood to run into the fluid in the tube. Now gently heat the dilated part of the lower end of the tube and seal off the narrow part below it. The air in the dilated part will cool and the blood-mixture will be drawn further into the tube. Now seal off the upper end Defibrinate the mixture by shaking the glass beads, also. and when this has been completed, swing round the tube rapidly so as to drive the defibrinated blood-mixture through the constriction into the lowermost part of the tube, so that it fills the dilated part and the narrow section above it. Heat the tube at the constriction above the column of fluid and seal off. The red corpuscles will settle to the flat bottom. Incubate the tube in a vertical position at a temperature of 35° to 38° C. In order to examine, open the tube, withdraw by a pipette the cells from the lower end, and seal again.

Maintenance of Parasitic Protozoa in Laboratory Animals.— In a laboratory strains of parasitic PROTOZOA may be maintained in suitable animals which have been found by previous examination not to harbour natural parasites of their own. This method serves the same purpose as cultivation, and it is easier to maintain a strain in an animal host than in a culture. Blood parasites, such as the malarial parasites and the piroplasmata, can be introduced by inoculating the blood from an infected to a clean host, and parasites that pass out in the



Fig. 2.—Sinton's apparatus for cultivation of malarial parasites in a small quantity of blood. The small Wright's capsule shown on the right contains the ascitic or hydroccele fluid. (After Sinton.)

encysted condition, such as COCCIDIA, can be mixed with the food and allowed to be ingested. The practice has been particularly successful in certain special cases.

The malarial parasites of birds can be maintained for a long time in canaries, sparrows, and other susceptible birds by inoculating blood from the one to the other. Some birds acquire heavy and fatal infections and others mild ones from which they recover. The piroplasmata of dogs are readily inoculable from one dog to another, but as fatal infections are often obtained, a number of animals will be required. Ticks fed on a dog will remain infective for long periods, and the parasites can thus be maintained in them. Strains of *Hæmoproteus columbæ* can be maintained in pigeons by breeding *Lynchia maura* in the cages in which they are kept. A fresh pigeon is introduced into the cage from time to time to hand over the infection from fly to fly. *Toxoplasma gondii* is also inoculable to a variety of laboratory animals.

Permanent Preparations of Fixed and Stained Protozoa.— Although it is important to study the living organisms in the first instance, the finer details of structure can only be studied in properly fixed and stained smears or films. Some of the methods of general application are described below.

Staining under the Cover-slip.—For staining large organisms, such as the Gregarines, the following method is used :—

Put wax feet at the corners of a square cover-slip, and invert it over a drop of the fluid containing the organisms. The wax feet should hold the cover-slip firmly to the slide. With a pipette run a little fixative at one side of the cover-slip, and draw it through by holding a piece of filter-paper at the opposite side. When the fixative has had time to act, wash it out by substituting another fluid (alcohol or water, as the case may be) and draw it through with filter-paper in the same manner. The stream should not be so violent as to wash away the organisms, but the substitution should be complete. Then run in the stain, allow it to act, and wash out and differentiate, if necessary controlling the process under the microscope. Dehydrate and then clear in clove-oil or xylol, and run in a very fluid Canada balsam. It is very important to see that the transfer from one fluid to another is not too rapid, as otherwise there is great risk of shrinkage, and also to see that the dehydration is complete.

The following is an indication of the length of time generally required, but should be regarded as no more than an indication. Fix in Bouin's fluid, 5 min.; wash in 70 per cent. alcohol, 5 min. (several changes); stain in borax-carmine, 5 min. or more; dehydrate with 70 per cent., 90 per cent., and absolute alcohol, 5 min. each, changing the absolute alcohol once or twice; clear by running in a mixture of clove-oil and absolute alcohol and then pure clove-oil; mount in Canada balsam by running the same under the cover-slip.

Preparation of Wet Fixed Films.—Films made on slides or cover-slips may be fixed with one of the fixing fluids, without being allowed to dry. After fixation the films are washed free of fixative, and stained and mounted like sections fixed on slides. The results shown by wet fixed films are far superior to those shown by dried films, described later. If the material to be examined is thick, it should be emulsified with physiological salt solution. In the case of blood, fluid from blood or serum culture media, tissues, fæces, in fact any liquid containing albuminous matter, the film will stick to the slide or the cover-slip during the processes of fixation and staining. The usual fixatives and stains are described hereafter.

Staining the Organisms fixed on a Cover-slip.—Make a number of smears of the fluid containing the organisms on cover-slips, and when the fluid has partially dried, invert the cover-slips and let them float on the surface of the fixative contained in a dish; or put a small drop of fluid containing the organism on a cover-slip and add with a pipette twice the quantity of hot fixative. When the organisms have been fixed on the cover-slips, pass successively through the washing fluid, alcohols, stain, clearing-fluid, etc., all these reagents being contained in shallow dishes. In all these subsequent stages put the cover-slips at the bottom of the fluid in the dish, with the face bearing the organisms upwards. Finally, remove the cover-slips and put them, face downwards, on slides on each of which a drop of Canada balsam has been put. In this way quite a large number of smears or preparations can be fixed and stained in practically the same time as would be taken to make one preparation.

Fixatives.—The following are commonly employed for fixing the PROTOZOA :—

(1) Concentrated solution of mercuric chloride, hot or cold: (2) Schaudinn's sublimate alcohol (2 parts saturated aqueous solution of mercuric chloride, 1 part absolute alcohol; immediately before use, add acetic acid to the quantity to be used to the strength of 5 per cent.): (3) Zenker's fluid (mercuric chloride 5 grm., potassium bichromate 2.5 grm., sodium sulphate 1 grm. in 100 c.c. of distilled water, with 2.5 to 5 per cent. of glacial acetic acid added before use):
(4) Bouin's fluid (saturated aqueous solution of picric acid 75 parts, formol 25 parts, and acetic acid 5 parts): (5) Bouin's alcoholic fixative (picric acid 1 grm., 80 per cent. alcohol 150 c.c., formol 60 c.c., acetic acid 15 c.c.): (6) formalin: and (7) vapour of 4 per cent. solution of osmic acid. Fixation is usually complete in 15 to 30 minutes.

Bouin's fixatives are the simplest to use, as the picric acid is more easily washed out after fixation than the mercuric chloride contained in the others. After Bouin's fixative, wash the films in several changes of distilled water till all the picric acid has been removed. In the case of alcoholic Bouin, if an aqueous stain is to be used, commence washing in 70 per cent. alcohol and, passing the film through graded alcohols, bring down to distilled water. After Schaudinn's fluid, wash in several changes of 70 per cent. alcohol to remove the mercuric chloride; then wash in 70 per cent. alcohol to which a few drops of Wiegert's iodine solution have been added; finally, wash in 70 per cent. alcohol to which a drop of 1 per cent. solution of sodium thiosulphate has been added, in order to remove all traces of iodine. Bring down through successive grades of alcohol into distilled water if it is desired to use a watery stain; a stay of a few minutes in each solution will suffice. After fixation with Zenker's fluid, wash successively with distilled water, distilled water with a few drops of iodine solution, distilled water with a trace of 1 per cent. solution of sodium thiosulphate, and distilled water.

Staining Methods.—The following stains are usually employed for staining PROTOZOA :—

Borax-carmine.—Fix in Bouin's fluid for 10 to 20 minutes according to bulk and permeability. Wash out in 70 per cent. alcohol (several changes). Stain in borax-carmine till thoroughly penetrated; 15 minutes are usually enough for small objects. Differentiate in acid alcohol (70 per cent. alcohol to which hydrochloric acid is added to the strength of 1 per cent.), controlling under the microscope. Dehydrate in 90 per cent. to absolute alcohol. Put in a mixture of cloveoil and absolute alcohol, equal parts. After a few minutes transfer to pure clove-oil, and leave there till cleared. Mount in Canada balsam.

Delafield's Hæmatoxylin.—Fix in Schaudinn's fluid. Wash in 30 per cent. alcohol, and bring down through 10 per cent. alcohol to distilled water. Add a few drops of Delafield's hæmatoxylin solution to a watch-glass full of distilled water. Leave in stain for a few minutes to an hour or more according to bulk. Bring up from distilled water through ascending grades of alcohol to 70 per cent. alcohol; differentiate in 70 per cent. acid alcohol. Dehydrate; clear; mount.

*Heidenhain's Iron Hæmatoxylin.*—Fix in Schaudinn's fluid for 10 to 30 minutes, according to the size and permeability of the object. Bring down to 30 per cent. and 10 per cent. alcohol to distilled water. Wash out the fixative thoroughly. Transfer to 4 per cent. solution of the violet crystals of iron alum (sulphate of iron and ammonium) in distilled water for  $\frac{1}{2}$  hour to 12 hours according to the size of the organisms. Stain in Heidenhain's aqueous hæmatoxylin solution (about 0.5 per cent.) for 30 minutes to several hours. Wash in distilled water. Differentiate in 1 per cent. iron alum solution till the granules in the nuclei are distinct, the films being removed into distilled water and examined on a slide with the microscope from time to time. When differentiation is complete, wash in several changes of distilled water and then for half an hour or more in running tap-water. Counterstain, if desired, with a 1 per cent. solution of eosin or orange G. Dehydrate by bringing through graded alcohols to absolute alcohol. Clear in xylol. Mount in Canada balsam.

Dobell's Iron Hæmatein.-Fix in Schaudinn's fluid. Bring down through 30 per cent. and 10 per cent. alcohol to distilled After washing, bring up through various grades of water. alcohol to 70 per cent., and from that transfer to 1 per cent. solution of iron alum in 70 per cent. alcohol for 10 minutes (the solution is made by dissolving 1 grui, iron alum in 23 c.c. warm distilled water and adding 77 c.c. of 90 per cent. alcohol). Rinse in 70 per cent. alcohol. Stain in 1 per cent. solution of hæmatein in 70 per cent. alcohol for 10 minutes. Rinse in 70 per cent. alcohol. Differentiate films in original alum solution and sections in 70 per cent. acid alcohol. Wash in several changes of 70 per cent. alcohol. Dehydrate; clear; mount. The whole process may be carried out in 30 minutes. Light green in 90 per cent. alcohol may be used as a counterstain.

Mayer's Hæmalum.—To prepare the stain dissolve 1 grm. of hæmatin in 50 c.c. of 90 per cent. alcohol, dissolve 50 grms. of alum in 1000 c.c. of water, and mix up the two solutions. Keep a crystal of thymol in the bottle to prevent growth of fungi, etc.

When using the stain, add a few drops of the stain to a Petri dish of distilled water, and leave the films in for several hours or overnight. Wash the films in running tap-water till they are quite blue. Dehydrate by bringing up though various grades of alcohol to absolute alcohol. Clear in xylol and mount in Canada balsam.

Mayer's Acid Hæmalum.—This strain is prepared from Mayer's hæmalum by adding acetic acid to a strength of 2 per cent. It is used in the same manner, but has less tendency to overstain.

If overstaining occurs with either of the above stains, place the films in acid alcohol. After they are decolorized, wash the films well in running water till the acid is completely neutralized and they are blue.

Mallory's Eosin and Methylene Blue.—This is recommended for sections of tissues containing parasites. Fix in Zenker's fluid. Wash out the fixative in running water for several hours. Stain in warm 5 per cent. aqueous solution of eosin for 20 minutes or longer, wash in water, and stain in Unna's alkaline methylene-blue solution diluted with 4 or 5 parts of water for 10 to 15 minutes. Wash in water, differentiate in 95 per cent. alcohol, controlling under a microscope, until sections are pinkish but nuclei deep blue. Dehydrate quickly and mount.

Mallory's Triple Stain.-This is also recommended the full

sections. Fix in Zenker's fluid. Thoroughly wash out the fixative for several hours in gently running water. Stain sections in 0.5 per cent. aqueous solution of acid fuchsin for 2 to 4 minutes, and transfer to the second solution (consisting of aniline blue soluble in water (Grübler) 0.5 grm., orange G (Grübler) 2 grm., 1 per cent. aqueous solution of phosphomolybdic acid 100 c.c.) for 10 to 20 minutes or longer. Wash and differentiate the sections in tap-water, dehydrate rapidly, clear, and mount. Lund (1933) recommends Zenker's fluid with 2.5 per cent. acetic acid, staining the sections for 2 minutes in Mallory No. 1 and 1 minute in Mallory No. II, then dipping rapidly into 95 per cent. and absolute alcohol, blotting quickly between each change, and then elearing in xylol for about 3 minutes.

Preparation of Dried Films on Slides.—Films of blood, intestinal contents or other fluids containing parasites are usually made for diagnostic purposes. Films of solid organs, such as the liver or the spleen, are made by smearing a piece of the organ lightly across the slide or dabbing the slide with the freshly-cut surface. The film is dricd as rapidly as possible by waving it in the air. Distortion and disintegration of the parasites may take place in the process of making the films and subsequent drying; to avoid this expose the film to osmic acid vapour for ten to fifteen seconds before allowing it to dry.

Thin Blood-films.-To make blood-films take perfectly clean glass-slides, free from any trace of grease, and lay them flat on a clean piece of paper. Sterilize a needle in the flame, and prick the dorsum of a finger just below the root of the nail. Pick up one of the clean slides, invert it, and towards one end just bring it in contact with the oozing blood. Too large a drop must not be taken. Re-invert the slide on the table and, taking another slide, with a smooth even edge as a spreader hold in at an angle of 45°, touching the drop so that a thin film of blood runs between the edge of the spreader and the slide. When the blood has spread along the edge, push the spreader fairly rapidly toward the other end. The blood must follow the spreader and not be pushed before it. A thin film will result covering about one-half of the slide. Allow the film to dry in air, covering it, if necessary, with a Petri dish to protect it from flies, dust, etc. In monsoon weather hæmolysis may take place before the film has time to dry. and at such times the film should be held, as soon as made, over a spirit-lamp flame to dry. Films are usually stained by one of the modifications of Romanowsky's stain.

su Staining the Dried Blood-films.—The films, as prepared wash in .vy be stained with the original Romanowsky's stain, an hour we of the many modifications, such as Leishman's stain or Giemsa's stain. The action of all such stains depends on certain loose chemical combinations. Medicinal (not pure) methylene-blue contains a number of oxidation products, the most important of which is methylene-azur. When watery or alcoholic solutions of methylene-blue and of eosin are mixed together a series of loosely combined chemical bodies are formed. These different compounds possess different affinities for different cell-structures, and thus differential staining results. The red blood-corpuscles stain a transparent pink or orange colour ; the nuclei of leucocystes, shades of violet ; eosinophile granules in the coarsely granular leucocytes, red ; the cytoplasm of malaria and other blood-inhabiting protozoal parasites a bright "Cambridge" blue, and their chromatin a bright ruby-red.

The Original Romanowsky's Stain.—In large laboratories the original Romanowsky's stain is still employed, as it is more economical than Leishman or Giemsa. Two stock solutions are needed. For preparing Solution A take medicinal (not pure) methylene-blue 1 grm., pure sodium carbonate 0.5 grm., distilled water 100 c.c., dissolve and place the stain either in the  $37^{\circ}$  C. incubator or in full sun-light for 2 or 3 days. The solution should acquire a deep purple colour before it is ready for use. For Solution B, dissolve eosin, extra B.A., water soluble 0.1 grm. in 100 c.c. of distilled water.

Fix the film for 5 minutes in methyl alcohol, or for 10 minutes in ordinary alcohol. Wash with distilled water and transfer to a clear Petri dish. Dilute 1 part of solution A with 19 parts of distilled water. Similarly dilute 1 part of solution B with 19 parts of distilled water. Mix equal parts of the diluted solutions A and B, and pour the mixed stain, immediately after mixing, into the Petri dish to cover the slide. Stain for half an hour or longer. On tilting the Petri dish a red stain should be seen at the edge, which will indicate that the staining is proceeding properly. Wash off the stain with distilled water and differentiate in a bath of distilled water. When the film commences to turn pink, remove from the bath and allow it to dry by slanting it against a vertical surface.

Leishman's Stain.—To prepare the stain thoroughly clean a glass (not porcelain) pestle and mortar and rinse out the mortar with a little pure methyl alcohol. Weigh out 0.15 grm. of Leishman's stain powder (Merck's or Grübler's). Measure into a perfectly clean glass cylinder 100 c.c. of methyl alcohol, purissimum, acetone-free (Merck's). Add a little of the alcohol to the stain in the mortar and grind. Add more alcohol and grind. Drain off the dissolved stain into a stoppered bottle. Continue to add the alcohol in portions and grind until every particle of the stain has gone into solution, and use the full 100 c.c. of methyl alcohol. Next ripen the stain by placing the bottle of stain overnight (but not longer) in the  $37^{\circ}$  C. incubator.

In order to stain, lay the blood-film, film side upwards, on a staining rack. Drop the stain on to the slide until the whole surface is covered with the stain. The methyl alcohol in the stain fixes the film. This takes half a minute only; after that interval drop on to the slide double the number of drops of pure distilled water. By tilting the end of the slide allow the water and stain to mix thoroughly, and allow the stain to act for 5 minutes or more. Wash the stain off by immersing the slide in distilled water. Put the slide to soak in a clean Petri dish containing fresh distilled water and rock gently. The film turns at first greenish-blue, then pink. This takes about half a minute or less. When the film is just turning pink remove the slide and lean it against a vertical surface to dry.

Giemsa's Stain.—To prepare the stain grind in a glass mortar with a glass pestle 3 grm. of azur-II-cosin and 0.8 grm. of azur-II (Grübler's or Merck's) into thorough solution in 250 c.c. of the purest anhydrous glycerine. Add 250 c.c. of the purest acetone-free methyl alcohol and mix thoroughly. Allow to stand overnight and next day filter the stain through filter paper. Prepared Giemsa's stain is stocked by many firms, and is also supplied by the Central Research Institute, Kasauli.

Fix the film by covering it for 3 to 5 minutes with pure methyl alcohol or by dipping it for 10 minutes into absolute alcohol, and wash thoroughly in distilled water. Dilute 1 part of Giemsa's stain with 14 parts of distilled water (10 or 15 drops in as many c.c. of distilled water), and pour it over the slide placed in a Petri dish. Stain for half an hour or longer. Remove the slide, flush with distilled water, and put it in a bath of fresh distilled water until it commences to turn pink. Remove the slide and let it dry by leaning it against a vertical surface.

The Panoptic Method of Staining.—Lay the slide on a staining rack and cover it for half a minute with undiluted Leishman's stain; dilute the stain with double the number of drops of distilled water, and allow to stain for 5 or 10 minutes. Wash the film with distilled water. Then lay the slide in a Petri dish and flood with diluted Giemsa's stain, one drop to each e.e. of water. Stain for 1 to 24 hours, covering the Petri dish to prevent evaporation. Wash the film with distilled water and transfer it to a bath of 1 in 1000 acetic acid. When the film begins to turn pink, remove, wash rapidly with distilled water, and slant the slide against a vertical surface to dry.

A more rapid alternative method is as follows :---Cover the film with undiluted Leishman's stain for half a minute ; dilute the stain with double the volume of diluted Giemsa's stain, one drop to each e.e. of water, and mix thoroughly. Differentiate in distilled water, as in the case of Leishman's stain.

Thick Blood-films.—The thick film method is specially applicable to those cases in which the patient has already taken a small dose of quinine and the malarial parasites are apt to be missed in thin films. Knowles (1928) recommends taking three thin films and one thick one from every suspected case of malaria. The thin films are stained and examined whilst the thick film is drying, and if no parasites are detected in the thin films the thick film may be proceeded with. Knowles and Das-Gupta (1924 a) advocate the following method for preparing thick films :-- Take a perfectly clean slide, prick the patient's finger, and bring the slide into contact with the issuing blood so that four drops are placed at the corners of a small square about half an inch across. The drops should not be too large nor too small. With a round needle pool the four drops so as to spread the film into an even thick film eovering the half-inch square. Puddling should be avoided and the film must not be made too thick. The film should be kept covered, and will take about two hours to dry. Lay the slide on a staining rack and dehæmoglobinize the film by gently flooding the slide with a mixture of 2.5 per cent. solution in distilled water of glacial acetic acid, 4 parts, and 2 per cent. solution in distilled water of crystalline tartaric acid, 1 part. The dehæmoglobinized film should have a greywhite colour. As soon as the process is complete drain off the fluid by gently tilting the slide. Next flood the slide with methyl alcohol and allow it to remain on for 3 to 4 minutes. Drain off the methyl alcohol and wash the film thoroughly in distilled water so as to remove every trace of acid. Stain with diluted Giemsa's stain, one drop to each c.c. of water, for 15 minutes or longer. Differentiate in the usual wav with distilled water. Do not blot the film, but let it dry by slanting it against a vertical surface.

SPOR.

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## Phylum **PROTOZOA.**

### Class SPOROZOA Leuckart, 1879.

Protozoa which are exclusively parasitic in their mode of life, and live in the cells or body fluids of Vertebrate or Invertebrate hosts. They produce resistant spores at some stage or another of their life-cycle.

The general organization of the class and the basis of classification into subclasses and orders has already been discussed. Following Reichenow, the class SPOROZOA is divided into four subclasses, as follows :---

1 (2	). Trophic and reproductive phases typi- cally distinct; trophozoite becoming fully developed before reproduction begins. Sporozoites are Gregarinulæ.	[Schaudinn, p. 50. Teiosporidia
2 (1	). Trophic and reproductive phases usually	
	overlap; the still growing or even quite	
	young trophozoite may begin to form	9
2 14	Spores with one or more thread-can-	э. Гр. 328
0 ( <del>*</del>	sules	Cnidosporidia Doflein.
4 (3	. Spores without thread-capsules	5.
5 (6	). Cysts forming long rod-like masses	
	(Miescher's tubes). Spores crescentic,	
	with one end rounded and the other pointed. Parasites of striped muscle of Vertebrates	[p. 36]. Sarcosporidia Balbiani,
6 (5)	. Spores large, containing a single volumi-	-
	nous nucleus. Simple type of develop-	[& Mesnil, p. 369.
	ment	Haplosporidia Caullery

# I. Subclass TELOSPORIDIA Schaudinn, 1900.

TELOSPORIDIA are SPOROZOA in which the trophic and reproductive phases are typically distinct, the animal becoming full grown before either asexual reproduction (*schizogony*) or sexual reproduction (*gamogony* or *sporogony*) begins.

The great majority are intracellular parasites, at least during part of their life-history. The parasites are uninucleate in the early period of growth. During growth (as in COCCIDIA and HEMOSPORIDIA), or after growth is completed (as in GREGARINIDA), the nucleus divides by repeated mitotic divisions, and the organism breaks up into as many daughter

individuals as the number of nuclei. This process of multiple fission may be an asexual process (schizogony), and the resulting daughter individuals (merozoites) will grow into adult trophozoites, or the daughter individuals may all be gametes. In the former case also, after schizogony has been repeated a number of times, some of the merozoites develop into gametocytes, which give rise to gametes. The gametes conjugate, the process being known as syngamy, and form zygotes. The zygote then becomes enclosed in a resistant eyst, known as the oöcyst. Inside the oöcyst the zygote may either divide directly into a number of motile vermiform bodies called sporozoites, or the zygote may first divide into separate bodies called sporoblasts, which become encysted in secondary cysts known as sporocysts; within each sporocyst the sporoblast gives rise to a number of sporozoites and a residual body. The mature occyst of this latter type thus contains a number of sporocysts, each containing many sporozoites.

The infection of a new host is brought about by the contaminative or inoculative method, and the sporozoites seek their way to the particular type of cell which they usually parasitize, viz., an epithelial cell (GREGARINIDA, COCCIDIA) or a blood-corpuscle (HÆMOSPORIDIA). The adult forms of GREGARINIDA are invariably extracellular or lumen dwelling, young growing stages alone being intracellular. Adult forms of Coccidia and Hæmosporidia are persistently intracellular. young, adult, and reproductive phases all occurring inside a host-cell. In the majority of GREGARINIDA (EUGREGARINARIA). the sporozoite grows directly into a gametocyte which produces gametes. In the SCHIZOGREGARINIDA schizogony also takes place. In the COCCIDIA and HEMOSPORIDIA regular alternation of the asexual method or schizogony, and the sexual method or sporogony, is the rule, and this alternation of generations frequently takes place in two distinct hosts. In GREGARINIDA and the HÆMOSPORIDIA the sporozoites are formed directly by divisions of the zygote; in COCCIDIA the zygote encysts and divides into sporoblasts which become sporocysts, and the occyst thus contains a number of sporocysts, inside which the sporozoites are developed. When sporocysts are present, the oöcyst is termed sporoeystid, and, according to the number present, is described as disporocystid, tetrasporocystid, or polysporocystid. The sporocysts are described as monozoic, dizoic, tetrazoic, octozoic, etc., according to the number of sporozoites present in each.

The subclass is divided into three orders, as follows :---

Mature	trophozoite	extracellular,	large ;	[em. Doflein, p. 52.
zygote	non-motile	sporozoites	within	
a spore		•••••	••••	Gregarinida A. Schneider, E 2
113. 209.

Mature trophozoite intracellular and small.	
Zygote non-motile; sporozoites within	Constate To Junet of 115
a spore	Coccidia Leuckart, p. 115
Zygote motile; sporozoites without	[sky, em. Donein, p. 209
envelope	Hæmosporidia Danilew-

# I. Order GREGARINIDA A. Schneider, emend. Doflein, 1901.

The GREGARINIDA are chiefly coelozoic or lumen-dwelling parasites of Invertebrates, especially Arthropods and Annelids, usually inhabiting the digestive tract, less frequently the coelome or the vascular system. They are typically intracellular only in the early part of their growth, i. e., in the



Fig. 3.—Life-cycle of a typical Eugregarine. A. syzygy; B, union of two sporonts; C, encystment of the two sporonts and nuclear multiplication in each; D, arrangement of the nuclei on the surface (pearl stage); E, gamete formation : F. gametes set free and their union in pairs; G, cyst containing the spores; H, liberation of the spores through the sporoducts. (From Reichenow, after Rähler, Schnitzler, and Schellack.)

trophozoite stage. Later they leave the epithelial cell and develop into more or less elongate motile adults, usually referred to as Gregarines. The vast majority of the GREGARI-NIDA do not show asexual reproduction or schizogony, and multiplication takes place solely by sporogony following upon gametogony. The adult Gregarines or sporonts are gametocytes, but do not show a differentiation into male or female gametocytes, which is a characteristic feature of the COCCIDIA and the HEMOSPORIDIA. The gametocytes associate

in pairs, and both produce an equal number of gametes, which are usually equal in size though not always in character, since those arising from one gametocyte may behave as male gametes and those from the other as female gametes. Conjugation thus takes place between similar or dissimilar gametes (*isogamy* or *anisogamy*). In a small group, however, schizogony takes place as well as sexual reproduction, and the order is divided into two suborders on this basis. Thus :

# I. Suborder EUGREGARINARIA Doflein, 1901.

This suborder includes the majority of the so-called Gregarines, which are usually parasites of Arthropods or Annelids. The sporocyst, after gaining entrance to a suitable host, germinates and sets free the sporozoites, which enter the epithelial cells of the digestive tract. There they grow large and protrude from the host-cells, remaining attached to them by an organ of attachment, called the epimerite, of varied form. These trophozoites sooner or later become detached from the cells of the host and move about in the lumen of the gut, where they are usually encountered as large and vermiform bodies exhibiting a gliding movement. In one group, the SEPTATA, the main body is divided into two parts, which are distinctly marked off from each other by an ectoplasmic septum. The smaller anterior part is known as the protomerite, and the larger posterior part the *deutomerite*. The latter contains a single nucleus. In the other group, the HAPLOCYTA, the body is not divided by a septum, and consists of a single chamber. In this latter group the sporozoites penetrate the wall of the gut and enter the body-cavity, forming cysts on the cœlomie side of the intestinal wall, or develop as free forms inside the seminal vesieles or other parts of the body-cavity.

The trophozoites increase in size and are then known as gametocytes; these then encyst in pairs. Within the cystmembrane each gametocyte gives rise to a large number of gametes, which may be isogamous or anisogamous. Each of the gametes formed from one gametocyte unites with one formed from the other, and a large number of zygotes are the result. Each zygote becomes surrounded by a resistant membrane (occyst or sporoeyst), and its contents produce usually eight sporozoites.

The suborder is divided as follows :---

[kester, p. 54.

Trophozoite single-chambered	Legion Haplocyta Lan-
Trophozoite divided by an ectoplasmic	[p. 89.
septum	Legion Septata Lankester,

# 1. Legion HAPLOCYTA Lankester, 1885

(= ACEPHALINA Kœlliker or MONOCYSTIDEA Stein+Lecudinidæ and other primarily Dicystid forms).

The EUGREGARINARIA are usually divided into ACEPHALINA and CEPHALINA on the basis of the absence or presence of an epimerite. Hesse (1909), Bhatia (1924), and Cognetti (1921, 1925, and 1926) have shown that a fairly large number of Monocystids also possess an organ of attachment or epimerite. An epimerite is also present in the family Lecudinidæ (=Doliocystidæ), but the body is not divided into a protomerite and a dentomerite. Brasil (1908 and 1909) discussed the relationship of Doliocystis with Lankesteria and other admittedly Monocystid forms. Minchin (1903) considered it purely a matter of definition whether Doliocystidæ should be considered as CEPHALINA without a septum, or as MONOCYSTIDEA with an epimerite. I have elsewhere (1924, p. 508) discussed the grounds for classifying the EUGREGARINARIA into HAPLOCYTA and SEPTATA, and for placing the Doliocystidæ under the former.

The legion HAPLOCYTA may be divided into two tribes, as follows :---

1.	Producing sporocysts with similar poles.	
	Generally coelomic parasites of Oligo-	[p. 54.
	chætes	Homopolarldea Bhatia,
2.	Producing sporocysts with dissimilar poles.	
	Intestinal or cœlomic parasites, usually of	[Bhatia, p. 82.
	marine animals	Heteropolaridea

# 1. Tribe HOMOPOLARIDEA Bhatia, 1930 (=Family HOMOPOLARIDÆ Dogiel).

Sporocysts with similar poles. Generally coelonic parasites of terrestrial Oligochætes. The tribe comprises eight families (*vide* Bhatia, 1930), of which representatives of six are at present known from India.

#### Identification Table of Families.

1	(12).	Sporocysts biconical, with similar, non-	
		appendiculate poles, octozoic	2.
2	(11).	Sporocysts not provided with spines at	
		either extremity	3.
3	(10).	Trophozoite simple	4.
4	(9).	Trophozoite solitary	5. [Bhatia, p. 55.
5	(6).	Trophozoite without any differentiation	Stein, emend.
		at the anterior end	Monocystidæ
6	(5).	Trophozoite with a differentiation at the	•
		anterior end	7.
7	(8).	Trophozoite with a conical or cylindro-	[Bhatia, p. 65.
		conical trunk at the anterior end	Rhynchocystidæ

MONOCYSTIS.

8	(7).	Trophozoite with a sucker-like differentia- tion at the anterior end	[Bhatia, p. 68. Stomatophoridæ
9	(4).	Adult trophozoites always associated in pairs	[Bhatia, p. 72. Zygocystidæ
10	(3).	Trophozoito branching at the anterior end	[Bhatia, p. 73. Aikinetocystidæ
11	(2).	Sporocysts provided with spines at either extremity	[Bhatia. Syncystidæ*
12	(1).	Sporocysts not biconical. Trophozoites with or without an epimerite	13.
13	(14).	Sporocysts oval or spherical. Gametes net differentiated	[Bhatia, p. 76. Dipiocystidæ
14	(13).	Sporocysts spherical. Gametes differenti- ated	[Poche. Schaudinellidæ*,

# 1. Family MONOCYSTIDÆ Stein, emend. Bhatia, 1930.

Trophozoites oval, spherical or elongated, without any differentiation at the anterior end, solitary. Sporocysts biconical, with similar, non-appendiculate poles, octozoic. The family includes four genera (*vide* Bhatia, 1930), of which three are known from India.

#### Key to Indian Genera.

1 (4). 2 (3).	Trophozoite ovoid or spherical Trophozoite ovoid	2. [p. 55. Monocystis Stein,
3 (2). 4 (1).	Trophozoite spherical Trophozoite olongated, cylindrical, like a Nematode worm	[p. 59. Apolocystis Cognetti, [p. 60. NEMATOCYSTIS Hesse.

#### Genus MONOCYSTIS Stein, 1848, emend. Hesse, 1909, and Cognetti, 1923.

Monocystis, Stein, 1848, pp. 183-223; Lankester, 1863, p. 93; 1866, pp. 23-8; 1872, pp. 342-51; Bütschli, 1880-2, p. 576; Ruschhaupt, 1885, pp. 1-38; Bosanquet, 1894, pp. 421-33; Labbé, 1899, p. 38; Cuénot, 1901, pp. 581-654; Cecconi, 1902 a, pp. 132-5; 1902 b, pp. 122-40; Minchin, 1903, pp. 154-66; Brasil, 1905 a, pp. 17-38; 1905 b, pp. 69-99; Hesse, 1909, p. 44; Hoffmann, 1909, pp. 139-66; Mulsow, 1911, pp. 20-55; Minchin, 1912, pp. 23, 174, 328, 331, 332, 333, 336, 339; Cognetti, 1923, pp. 250-3; Berlin, 1924, p. 14; Cognetti, 1925, p. 229; Wenyon, 1926, p. 1147; Calkins & Bowling, 1926, pp. 385-99; Knowles, 1928, p. 499; Reichenow, 1929, p. 886; Bhatia, 1929, p. 122; 1930, p. 158; Kudo, 1931, pp. 57, 74, 293; Calkins, 1933, pp. 99, 100, 309, 559.

Trophozoites ovoid, without marked differentiation anteriorly, often provided at the anterior pole with a mucron, solitary. Sporocysts as defined for the family.

<sup>\*</sup> placed after the name of a family indicates that no representative of the family has as yet been recorded from India, Burma or Ceylon.

#### SPOROZOA.

#### Key to Indian Species.

1 (3).	Nucleus spherical, with a large central	<u> </u>
2.	karyosome Trophozoites fusiform or variable in form, often with a clear cylindrical process at one end, $100-150 \mu$ by $30-40 \mu$ . Gameto- cysts spherical, $80-100 \mu$ in diameter	2. [p. 56. <i>M. beddardi</i> Ghosh,
3 (1).	Nucleus spherical or oval, with an irregu- lar or eccentric karvosomo	4.
4 (6).	Nucleus with a large irregular karyo- some, consisting of several masses	5.
5.	Trophozoites oval, or elongated and club-shaped, $40-80 \mu$ by $12-30 \mu$ . Gametocysts irregularly hemispherical, $70-80 \mu$ in diameter.	[p. 57. M. bengalensis Ghosh,
6 (4).	Nucleus spherical or oval, with an eccentric karvosome	7.
7 (8).	Trophozoites rounded or oval when young, fusiforin when mature, $100 \mu$ in length. Gametocysts sphorical or oval $44 \mu$ is diameter	[p. 58. M. Hoidi Ghosh
8 (7).	Trophozoites variable in form, attaining a size up to $220 \mu$ by $50 \mu$ . Gameto- cysts spherical, $80 \mu$ in diameter	[p. 58. M. pheretimi Bh. & Ch.,

1. Monocystis beddardi Ghosh. (Fig. 4.)

†Monocystis beddardi, Ghosh, 1923, pp. 425-7, figs. 9-13. Monocystis beddardi, Bhatia, 1929, p. 123.

Trophozoites elongately fusiform when young, fusiform or variable in form when mature. Body often with a clear



Fig. 4.—Monocystis beddardi Ghosh. (After Ghosh.)

cylindrical process at one end. Surface smooth. Ectoplasm very thin. Endoplasm highly granular, with paramylon grains. Change of shape involves the formation of a bulbous swelling, which occupies a large portion of the body, with

<sup>†</sup> prefixed to a reference indicates that the record of the species from India, Burma or Ceylon is based on it.

rapid in-pouring of the paramylon grains into it, and the swelling gradually disappears, to be formed again; the parasite now assumes a rounded shape, with or without a narrow process at one or both ends. Nucleus spherical, with a large central karyosome. Gametocyst spherical, with hemispherical or irregularly oval gametocytes. Sporocysts typical.

Dimensions.—Trophozoite  $100-150 \mu$  in length and  $30-40 \mu$  in width; gametocysts  $80-100 \mu$  in diameter; sporocysts  $12 \cdot 5 \mu$  in length.

Remarks.—Dr. K. N. Bahl sent me some preparations of the contents of the seminal vesicles of *Eutyphæus* sp. In these are a large number of trophozoites and other stages of a Monocystid referable to M. beddardi Ghosh. There are, in addition, numerous specimens referable to Dirhynchocystis globosa (Bhatia & Chatterjee). Ghosh in his description of M. beddardi describes the parasite as assuming a rounded shape, with or without a narrow process at one or both ends. It is possible that he found specimens of D. globosa occurring along with M. beddardi and considered them merely as contracted stages of the latter. But the processes in Dirhynchocystis are clearly marked off from the body, and not as represented by Ghosh in his figs. 12 and 13.

Habitat.—Seminal vesicles of Eutyphœus nicholsoni (Beddard): BENGAL, Calcutta; Eutyphœus sp.: UNITED PROVINCES, Lucknow.

#### 2. Monocystis bengalensis Ghosh. (Fig. 5.)

†Monocystis bengalensis, Ghosh, 1923, pp. 423-5, figs. 1-7. Monocystis bengalensis, Bhatia, 1929, p. 123.

Trophozoites oval when young, later elongated and clubshaped. Anterior end wide and rounded. Posterior end



Fig. 5.—Monocystis bengalensis Ghosh. A, trophozoite; B, cyst with two sporonts. (After Ghosh.)

narrow and rounded. Surface smooth. Ectoplasm very thin. Myonemes barely visible in stained specimens. Endoplasm highly granular, with large irregular-shaped paramylon grains. Nucleus rounded, with a large irregular karyosome, consisting of several compact masses. Movement slow and peristaltic. Gametocytes rounded or oval. Gametocysts irregularly hemispherical, apposed to each other by their flat surfaces. Sporocysts and sporozoites typical.

Dimensions.—Trophozoite  $40-80\,\mu$  in length,  $12-30\,\mu$  in width; gametocysts  $70-80\,\mu$  in diameter.

Habitat.—Seminal vesicles of Pheretima posthuma (L. Vaill.): BENGAL, Calcutta.

#### 3. Monocystis lloidi Ghosh.

Monocystis lloidi, Ghosh, 1923, p. 425. Monocystis lloidi, Bhatia, 1929, p. 123.

Trophozoites rounded or oval when young; somewhat fusiform, about one-third longer than broad, when mature. Anterior end more rounded than the posterior. Surface smooth. Ectoplasm distinct and comparatively thick. Endoplasm finely granular, with scattered, coarse granules. No distinct paramylon grains. Nucleus with a small eccentric karyosome. Movement brisk and peristaltic. Gametocysts spherical or oval. Sporocysts and sporozoites typical.

Dimensions.—Trophozoite  $100 \mu$  in length; gametocysts  $84 \mu$  in diameter.

Habitat.—Seminal vesicles of Pheretima posthuma (L. Vaill.): BENGAL, Calcutta. Rare.

### 4. Monocystis pheretimi Bhatia & Chatterjee. (Fig. 6.)

\*Monocystis pheretimi, Bhatia & Chatterjee, 1925, pp. 199-200, figs. 20, 21, 28, 29.

Monocystis pheretimi Bhatia, 1929, p. 123.

Trophozoites variable in form, spherical, ovoid, ellipsoidal



Fig. 6.-Monocystis pheretimi Bh. & Ch. (After Bhatia and Chatterjee.)

#### APOLOCYSTIS.

or dumbbell-shaped, showing active movements. Paraglycogen grains few. Epicyte thin, without striations. Sareocyte well developed. Myocyte fibrils delicate. Endoplasm excavated by vacuoles. Nucleus generally spherical, sometimes ovoidal, with a single spherical karyosome, placed eecentrically. Gametoeysts spherical. Sporocysts and sporozoites typical.

Dimensions.—Trophozoites attain a size up to  $220\mu$  by  $50\mu$ ; gametocysts  $80\mu$  in diameter.

Habitat.—Seminal vesicles of Pheretima posthuma (L. Vaill.): PUNJAB, Lahore; BOMBAY, Bombay.

#### Genus APOLOCYSTIS Cognetti, 1923.

Apolocystis, Cognetti, 1923, p. 253; 1925, p. 231; Reichenow, 1929, p. 887; Bhatia, 1929, p. 124; 1930, p. 158; Troisi, 1933, pp. 336-42; Calkins, 1933, p. 559.

Trophozoites spherical, without any marked principal axis, and without polar differentiation, solitary. Sporocysts as in the family.

#### 5. Apolocystis matthaii (Bhatia & Setna). (Fig. 7.)

†Monocystis matthaii, Bhatia & Setna, 1926, pp. 362-4, figs. 1-4, 25-27. Apolocystis matthaii, Bhatia, 1929, p. 124.

Trophozoites most commonly spherical, sometimes ovoid or kidney-shaped, not covered by hairs. Epicyte thin and not showing any meridional striations. Sarcocyte hyaline and little developed. Myocyte fibres not distinguishable. Endoplasm very dark, being finely granular, the alveoli



Fig. 7.-Apolocystis matthaii (Bh. & Set.). (After Bhat a and Setna.)

compactly arranged and with large grains of reserve material. Nucleus spherical, with single spherical karyosome, placed eccentrically. Gametoeysts ellipsoidal.

Dimensions.—Trophozoites attain a size up to  $238 \mu$ ; gametocysts up to  $468 \mu$  in their longer diameter.

#### SPOROZOA.

*Remarks.*—Instances of solitary encystment and, more rarely, cysts containing three individuals were encountered. I have emended the spelling of the specific name to make it conform correctly to that of the person to whom the species is dedicated.

Habitat.—Seminal vesicles of Megascolex trilobatus (Steph.): BOMBAY, Bombay.

#### Genus NEMATOCYSTIS Hesse, 1909.

Nematocystis, Hesse, 1909, p. 45; Cognetti, 1918, p. 149; 1921 a. pp. 153-4; 1921 b, pp. 552-4; Berlin, 1924, pp. 67-8, 74, 83-7; Cognetti, 1925, p. 231; Bhatia & Chatterjee, 1925, pp. 193-7; Bhatia & Setna, 1926, pp. 364-7; Reichenow, 1929, p. 887; Bhatia, 1929, pp. 124-5; 1930, p. 158; Calkins, 1933, p. 559.

Trophozoites elongated, cylindrical, shaped like a Nematode worm, attaining even up to 5 mm. in length. Solitary. Sporocysts as in the family.

#### Key to Indian Species.

1 (6).	Trophozoite tapering at one or both ends.	2.
2 (3)	Trophozoite tapering at either end.	
	Nucleus long, fusiform, with two	[p. 60.
	karyosomes	N. hessei Bh. & Ch.,
3 (2).	Trophozoite with anterior end rounded,	
	posterior end pointed	4.
(5).	Nucleus long, fusiform, with single karyo-	[p. 62.
	some	N. lumbricoides Hesse,
5 (4).	Nucleus elongated oval, with several	[Bh. & Ch., p. 62.
	karyosomes	N. plurikaryosomata
6 (1).	Trophozoite usually rounded at both	
	ends	7.
7 (8).	Anterior pole without epimeritic denti-	
	culations, posterior pole not covered	
	by fine hairs. Nucleus usually in the	
	middle of the body, with single karyo-	[Set., p. 63.
	some	N. stephensoni Bh. &
8 (7).	Anterior pole with epimeritic denticula-	-
• •	tions, posterior pole covered by fine	
	hairs. Nucleus near one end of the	[p. 64.
	body, with single karyosome	N. vermicularis Hesse.
	·· · ·	

#### 6. Nematocystis hessei Bhatia & Chatterjee. (Fig. 8.)

Nematocystis hessei, Bhatia & Chatterjee, 1925, pp. 194-5, pl. viii, figs. 11, 11 a, 12; pl. ix, fig. 24.
 Nematocystis hessei, Cognetti, 1925, p. 231; Bhatia, 1929, p. 125.

Trophozoite elongated like a worm and tapering at either end. No epicytal striations. Nucleus long and fusiform, with two karyosomes, which are sometimes very unequal in size.

Dimensions.—Up to  $552\mu$  in length by  $42\mu$  in thickness.

Remarks.—The species in some respects resembles N. anguillula, but differs from it in the absence of epicytal striations and in the nuclear structure. The nucleus is situated near the centre of the parasite and is long and fusiform; it may attain a length of  $79\mu$ . The two karyosomes may be equal



- Fig. 8.—Nematocystis hessei Bh. & Ch. (After Bhatia and Chatterjee.) Fig. 9.—Nematocystis lumbricoides Hesse. (After Bhatia and Chatter-
- jee.) Fig. 10.—Nematocystis plurikaryosomata Bh. & Ch. (After Bhatia and
- Fig. 10.—Nematocystis plurikaryosomata Bh. & Ch. (After Bhatia and Chatterjee.)

or very unequal in size, and are situated near the two ends of the nucleus.

Habitat.—Sominal vesicles of Pheretima heterochæta (Mchlsn.): PUNJAB, Lahore.

#### 7. Nematocystis lumbricoides Hesse. (Fig. 9.)

Nematocystis lumbricoides, Hesse, 1909, pp. 153-5, figs. lxv, lxvi.

- \*Nematocystis lumbricoides, Bhatia & Chatterjee, 1925, p. 196, pl. viii, fig. 17.
  - Nematocystis lumbricoides, Cognetti, 1925, p. 231; Bhatia, 1929, p. 124.

Trophozoite shaped like an earthworm, swollen in the middle and narrower at each end, anterior end somewhat rounded and posterior end more pointed. Nucleus long and fusiform, with a single karyosome.

Dimensions.—Up to 1.5 mm. in length and  $60\mu$  in thickness. Remarks.—Specimens examined by Bhatia and Chatterjee from Pheretima heterochæta (Mchlsn.) at Lahore differed from those described by Hesse from Helodrilus caliginosus Savig. in France in that the size did not exceed 1 mm. by  $31\mu$ , the epicyte was not ornamented with parallel longitudinal striations, and the nucleus was variable in position and contained a single large oval karyosome placed eccentrically.

Habitat.—Seminal vesicles of Pheretima heterochæta (Mchlsn.): PUNJAB, Lahore.

#### 8. Nematocystis plurikaryosomata Bhatia & Chatterjee. (Fig. 10.)

†Nematocystis plurikaryosomata, Bhatia & Chatterjee, 1925, pp. 195-6, pl. viii, figs. 13-16; pl. ix, fig. 25.

Nematocystis plurikaryosomata, Cognetti, 1925, p. 231; Bhatia, 1929, p. 125.

Trophozoite with a long and extremely deformable body. No epicytal striations. Nucleus elongate and oval, with several small karyosomes.

Dimensions.—Up to more than 1 mm. in length and  $100\mu$  in thickness. Gametocysts small, attaining a size of  $140\mu$  in diameter. Spores  $6.5-8.5\mu$  by  $3.4\mu$  in size.

Remarks.—The body of the parasite is very deformable and shows constrictions and bulgings during the progression of the parasite, the granular cytoplasm, together with the nucleus, appearing to flow from one pole to the other. The parasite differs from N. magna in that it lives free in the seminal vesicles, has no polar ornamentations, no hairs round its posterior pole, and in its nuclear structure. In stained preparations the paraglycogen grains are seen to be aggregated along the central region, leaving a clear peripheral zone. The nucleus is variable in position and contains 6 to 12 large, spherical karyosomes; it measures  $60\mu$  along its long axis.

Habitat.—Seminal vesicles of Allolobophora (Eisenia) fætida (Savig.): PUNJAB, Kasauli.

#### 9. Nematocystis stephensoni Bhatia & Setna. (Fig. 11.)

\*Nematocystis stephensoni, Bhatia & Setna, 1926, pp. 364-7, pls. xi, xii, figs. 5-17; pl. xiii, figs. 28, 29. Nematocystis stephensoni, Bhatia, 1929, p. 125.

Trophozoite of an elongate cylindrical form, short and swollen or elongated and thin, with blunt extremities. Body highly deformable, presenting a number of swellings and constrictions. Epicyte shows fine longitudinal striations.



Fig. 11.—Nematocystis stephensoni Bh. & Set. (After Bhatia and Setna.)

Nucleus elliptical, with a single central karyosome, which consists of a central doeply staining portion surrounded by a vacuolated layer. Gametocysts oval. Gametocytes equal in size; gametes similar.

Dimensions.—Trophozoite up to 1.26 mm. in length and 100  $\mu$  in thickness; gametocysts from 153  $\mu$  by 100  $\mu$  to 314  $\mu$  by 246  $\mu$ ; zygotes 8  $\mu$ ; sporocysts 15.5  $\mu$  by 8.1  $\mu$ .

Remarks.—In the living condition the parasite is quite opaque and capable of contracting and elongating so rapidly as to produce constrictions and bulgings, giving it the same appearance as N. magna and N. plurikaryosomata. The opacity of the granular cytoplasm is so great that the nucleus is only faintly indicated.

In the stained preparations the cytoplasm exhibits the usual structure. The nucleus is generally situated about the middle of the body, but is often found nearer one or the other extremity; this is due to the movements of the endoplasm. Its elongated axis generally lies parallel to the long axis of the body. The shape and structure of the nucleus are very characteristic. The nucleus is of an elongate, regularly ellipsoid form, and consists of (i) a well-defined nuclear membrane, (ii) a broad zone of peripheral achromatin, in which the network is not distinguishable, and over which fine chromatin grains are dispersed, and (iii) a large central, spherical or oval karyosome, which is like an entire nucleus in structure and appears very much like a nucleus within a nucleus. The nucleus thus appears to consist of two concentric rings and a deeply staining homogeneous mass in the centre. The inner ring and the central homogeneous mass together constitute the karvosome.

Development is on the usual lines, and different stages have been described by Bhatia and Setna (1926).

Habitat.—Seminal vesicles of Eutyphœus incommodus (Beddard): PUNJAB, Kasauli.

#### 10. Nematocystis vermicularis Hesse. (Fig. 12.)

Nematocystis vermicularis, Hesse, 1909, pp. 155-8, figs. lxvii-lxx Berlin, 1924, pp. 68, 74.

\*Nematocystis vermicularis, Bhatia & Chatterjee, 1925, p. 197. Nematocystis vermicularis, Bhatia, 1929, p. 124.

Trophozoite with a fusiform, rather thick body, rounded at both extremities. The anterior pole ornamented with a cap formed of small cylindrical prolongations placed side by side, posterior pole covered by fine hairs directed backwards. Ectoplasm thin, cuticular ornamentations not visible. Endoplasm rather rich in chromatoid granulations. Nucleus ellipsoidal, situated near one of the extremities of the body, and containing a single karyosome.

Dimensions.—Scarcely 1 mm. in length and  $100 \mu$  in thickness. Remarks.—The single specimen obtained by Bhatia and Chatterjee from the seminal vesicles of *Pheretima barbadensis* (Beddard) closely resembled Hesse's fig. lxviii a of the species as described from *Helodrilus longus* Ude. The size of the specimen was, however, much less, being only  $446 \mu$  in length by  $69 \mu$  in thickness. The nucleus was situated more towards the blunt end of the parasite and measured  $38.5\mu$  along its long axis. There was a central spherical karyosome.



Fig. 12.—Nematocystis vermicularis Hesse. (After Hosse.)

Habitat.—Seminal vesicles of Pheretima barbadensis (Beddard): PUNJAB, Lahore.

# 2. Family RHYNCHOCYSTIDÆ Bhatia, 1930.

Trophozoites ovoid, spherical or clongated, with a conical or cylindro-conical trunk at the anterior end, solitary. Sporocysts biconical, with similar non-appendiculate poles, octozoic.

#### Genus RHYNCHOCYSTIS Hesse, 1909.

Rhynchocystis, Hesse, 1909, p. 45; Cognetti, 1911, p. 207; Berlin, 1924, p. 68; Cognetti, 1925, p. 232; Bhatia & Chatterjee, 1925, p. 190; Bhatia & Setna, 1926, p. 370; Reichenow, 1929, p. 887; Bhatia, 1929, p. 129; 1930, p. 158; Calkins, 1933, p. 559; Troisi, 1933, pp. 327-34.

Trophozoites ovoid or cylindroid. Anterior pole provided with a metabolic epimerite, which is most frequently elongated into a conical or cylindro-conical trunk. Sporocysts with characters of the family.

#### Key to Indian Species.

1 (3).	Hairs present on both the mucron and the posterior end of the body	2.
2.	Form variable, pear-shaped, spherical	
	or gregariniform. Epimerite meta-	
	bolic, a conical or hemispherical mu-	
	cron. Nucleus rounded, variable in	[p. 67.
	position	R. cognettii Bh. & Ch.,
3 (1).	Hairs not present on any part of the	
• •	body	4.
4 (5).	Form elongate, pear-shaped, anterior	
. ,	end broader, with a nipple-shaped epi-	
	merite. Nucleus oval, usually in	[Set., p. 67.
	posterior half of the body	R. mamillata Bh. &

F

5 (4).	Form elongate, cylindrical, with cylin-	
	dro-conical epimerite marked with	
	distinct epicytal striations. Nucleus	
	oval, usually about the middle of the	[p. 66.
	body	R. awatii Bh. & Set.,

#### 11. Rhynchocystis awatii Bhatia & Setna. (Fig. 13.)

*†Rhynchocystis awatii*, Bhatia & Setna, 1926, pp. 371-3, pl. xii, figs. 23, 24; pl. xv, figs. 34, 35.
 *Rhynchocystis awatii*, Bhatia, 1929, p. 125.

Trophozoite with an elongate cylindrical body, the anterior end generally provided with a cylindro-conical epimerite. Epicyte ornamented with fine longitudinal striations, which are more distinct and spaced out over the epimeritic region. Sarcocyte poorly developed and free from granules. Nucleus oval, with a large eccentric karyosome, and generally placed about the middle of the body.

Fig. 13.



Fig. 13.—Rhynchocystis awatii Bh. & Set. (After Bhatia and Setna.) Fig. 14.—Rhynchocystis cognettii Bh. & Ch. (After Bhatia and Chatterjee.)

Fig. 15.—Rhynchocystis mamillata Bh. & Set. (After Bhatia and Setna.)

66

Dimensions.—Maximum length  $400 \mu$ ; maximum width, in shorter and thicker individuals,  $50 \mu$ .

Remarks .-- This appears to be a rare parasite of its host, being found only in a few specimens and in extremely small numbers. The form of the trophozoite varies considerably ; some are short and thick, while others are long and vermiform. An epimerite is not found in all individuals, but, when present, it is a distinct club-shaped structure, dilated at its distal extremity and narrower at its base. The ectocyte is thin. The entocyte is finely granular. The nucleus lies more often in the middle, though in some cases it is found to be situated near the anterior end. It is oval in form, with its long axis lying along the length of the parasite. The nucleus usually measures  $17.5\mu$  in its longer and  $10.5\mu$  in its shorter diameter, and contains a single large eccentrically placed karvosome. Some individuals show end to end syzygy ; sometimes the satellite retains its epimerite and is fixed by it to the posterior end of the primite.

Habitat.—Seminal vesicles of Pheretima elongata (E. Perr.): BOMBAY, Bombay.

12. Rhynchocystis cognettii Bhatia & Chatterjee. (Fig. 14).

†Rhynchocystis cognettii, Bhatia & Chatterjee, 1925, pp. 190-3, pl. vii, figs. 1-10; pl. ix, figs. 22, 23.
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Rhynchocystis cognettii, Bhatia, 1929, p. 125.

Trophozoite with an epimerite. Shape of the body variable, pear-shaped, spherical or gregariniform, and the anterior end provided with a mucron surrounded by a crown of sarcoeyte. Hairs found only in the region of the mucron and sometimes at the posterior end also. Nucleus generally spherical, with a single spherical karyosome placed eccentrically and surrounded by a clear white halo. The nucleus varies in its position, but is never situated in the epimeritic region. Gametocytes are nearly equal in size.

Dimensions.—Maximum size of the trophozoite  $129\mu$  by  $46 \mu$ ; gametocysts are more or less ovoid and measure up to  $129\mu$  by  $81\mu$ .

Habitat.—Seminal vesicles of Allolobophora caliginosa (Savig.): PUNJAB, Kasauli.

#### 13. Rhynchocystis mamillata Bhatia & Setna. (Fig. 15.)

†Rhynchocystis mamillata, Bhatia & Setna, 1926, pp. 370-1, pl. xii, figs. 21, 22; pl. xiv, figs. 32, 33. Rhynchocystis mamillata, Bhatia, 1929, p. 125.

Trophozoite with an elongate pear-shaped body, the anterior end broader and provided with an epimerite consisting of a nipple-shaped mucron surrounded at its base by a ring in which the sarcocyte is well developed. Movements of the trophozoite very slow. Body not covered with hair. Nucleus oval and situated in the posterior half of the body.

Dimensions.—The trophozoite may attain a maximum length of  $126\,\mu$ , maximum width  $52\cdot5\,\mu$ ; nucleus  $14-15\cdot9\,\mu$  in length and  $8-11\cdot3\,\mu$  in width. Cysts and spores not identified.

Remarks.—R. mamillata differs from R. cognettii in being more constant in form and in not exhibiting the changes shown by the latter species. It differs from R. pilosa by its constant form and by the complete absence of hair covering the body. The relative dimensions of the trophozoite, nucleus, and karyosome differ markedly from R. awatii, which is found in the same host-species, the nucleus being larger in trophozoites of corresponding size.

The sperms of the host, sticking close together to the epimerite, form a thick investment round it. The ectoplasm is not thick, but the epicyte is fairly well developed. The endoplasm, with the contained granules, is moved about in the interior of the body from pole to pole, without affecting the external form of the body. The nucleus generally lies in the posterior half of the body, rarely in the middle or the anterior half; it is distinctly oval, and contains a single spherical or slightly oval karyosome, which is placed eccentrically.

Habitat.—Seminal vesicles of Pheretima elongata (E. Parr.): BOMBAY, Bombay.

# 3. Family STOMATOPHORIDÆ Bhatia, 1930.

Trophozoites ovoid, spherical, cylindrical or cup-shaped, with an anterior sucker-like epimeritic organ; solitary. Sporocysts navicular, with similar non-appendiculate and truncated poles; octozoic.

The family includes seven genera (vide Bhatia, 1930), of which only one is at present known from India. All the genera are parasitic in various species of *Pheretima* only.

# Genus STOMATOPHORA Drzewecki, 1907, emend. Hesse, 1909, and Bhatia, 1924.

Stomatophora, Drzewecki, 1907, pp. 218-46; Hesse, 1909, pp. 45, 161-87; Bhatia, 1924, pp. 481-512, pl. xxiii, figs. 1-31; Bhatia & Setna, 1926, pp. 367-8; Reichenow, 1929, p. 887; Calkins, 1933, p. 560; Ray & Chatterjee, 1936, p. 345.

Trophozoites ovoid or spherical. Anterior end provided with a sucker-like epimeritic organ with or without a central mucron. Sporocysts typical of the family.

#### Key to Indian Species.

1 (4).	Form with elongate cylindro-conical	0
2 (3).	Sucker an inverted balloon-shaped de- pression with a central mucron and	2. [p. 69.
3 (2).	radial epicytal striations Sucker cup-shaped, with a central mucron surrounded by a crown of	S. bulbifera Bh. & Set., [p. 69.
4 (1).	Form plate- or disc-like, marked by furrows and divided into a number of	S. coronata (Hesso),
	like, with a central mucron	S. diadema Hosse, p. 71.

#### 14. Stomatophora bulbifera Bhatia & Setna. (Fig. 16.)

†Stomatophora bulbifera, Bhatia & Setna, 1926, pp. 368-70, pl. xii, figs. 18-20, ; pl. xiv, figs. 30, 31. Stomatophora bulbifera, Bhatia, 1929, p. 125.

Trophozoite with an elongate cylindro-conical body, a broad and round anterior end and a narrow and pointed posterior end. Body not marked by any furrows or epicytal striations. The sucker at the anterior end is an inverted balloon-shaped depression with a central mucron and radial epicytal striations. Nucleus, oval with a large spherical karyosome.

Dimensions.—Length of trophozoite  $56-119\mu$ , width  $24\cdot 5-52\cdot 5\mu$ ; nucleus  $10\cdot 5-15\cdot 7\mu$  by  $7-11\cdot 3\mu$ .

*Remarks.*—The greatest width of the organism is at onc-third the length of the body from the anterior end. The sucker is like an inverted balloon in form, with the epicyte lining it and showing striations diverging from a deeply stained central mass, which corresponds to the mucron but is not raised. Over the body the epicyte is thin, and the sarcocyte forms a thick hyaline and transparent layer. The endoplasm is alveolar, and numerous paramylon grains are found adhering to the walls of the alveoli. Nucleus with its long axis parallel to the long axis of the body.

Habitat.—Seminal vesicles of Pheretima elongata (E. Perr.) BOMBAY, Bombay.

#### 15. Stomatophora coronata (Hesse). (Fig. 17.)

Monocystis coronatus, Hesse, 1904, p. 268.

Stomatophora coronata Drzewecki, 1907, pp. 218-46; Hesse, 1909, pp. 161-87, figs. lxxiv-lxxix; pl. ii, figs. 40-73; pl. vi, figs. 136-70; pl. vii, figs. 187-93; Sokolow, 1911, p. 292; Minchin, 1912, p. 328.

† Stomatophora coronata, Bhatia, 1924, pp. 499-507, pl. xxiii, figs. 20-5.
 Stomatophora coronata, Cognetti, 1925, p. 233; Bhatia, 1929, p. 125; Reichenow, 1929, p. 887.

Trophozoite with an oval or ellipsoidal body, rounded at

both ends. Anterior end bears a sucker, formed as a spherical cavity with a cap cut off from the upper pole, and with a central mucron arising from the proximal pole as a projection into the cavity; the sucker is surrounded by a crown of a variable number of petals formed of hyaline sarcocyte covered over



Fig. 16.—Stomatophora bulbifera Bh. & Set. (After Bhatia and Setna.)
Fig. 17.—Stomatophora coronata (Hesse). (After Bhatia.)
Fig. 18.—Stomatophora diadema Hesse. (After Bhatia.)

by epicyte marked by striations. Gametocysts generally spherical or slightly ellipsoidal. Sporocysts navicular, but their ends are drawn out and truncated, presenting a buttonlike flattening, sometimes sticking end to end to form chains. Dimensions.—Maximum size of trophozoite  $180 \mu$  by  $130 \mu$ , average  $80 \mu$  by  $60 \mu$ ; gametoeysts measure  $60-70 \mu$  by  $50-60 \mu$ ; sporoeysts  $9-11 \mu$  by  $5-6 \mu$  and  $7-7.5 \mu$  by  $3 \mu$ .

Remarks.—The organism exhibits active though by no means rapid movements of the body, thus causing alterations in form from moment to moment. The body is generally expanded at the anterior end so as to look like a top or a flowervase. It may then show a constriction round the middle of the body and assume an hour-glass-like shape, and later a more regular outline again. In 1924 I studied the structure of the sucker and the crown of petals, and amended the diagnosis of the species as given by Hesse (1909).

There is generally a small difference in size between the gametocytes enclosed in a cyst, but the gametes do not show anisogamy.

Habitat.—Seminal vesicles of Pheretima barbadensis (Beddard): PUNJAB, Lahore.

#### 16. Stomatophora diadema Hesse. (Fig. 18.)

Stomatophora diadema, Hesse, 1909, pp. 187-9, pl. ii, figs. 66-8. †Stomatophora diadema, Bhatia, 1924, pp. 503-7, pl. xxiii, figs. 26-31. Stomatophora diadema, Cognetti, 1925, p. 233; Bhatia, 1929, p. 125. †Stomatophora sp., Ray & Chatterjee, 1936, p. 345.

Trophozoite with the form of a sphere flattened and compressed between its two poles, so as to resemble a plate or disc marked by furrows and divided into a number of irregular lobes, some of which are considerably larger than others. The sucker is a shallow-like cup depression in the centre of the body, and shows a central conical projection or mucron. Nucleus generally rounded, sometimes oval, eccentric in position, with a large karyosome. Gametocysts usually spherical. Sporocysts navicular, with extremities rather drawn out and truncated at the ends.

Dimensions.—Maximum size of trophozoite  $105 \mu$ ; gametocysts  $125-170 \mu$  in diameter; sporocysts  $8-12 \mu$  by  $3-4 \mu$ .

Remarks.—In 1924 I discussed the morphology of the sucker in S. simplex, S. coronata, and S. diadema, and came to the conclusion that, unlike the epimerite of the Polycystid Gregarines, the sucker is not present in the earlier stages of development in any of the species of Stomatophora, and that during the growth of the trophozoite a simple cup-like sucker is formed by a flattening and inpushing of the anterior end of the body, the median projection or mucron being thus carried to the bottom of the eup-like depression. This process would seem to cause the epicytal striations to appear first on the surface of the sucker and extending from the central mucron to certain definite points on the circular border of the aperture;

later they extend beyond over smaller or larger portions of the body. In S. simplex these striæ do not extend beyond the region of the sucker; in S. coronata they extend over a small area of the body immediately surrounding the sucker, thus forming a crown of petals; and lastly, in S. diadema they extend not only over the whole of the body, but some of them deepen and involve the deeper layers of the ectoplasm, causing the body to be cleft into lobes. Ray and Chatterjee (1936) have also studied the structure of the suckers of Stomatophora, but their detailed observations have not yet been published.

Habitat.—Seminal vesicles of Pheretima barbadensis (Beddard): PUNJAB, Lahore; seminal vesicles of Pheretima posthuma (L. Vaill.): BENGAL, Calcutta.

# 4. Family ZYGOCYSTIDÆ Bhatia, 1930.

Adult trophozoites always associated in pairs or groups of three. Sporocysts biconical, octozoic.

#### Genus EXTREMOCYSTIS Setna, 1931.

Extremocystis, Setna, 1931, pp. 206-9, pl. vi, figs. 2-6.

Adult trophozoites elongate, resembling *Nematocystis*; always associated in pairs, attachment being end to end.

#### 17. Extremocystis dendrostomi Setna. (Fig. 19.)

*Extremocystis dendrostomi*, Setna, 1931, pp. 206-9, pl. vi, figs. 2-6; p. 325.

The only known species in the genus.

Adult trophozoites have the characters given above; attachment is brought about by one end of an individual



Fig. 19.—Extremocystis dendrostomi Setna. (After Setna.)

fitting into a concave depression of the other. Nucleus ellipsoidal, greatly elongated, generally lying near that end of the trophozoite which joins with the other trophozoite. Dimensions.—Maximum size 130  $\mu$  by 19  $\mu$ , minimum 70  $\mu$  by 12 $\mu$ .

Remarks.--The parasites lie coiled and contorted among the cœlomic corpuscles and the genital products of the host, and are never attached to the body-wall or the gut. There is no organ of attachment to the host. The body is cylindrical, resembling an elongate Nematode worm, with more or less parallel sides and tapering slightly towards the extremities of the united pair. Attachment takes place by means of the tapering end of an individual fitting into a regular concave hemispherical depression of the other. The ratio of width to length of each individual is about 1:6. Ectoplasm is thin. Endoplasm is coarsely granular, and the stream of granules flows very swiftly and uniformly from one end of the trophozoite to the other, going backwards and forwards in a straight line. The nucleus measures  $17 \mu$  by  $6 \mu$ , and lies with its long axis parallel or slightly inclined to the sides of the body.

The associating pairs become short and pear-shaped and later rounded off. Gametocysts not found. Sporocysts spindle-shaped, with the two ends finely pointed,  $29-35 \mu$  by  $6\cdot 4 \mu$ .

Habitat.—Cœlomic cavity of Dendrostoma signifer Sel. & de Man : ANDAMANS, Port Blair.

# 5. Family AIKINETOCYSTIDÆ Bhatia, 1930.

Trophozoites solitary or adherent, branching dichotomously, the branches serving for the attachment of the parasite to the host. Sporocysts as in *Monocystis*.

#### Genus AIKINETOCYSTIS Gates, 1926.

Aikinetocystis, Gates, 1926, pp. 400-4; Bhatia, 1930, p. 160.

Trophozoites cylindrical or columnar, with a characteristic regular, dichotomous branching at the attached end, with sucker-like bodies borne on the ultimate branches; solitary or in groups of three to eight. Sporocysts as in *Monocystis*. Cœlomic parasites in various species of *Eutyphœus*.

#### 18. Aikinetocystis singularis Gates. (Fig. 20.)

†Aikinetocystis singularis, Gates, 1926, pp. 400-4, figs. 1-3. Aikinetocystis singularis, Bhatia, 1929, p. 126; 1930, p. 160.

Trophozoite cylindrical or columnar, with a characteristic regular, dichotomous branching at the attached end; fixation

#### SPOROZOA.

to the host by means of sucker-like bodies borne on the ultimate branches. Ectoplasm shows longitudinal and transverse striations. The transparent fluid endoplasm is densely packed with ovoid paraglycogen granules. Nucleus is a large ovoid body containing a spherical eccentrically situated karyosome.

Dimensions.—Trophozoite may be as long as 4 mm., nucleus 640–860  $\mu$  in length; gametocysts about 620  $\mu$  in length; sporocysts of two sizes, the larger 20–23  $\mu$  long, the smaller 7–9  $\mu$  long.

*Remarks.*—The parasite presents a smooth creamy-white appearance or, very rarely, a light brownish tinge. The large unattached end is bluntly rounded. At a greater or less distance from this end the body branches into two rami,



Fig. 20.—Aikinetocystis singularis Gates. n, nucleus; K, karyosome; Pr, primary ramus; Sr, secondary ramus. (After Gates.)

each of which divides to form two smaller secondary rami, This branching continues until eight or sixteen small ramuli are produced. The dichotomy is regular. The ramuli bear groups of irregularly ovoid, sucker-like objects by means of which the parasite is attached to the host. Pairs of animals are frequently found attached to each other near the rounded free ends. Groups of three to eight animals, similarly adherent to each other, are also found.

Living detached specimens show two kinds of movements. Waves of peristaltic contraction pass along the trunk and rami of the first order, producing a churning of the endoplasmic contents in the trunk, from the trunk into the primary rami, and back into the trunk; the nucleus is squeezed to and fro from one end of the trunk to the other, or even passing into one of the primary rami and returning into the trunk. The second kind, described as rotation movements, result in a spiral twisting of the primary rami and their branches on their own axes.

*Monocystis*-like sporocysts, in masses visible to the naked eye, are present in the anterior segment of the host or can be found by scraping the body-wall. Ovoid gametocysts were found in a few hosts.

Habitat.—Cœlomic cavities of Eutyphœus foveatus (Rosa) (most commonly) and E. spinulosus Gates, E. rarus Gates, and E. peguanus Gates (more rarely): BURMA.

#### Genus NELLOCYSTIS Gates, 1933.

Nellocystis, Gates, 1933, pp. 508-11.

Trophozoites solitary or in aggregates of two or three individuals. Anterior end stalk-like and branching into several short filaments which attach the parasite to the host. Sporocysts as in *Monocystis*.

#### 19. Nellocystis birmanica Gates. (Fig. 21.)

Nellocystis birmanica, Gates, 1933, pp. 508-11, figs. 1-8.

Trophozoites club-shaped, the posterior end enlarged, the anterior end narrowing to an elongate stalk. The anterior end of the stalk branches into several short filaments, which attach the parasite to the host. Within the posterior portion is a vacuolar cavity containing a fluid in which there is a single dark, regularly spheroidal, granular mass containing a single ovoidal nucleus with a single karyosome. Within the stalk, near the vacuolar cavity, is a second nucleus, which contains, in addition to a large karyosome, a smaller, slightly bent, rod-like body. This stalk-nucleus is placed with its long axis along the long axis of the stalk. In some specimens the stalknucleus is transverse to the long axis of the stalk, and the single large karyosome is replaced by a number of smaller granules, scattered throughout the nucleus. Sometimes the single vacuolar cavity contains two discrete, ovoidal masses of unequal size, each containing a nucleus with a single karvosome, and the stalk-nucleus is granular and flattened on the side of the vacuolar cavity. Aggregates of two or three individuals, with as many stalks for attachment, are also found, and each shows the structure as seen in a solitary individual. Sporocysts are pseudonavicellar (homopolar), and were seen to contain a single nucleus, eccentrically located within the finely granular contents.

*Remarks.*—The Gregarines when present are said to occur in large number, 50 to 90 per segment for a number of segments.

The size of the parasites was not determined. The parasites were observed in the fluid used for preserving the host, without clearing or staining them. In the absence of proper protozoological technique it is by no means certain that the branching stalk was not some tissue of the host, or that the organism is really binucleate. Also what is described by the author of the species as bi-stalked and tri-stalked forms may, perhaps,



Fig. 21.—*Nellocystis birmanica* Gates. *a*, portion of a solitary individual; *b*, portion of the anterior end, more highly magnified; *c*, bi-stalked form; *d*, tri-stalked form. (After Gates.)

have been syzygies or gametocytes in a process of association The organism is so remarkable that it would be well worth further detailed study.

Habitat.—Cœlom of the earthworm, Pheretima compta Gates : BURMA, Toungoo Hills.

# 6. Family DIPLOCYSTIDÆ Bhatia, 1930.

Trophozoites fusing in pairs to form spherical gametocysts. Sporocysts spherical or oval; octozoic.

The family includes two genera, *Lankesteria* and *Diplocystis*; only the former is known from India so far.

#### Genus LANKESTERIA Mingazzini, 1891.

Lankesteria, Mingazzini, 1891, p. 407, ; 1893, pp. 50, 63; Labbé, 1899, p. 46; Minchin, 1912, pp. 327, 329; Wenyon, 1926, pp. 1121-4, fig. 465; Reichenow, 1929, pp. 889-90; Bhatia, 1930, p. 161; Ray, 1933, pp. 392-6; Calkins, 1933, p. 559.

Trophozoites more or less spatulate or leaf-shaped, of small size. Epimerite small, described as an anterior pseudopodiumlike process. Gametocysts spherical, produced by association of two individuals after contraction. Sporocysts oval, octozoic. Intestinal parasites of Tunicates, Insects, etc.

#### Key to Indian Species.

1 (4).	Trophozoite elongate	2.
2 (3).	Trophozoites $50-200 \mu$ . Epimerite	
• /	simple or umbrella-shaped. Nucleus	
	spherical, with a single karvosome.	
	Gametocysts spherical. Sporocysts	
	spindle-shaped	L. culicis (Ross), p. 77.
3 (2).	Trophozoite with strongly thickened.	(
• •	often spherical anterior end. Nucleus	
	large, with one, two, or many karvo-	[p. 8].
	somes	L. tripteroidesi. sp. nov.
4 (1).	Adult trophozoite egg-shaped or pear-	,
- (-)-	shaped. $101.4 \mu$ by $78 \mu$ . Nucleus	
	large, spherical, with a single karvo-	
	some. Gametocysts spherical or broad-	[Swaminath], n. 79.
	ly oval. Sporocysta spindle-shaped	L. mackiei (Short &
	-, apsiss, sib opinate bioped it	Let montor (Dalor 0 co

#### 20. Lankesteria culicis (Ross). (Fig. 22.).

†Gregarina culicis, Ross, 1895, p. 346; 1898, p. 147; 1906, pp. 102, 103.

Gregarine sp., Marchoux, Salimbeni, and Simond, 1903, p. 713.
 Lankesteria culicis, Wenyon, 1911 a, p. 273; 1926, pp. 1121-4, fig. 465; Knowles, 1928, pp. 495-9; Reichenow, 1929, p. 890; Lan-Chou, 1930, pp. 301-2, pl. xii, figs. 1-20.
 †Lankesteria culicis, Ray, 1933, pp. 392-6, pl. xxiii.

Young trophozoites intracellular in the epithelial cells of the stomach of the host-larva, developing a simple pseudopodiumlike epimerite, by which the full-grown trophozoite remains attached to the epithelial cell while hanging into the lumen of the stomach. Eventually trophozoites become free and move about amongst the intestinal contents. The free-living forms have a granular cytoplasm, and at the anterior end the remains of the organ of fixation, or epimerite, can be detected. There is a large central spherical nucleus, with usually a single large karyosome.

When the larva becomes a pupa, the trophozoites leave the gut and enter the Malpighian tubes, within which they associate in pairs and form large spherical gametocysts. The gametes produced by the two Gregarines are of the same size, but differ as regards their nuclei; those produced by one have



Fig. 22.—Diagram of the life-cycle of Lankesteria culicis (Ross).
A, escape of eight sporozoites from oöcyst, and infection of intestinal cells of mosquito larva; B, growth of young Gregarine till it protrudes from the cell though remaining attached; C, free trophozoite in the lumen of intestine; D, association of two sporonts in the Malpighian tube; E-G, associated sporonts in gametocyst, showing nuclear multiplication; H, nuclei arranged on surface of body; I-J, formation of gametes; K, fusion of gametes; L, zygotes elongated and encysted in očcysts, in which nuclear multiplication and formation of eight sporozoites is taking place. (After Wenyon.) (×c. 1000.)

large nuclei and may be regarded as female gametes, while those by the other have small nuclei and may be regarded as male gametes. The zygotes become elongated and develop into spindle-shaped sporocysts; and in each sporocyst eight sporozoites are developed. When the adult mosquito hatches from the pupal case, its Malpighian tubes contain gametocysts filled with sporocysts. The gametocysts then rupture, sporocysts escape into the cavity of the Malpighian tubes, and make their way to the intestine, whenee they pass into water to await ingestion by other larvæ.

Dimensions.—A free-living trophozoite measures from 50–200 $\mu$  in length; a mature sporocyst measures  $10\mu$  by  $6\mu$ .

Remarks.—Ray (1933) has published his observations on this form. According to him two types can be readily distinguished. Those which infest the œsophagus and anterior part of the mid-gut, where the chitinous lining is thin and smooth, are always of intracellular habit and possess very feebly developed epimerites; while those in the posterior part of the mid-gut, where the epithelium is covered by a thickridged chitin, bear a definite epimerite and are all growing extracellularly, only anchored to the epithelium by the epimerite embedded in the host-cell. The epimerite is shaped like an open umbrella connected by a very short handle. The youngest forms in both cases measure  $10\mu$  by  $4-5\mu$ . At the anterior end of the body there is a clear cone-like zone, which takes up a dark, homogeneous stain, and which, according to Ray, is comparable with the protomerite of septate Gregarines. Such differentiation of the anterior portion of the eytoplasm is, however, not unknown in other Monocystids possessing an epimerite (vide Bhatia and Setna, 1927, figs. 18, 19, & 23), and unless there is a definite septum the organism cannot be placed among the SEPTATA.

The intracellular forms, when they become too large to remain within the host-cells, enter the gut-lumen, where they measure  $150-194 \mu$  by  $31-34 \mu$ . The extracellular forms attain the same size and, when mature, become detached, leaving the epimerite embedded in the epithelium.

Habitat.—Stomach of the larva, and Malpighian tubes of pupa and adult mosquito, Aëdes (Stegomyia) ægypti (Linn.): INDIA; and Aëdes (Stegomyia) albopictus Skuse: BENGAL, Calcutta.

21. Lankesteria mackiei (Short & Swaminath). (Fig. 23.)

Certain parasites, Mackie, 1915, p. 948.

Monocystis mackiei, Short & Swaminath, 1927, pp. 539-52, pls. li-

liv, figs. 1-18.

A Gregarine, Knowles, 1928, p. 501.

Lankesteria mackiei, Reichenow, 1929, p. 890.

The young intracellular stage occurs in a cell of the intestinal epithelium of the larva of a sand-fly (*Phlebotomus*), and is at first spherical, but later becomes more or less triangular, with rounded angles, measuring about  $23 \cdot 4\mu$  in length when about to leave the host-cell. The adult Gregarine occurs in the lumen of the gut or body-cavity of the larva, but only in the body-cavity of the pupe and the adult fly. It is egg-shaped or pear-shaped, but it can voluntarily lengthen or shorten the body and is considerably compressible. Epicyte well developed and marked with longitudinal striations. Endoplasm very markedly granular. Nucleus a large spherical

Fig. 24.

Fig. 23.

Fig. 23.—Lankesteria mackei (Short & Swaminath). (After Short & Swaminath.)
 Fig. 24.—Lankesteria tripteroidesi, sp. nov. (After Guenther.)

or subspherical body, measuring about  $30\,\mu$  in its longest diameter, eccentrically placed, with a large densely-staining spherical or subspherical karyosome, measuring 8–10  $\mu$  in diameter. Gametocytes somewhat elongated and crescentshaped, with blunt extremities. They are similar, and a pair become apposed and surrounded by a cyst. Gametocysts spherical or broadly oval. Hundreds of gametes are formed in each gametocyte and are similar in form and size. Zygotes secrete an impermeable membrane round themselves and become sporocysts. The sporocysts, when released from the original gametocyst, are broadly spindle-shaped, terminating in a knob-like projection at each end. Each sporocyst contains eight sporozoites, applied closely to one another in a tight bundle. Sporocyst ruptures at one or both poles while in the alimentary canal of the larval sand-fly, and the liberated sporozoites attack the epithelial cells of the gut.

Dimensions.—Young trophozoite (intracellular)  $23.4\mu$  in length; adult trophozoite (free in gut)  $101.4\mu$  in length by  $78\mu$  in greatest breadth; nucleus  $30\mu$  in length; karyosome 8–10 $\mu$  in diameter; gametocyst  $66.3-105.1\mu$  in length; sporocyst  $9.6\mu$  by  $5.8\mu$ .

Remarks.—The complete life-cycle of the Gregarine has been described by Short and Swaminath and correlated with the different stages in the life-history of the host. The sporocysts of the gregarine are passed out with the eggs by an adult sand-fly. As the larvæ hatch out, the sporocysts are taken into their alimentary canal with their first food. The sporozoites invade the epithelial cells, an intracellular stage is passed, and the destruction of the host-cells releases the Gregarines either in the alimentary canal or the body-cavity of the larva. The growth of the larvæ is accompanied by the growth of the Gregarines, but in the pupæ the Gregarines in the alimentary canal have completely disappeared, and in both pupæ and adults adult Gregarines are only found in the body-cavity, where all the other stages of gametogony and sporogony take place.

Habitat.—Alimentary canal and body-cavity of the larva, and in the body-cavity of the pupa and adult of the sand-fly, *Phlebotomus argentipes* Ann. & Brun: ASSAM; also in *P. papatasii* Scop., bred out in the laboratory: BENGAL, Calcutta.

#### 22. Lankesteria tripteroidesi, sp. nov. (Fig. 24.)

<sup>†</sup>A Monocystid Gregarine, Guenther, 1914, pp. 264–7, 5 text-figs. A Gregarine, Wenyon, 1926, p. 1149.

Trophozoite elongate, with a strongly thickened anterior end which is often spherical and may project laterally. Cytoplasm coarsely alveolar and filled with granules. There are a few vacuoles at the anterior end. Nucleus large, with a slimy protoplasmic thread extending forwards and backwards from it. The nucleus contains large dark nucleoli, which may be one, two, or many in number.

Habitat.—Body-cavity, respiratory tubes, and anal gills of the larva of the mosquito, *Tripteroides dofleini* (Guenther). (=Ficalbia dofleini (Guenther)): CEYLON.

SPOR.

# 2. Tribe HETEROPOLARIDEA Bhatia, 1930 (including Family CHOANOSPORIDÆ Dogiel).

Intestinal or cœlomic parasites usually of marine animals, such as Polychætes, Nemertines, Sipunculids, Echinoids, and Ascidians. Sporocysts with dissimilar poles. The tribe comprises four families (vide Bhatia, 1930), of which only one is known, so far, to be represented in India.

#### Identification Table of Families.

1 (2)	. Sporocyste provided with a typical funnel- like opening at one end	[Woodcock. Urosporidæ*
2 (1)	. Sporocysts not provided with a funnel-like opening at one end	3.
3 (6)	. Sporocysts oval, with a thickening at one	4 [Kamm n 99
4 (5)	Epimerite simple and deformable	Lecudinidæ
5 (4)	Sporonts held together by a ball-and-socket joint	{J. S. Huxley. Ganymedidæ*
6 (3)	Sporocysts spindle-shaped, with one side slightly more prominent than the other. Gametocysts elongated, sausage-like	{Bhatia. Allantocystidæ *

#### INCERTÆ SEDIS.

Sporocysts not know	vn. Body non-coptate,	
with rudimentary	opimorito. Develop-	[Henry.
ment intracellular.	Sporonts associative	Kofoidinidæ*

# Family LECUDINIDÆ Kamm, 1922, emend. Reichenow, 1929.

#### (Syn. DOLIOCYSTIDÆ Labbé, 1899.)

Body non-septate, distinguishable from the Monocystids by the protoplasm in the anterior portion possessing finer granules. A typical epimerite for attachment to the wall of the gut of the host is present, but may be lost in fully grown individuals. Syzygy does not occur. Sporocysts oval, with a thickening at one pole. Parasites of marine Annelids.

#### Key to Indian Genera.

1 (2).	Epimerite invaginable, assuming a	[p. 83.
•••	variety of forms	LECUDINA Mingazzini,
2 (1).	Epimerite of a definite form	3.
3 (4).	Epimerite bulb-like at the end of a long	[p. 85.
• •	rigid style	BHATIELLA Setna,
4 (3).	Epimerite funnel-like, on a long tubular	[p. 85.
• •	stalk	FERRARIA Setna,

#### Genus LECUDINA Mingazzini, 1891.

Gregarina (part), Kölliker, 1848, p. 35. Locudina, Mingazzini, 1891, p. 469. Doliocystis, Léger, 1893, pp. 204-6. Lecudina + Ophiodina, Mingazzini, 1893, p. 51. Ophioidina, Minchin, 1903, p. 196. Doliocystis, Minchin, 1903, pp. 202-3. Lecudina, Kamm, 1922, pp. 10, 23-6; Reichenow, 1929, p. 892; 1932, pp. 31-2; 1935, p. 368.

Body cylindrical or ovoid, cytoplasm of the anterior portion distinctly marked off by the possession of fine granules. Epimerite caducous or invaginable, able to take for any single species only a variety of well-determined forms. Intestinal parasites of Polychætes.

#### 23. Lecudina brasili Ganapathy & Aiyar. (Fig. 25.)

†Lecudina sp., Ganapathy & Aiyar, 1937, p. 292.

Youngest stages intracellular within the gut-epithelium, ovoid in outline, with a slight concavity at one end. The fullgrown trophozoite is widest at about a third of its length from the anterior end, and the nucleus, which is spherical, is situated at that level. The pellicle is uniformly thick and the eytoplasm granular, except in the prolongation at the anterior end. These trophozoites lie in the lumen of the gut, attached to the epithelial cells by an epimerite. In the fully evaginated condition the epimerite has a truncated base. with a slender prolongation ending in a darkly staining anchorplate. The pellicle does not appear to be continued over the epimerite, which has therefore been interpreted as endoplasmic origin and is capable of retraction and evagination. in The epimerite is entirely extracellular. In the retracted state the anterior region of the organism presents a slight concavity. When the organism detaches itself the anchor-plate is left behind, and a bubble of cytoplasm appears to protrude from the anterior end; this is in reality the evaginated epimerite after it has become detached from the gut-epithelium. In sections the whole epimerite, excluding the anchor-plate, shows faint longitudinal striations. Association takes place between two mature trophozoites, which become surrounded by a spherical gametocyst. Two kinds of gametes are produced and conjugation is anisogamous. Sporocysts are oval in outline, with a characteristic thickening at one pole. Eight sporozoites are developed inside each spore.

Dimensions.—Trophozoites measure  $150\mu$  by  $30\mu$ , length of epimerite  $10-12\mu$ ; gametocysts measure  $75\mu$  in diameter; sporocysts measure  $6\mu$  by  $4\mu$ .

Remarks.—Ganapathy and Aiyar (1937) have recently communicated a paper describing the different stages in the

life-cycle of the parasite. Only a brief abstract has been published so far, but through the courtesy of the authors I have been able to consult the paper before it appears in print. As first shown by Brasil (1908, 1909), there is a well-defined "epimerite," but it differs from that organ as it occurs in the Cephaline Gregarines. It is an invaginable apparatus, which is sometimes like a conical trunk as in *Leculina aphroditæ* (Lankester) or in *L. polydoræ* (Léger) and sometimes like



Fig. 25.—Lecudina brasili Ganapathy & Aiyar. Trophozoite attached to the gut-wall. (After Ganapathy and Aiyar.)

a spherical button as in L. elongata (Mingazzini), but the form of the trophozoite is different from any of those species, and resembles that of L. pellucida (Kölliker), in which the epimerite is described as a simple small papilla. I do not agree with the authors in thinking that the epimerite is endoplasmic. Their figure shows that the thick pelliele does not extend forward over the epimerite, but they describe striations which are certainly epicytal. The so-called epimerite is merely a deformable prolongation of the anterior end, and when the anchor-like portion breaks off there is a large wound through which the entocyte would seem to flow out. The trophozoites are parasitized by *Metchnikovella* sp.

Habitat.—Intestine of Lumbriconercis sp. : MADRAS, Madras.

#### Genus BHATIELLA Setna, 1931.

Bhatiella, Setna, 1931, p. 203.

Epimerite in the form of a bulb-like structure at the end of a long rigid style.

#### 24. Bhatlella morphysæ Setna. (Fig. 26.)

†Bhatiella morphysæ, Setna, 1931, pp. 203-4, pl. v, figs. 1, 2.

Solitary, non-septate Gregarine, with a pear-shaped body, widest just behind the middle. Epimerite in the form of



Fig. 26.-Bhatiella morphysæ Setna. (After Setna.)

a distinct bulb-like structure at the extreme tip of a long rigid style. Dehiscence by simple rupture, no sporoducts.

Dimensions.—Trophozoite, maximum 200  $\mu$  by 103  $\mu$ , minimum 100  $\mu$  by 40 $\mu$ .

*Remarks.*—The epimerite is about one-fourth the total length of the body, and its style is broad at the base and slender at the apex. The protoplasm does not show finer granules in the anterior portion of the body. The nucleus is ellipsoidal, situated in the posterior half of the body, and contains a relatively large karyosome.

Habitat.—Mid-gut of Morphysa sanguinea Montague (= M. furcellata Crossland) : ANDAMANS, Port Blair.

#### Genus FERRARIA Setna, 1931.

Ferraria, Setna, 1931, p. 205.

Epimerite a wide-mouthed, funnel-like structure on a long tubular stalk.

*Remarks.*—Setna (1931) has described the Gregarine as septate, and placed it in the family Polyrhabdinidæ Kamm. Reichenow (1929), however, considers that the presence of a septum in *Polyrhabdina* as described by Kamm is an error, and the septate appearance is due to the protoplasm in the anterior portion of the body being more finely granular. He has consequently amalgamated the family Polyrhabdinidæ with Lecudinidæ. Setna describes and figures the protoplasm as dark and finely granular throughout, and indicates the septum as a clear area, which may be an artefact.

#### 25. Ferraria cornucephali Setna. (Fig. 27.)

*Ferraria cornucephali*, Sotna, 1931, pp. 205-6, pl. v, figs. 3-5; pl. vi, fig. 1.

A solitary, rather stout-bodied Gregarine. Epimerite a widemouthed, funnel-like structure on a long, slender, tubular stalk. Nucleus large and spherical, with a large central karyosome. In the digestive tract of a polychæte, quite commonly attached to the intestinal wall, or free in the lumen of the gut.



Fig. 27.-Ferraria cornuccphali Setna. (After Setna.)

Dimensions.—Trophozoite. maximum 300  $\mu$  by 91  $\mu$ , minimum 243  $\mu$  by 81  $\mu$ .

Remarks.—Setna has described the form as a septate Gregarine, but this is probably an error (see remarks under the genus). He gives ratio of length of protomerite to total length of trophozoite as 1:4: width of protomerite to width of deutomerite as 1:1.6. The anterior part (so-called protomerite) is hemispherical to subglobular, widest behind. The posterior part (so-called deutomerite) is elongated, cylindrical and ovoidal, widest about its middle and well rounded posteriorly. There is little or no constriction at the septum, which appears to be indicated by a clear area between the anterior and the posterior portions.

Two distinct types of gametocysts and sporocysts were encountered in the mid-gut of the host, but neither was definitely associated with the trophozoites of this or of the preceding species. One type consisted of spherical gametocysts, measuring 90-100  $\mu$  in diameter, and full of oval sporocysts, measuring 10  $\mu$  by 4.5  $\mu$ . Sporocysts escape by rupture of the gametocysts. The other type of gametocyst contained a mass of very peculiar sporocysts,  $20 \mu$  by  $4 \mu$  in size, and with rounded anterior and posterior ends; each sporocyst is bilaterally symmetrical, covered by a thick membrane, and contains a well-defined nucleus.

Habitat.—Mid-gut of Morphysa sanguinea Montague (=M. furcellata Crossland): ANDAMANS, Port Blair.

#### INCERTÆ SEDIS.

#### Genus DIRHYNCHOCYSTIS Cognetti, 1921.

Dirhynchocystis, Cognetti, 1921 a, p. 150; 1925, pp. 229, 233. Echinocystis, Bhatia & Chatterjee, 1925, p. 197. Dirhynchocystis, Bhatia, 1929, p. 125; Reichenow, 1929, p. 887; Bhatia, 1930, p. 165.

Solitary. Body ovoid, with two subcylindrical appendages, arising from opposite sides. Sporocysts biconical, with similar poles.

*Remarks.*—Cognetti (1921) describes the two spine-like prolongations as anterior and posterior, and regards each as comparable to the trunk-like process arising from the anterior end of *Rhynchocystis*. To me these spines suggest a strong comparison with the antero-lateral spines of *Ancora*, from which the form can be derived by shortening and rounding off the body.

26. Dirhynchocystis globosa (Bhatia & Chatterjee). (Fig. 28.)

+Echinocystis globosa, Bhatia & Chatterjee, 1925, pp. 197-9, pl. viii, figs. 18, 19; pl. ix, figs. 26, 27.

Dirkynchocystis globosa, Cognetti, 1925, p. 233; Bhatia, 1929, p. 125; Reichenow, 1929, p. 887; Bhatia, 1930, p. 165.

Trophozoite possessing a more or less spherical body, with two spine-like structures radiating from the surface. Nucleus



Fig. 28.—Dirhynchocystis globosa (Bh. & Ch.). (After Bhatia and Chatteriee.)

large, generally ovoid, and containing a single karyosome. Sporocysts biconical, with two similar truncated poles. Dimensions of the trophozoite  $74\,\mu$  by  $65\,\mu$ .
Remarks.--Infection is very rare. The young trophozoite within the blastophore has no spine-like structures. The adult trophozoite lives free in the seminal vesicles and possesses the spines: it has a dark aspect in the living condition on account of reserve of paraglycogen granules, and shows slow movements. Form of the adult is like a globe drawn out along one axis. The spine-like structures taper gradually towards the distal end, and consist of a non-granular endoplasm covered by sarcoeyte and epicyte, thus resembling in structure the trunk-like epimerite of Rhynchocystis: they disappear during association. Nucleus large, slightly elongate and oval, generally situated near the middle of the body, and may attain a length of  $24\mu$ : it contains a single large central karyosome. Sporocysts, doubtfully belonging to this species, were unusually large, measuring  $28 \mu$  by  $14 \mu$ , and one of the sporocysts contained only two sporozoites.

Habitat.—Seminal vesicles of *Pheretima posthuma* (L. Vaill.): PUNJAB, Lahore; BOMBAY, Bombay. Seminal vesicles of *Eutyphœus* sp.: UNITED PROVINCES, Lucknow.

# Genus GRAYALLIA Setna, 1927.

Grayallia, Setna, 1927, pp. 335-7; Bhatia, 1929, p. 126; 1930 p. 165; Reichenow, 1929, p. 887.

Monocystid with four spines at either end. The spine-like prolongations arise from the surface of the body at a little distance from the extremities : they are broader at the base than at the apex and are subequal.

# 27. Grayallia quadrispina Setna. (Fig. 29.)

†Grayallia quadrispina, Setna, 1927, pp. 335-7, figs. 1-3.

The trophozoite resembles *Nematocystis* in form, with four spine-like processes arising from the surface of the body



Fig. 29.—Grayallia quadrispina Setna. s, one of the spines. (After Setna.)

at either end and radiating backwards. Body chalky-white in colour and highly deformable. Nucleus large and oval, containing a variable number of karyosomes. Cysts and sporocysts not identified.

Dimensions.—Size 720–1270 $\mu$  in length by 35–50 $\mu$  in width. Remarks.—The endoplasm of the parasite is very granular and mobile and, owing to its very active movements, the parasite assumes a variety of appearances. When extended, the breadth is comparatively uniform, but when the parasite contracts a characteristic shape is assumed. The epicytal striations are well developed over the general body surface; they are faintly visible, and lie close together on the spines. The sarcocyte is feebly developed and the myocyte is also thin; both these layers are continued into the spines, but the endoplasm is not. The spines are about  $12\mu$  in length and are capable of movement, being swung actively from side to side or held straight against the body. In the endoplasm are lodged paraglycogen bodies and many small, oval, and sometimes rod-shaped bodies, which may be parasites.

Habitat.—Seminal vesicles of Pheretima heterochæta (Mchlsn.): BOMBAY, Bombay.

# 2. Legion SEPTATA Lankester, 1885

# (=CEPHALINA Delage, excluding Doliocystidæ, or POLY-CYSTIDEA, sensu stricto).

The body is divided into the *protomerite* and *deutomerite* by an ectoplasmic septum, and an organ of attachment, known as the *epimerite*, is always present, at least in the carlier stages. An end-to-end association of two or more trophozoites (*syzygy*) is common: in such the anterior individual is called the *primite*, and the posterior ones the *satellites*. Parasites of the digestive tracts of Invertebrates, especially Arthropods.

Labbé's classification (1899) into families is generally followed with modifications. Kamm (1922) divides the group into twelve families. Reichenow (1929) has combined two of these, Lecudinidæ and Polyrhabdinidæ, into one, which has been transferred to HAPLOCYTA in this work. Reichenow has also transferred the family Porosporidæ to this group, while two new families, Monoductidæ and Hyalosporinidæ, have recently been described by Ray and Chakravarti (1933), and a third, Kofoidinidæ, by Henry (1933). Of these there does not seem to me to be sufficient justification for separating the Hyalosporinidæ from the Stenophoridæ, and Kofoidinidæ is placed under HAPLOCYTA in this work. There are thus eleven families in this legion.

# SPOROZOA.

# Identification Table of Families.

$     \begin{array}{ccc}       1 & (8) \\       2 & (7)     \end{array} $	. Sporonts in associations or solitary . Sporonts in associations of 2 or 3	2. 3.
3 (4)	Associations of 2; form syzygies early. Development intracellular. Epimerite	
	cysts dehisce by simple rupture. Sporo-	[Kamm.
4 (3)	cyst ovoidal, with equatorial line Associations of 2 or 3. Development	Cephaloidophorldæ*
5 (6)	extracellular	5.
• (•)	septum in satellites. Gametocysts de-	
	ellipsoidal. Entire life-cycle in a single	[Léger.
6 (5).	Epimerite absent or weakly developed.	Dlaymophylaæ *
	Gametocysts dehisce by simple rup-	
<b>F</b> (0)	Cycle shows alternation of hosts	Porosporidæ* Labbé.
1 (2)	to 12. Epimerite simple, symmetrical.	
	Development extracellular. Gameto- cysts dehisce by simple rupture or by	
	spore-ducts. Sporocysts oval or	[p. 96. Cragorinidos Labbá
8 (1)	Speronts solitary	9.
9 (10)	velopment intracellular. Gametocysts	
	dehisce by simple rupture. Sporo- cysts ovoidal, with equatorial line, not	[Dub., p. 9].
10 (9)	extruded in chains	Stenophoridæ Lég. &
11 (20)	opment extracellular	11.
11 (20)	or by means of a lateral pseudocyst	12.
12 (17). 13 (14).	Gametocysts dehisce by simple rupture. Epimerite a large cup bordered with	13.
	hooks and placed on a long, slender neck. Sporocysts crescentic	[Léger. Menosporidæ*
14 (13).	Epimorite without a long neck. Sporo-	15
15 (16).	Sporocysts irregular, biconical or cylin-	[Léger, p. 107.
16 (15).	Sporocysts with equatorial and polar	Acunocepnalidæ [Léger.
17 (12).	spines	Acanthosporidæ *
18 (19)	lateral pseudocyst or by simple rupture.	18.
10 (10).	form or root-like prolongations. Sporo-	[Léger, p. 108.
19 (18).	Epimerite symmetrical, with or without	Daciylophoridæ
	appendages. Sporocysts pouch-like, brown or black, in chains	[A. Schneider. Stylorhynchidæ *
20 (11).	Gametocysts dehisce by a single elon- gated spore-duct	21.
21.	Epimerite a small elevation with prongs	,
	Sporocysts compressed, broadly spindle-	[0]
	snaped, with median ridge on dersal surface	Unak., p. 109. Monoductidæ Ray &

# 1. Family STENOPHORIDÆ Léger & Duboscq, 1904.

Development intracellular. Trophozoites solitary. Epimerite absent or of a simple structure. Gametocysts dehisce by simple rupture. Sporocysts ovoidal, with broad epispore and with equatorial line. Not extruded in chains. Parasitic in Diplopods.

Key to Indian Genera.

1 (2).	Epimerite absent or rudimentary. Sporo-	[p. 91.
	cyst ovoidal, with equatorial line	STENOPHORA Labbé,
2 (1).	Epimerite small, tongue-like, bordered by a collar at its base. Sporocyst oval, with a surrounding hyaline membrane	[Chak., p. 92. Hyalosporina

#### Genus STENOPHORA Labbé, 1899.

Gregarina (part), Frantzius, 1848, pp. 191-4. Stenocephalus, Aimé Schneider, 1875, p. 584.

Stenophora, Labbé, 1899, p. 15; Minchin, 1903, p. 198; Légor & Duboscq, 1904, pp. 361-79; Watson, 1916, p. 48; Kamm, 1922, p. 10; Reichenow, 1929, p. 894; Kudo, 1931, p. 297; Calkins, 1933, p. 561.

Trophozoites solitary. Epimerite absent or a mere knob. Gametocysts open by simple rupture. Sporocysts oval, with a broad epispore and with an equatorial line. Not extruded in chains.

## Key to Indian Species.

Epimerite round ; protomerite bottle-shaped,	
with a process at its posterior end;	[p. 91.
deutomerite ellipsoidal. Nucleus elliptical.	S. ellipsoidi Chak.,
Epimerite conical or rounded; protomerite	
rounded anteriorly and flattened at the	
septum; deutomerite elongated, tapering	(p. 92.
posteriorly. Nucleus spherical	S. khagendræ Ray,

## 28. Stenophora ellipsoldi Chakravarti. (Fig. 30.)

†Stenophora ellipsoidi, Chakravarti, 1934, pp. 164-8, figs. 1-6.

Young trophozoite intracellular, with a round epimerite and a bottle-shaped protomerite; not known to penetrate beyond the nucleus of the epithelial cells. Sporonts ellipsoidal. Epimerite persists for some time. Protomerite in the adult has its posterior end drawn out into a small blunt process which produces a depression in the septum. Deutomerite ellipsoidal, grows more rapidly than the protomerite. Nucleus elliptical and variable in position. Gametocysts spherical, develop outside the body of the host, and dehisce by simple rupture. Sporocysts spindle-shaped, octozoic. Dimensions.—Trophozoite, young  $8-10.25\mu$  in length by  $4\mu$  in width; sporonts  $250-372\mu$  by  $50-95\mu$ ; gametoeysts  $118-172\mu$ ; sporocysts  $10\mu$  by  $4\mu$ .

Habitat.---Mid-gut of a Millipede, Diplopoda sp. : BENGAL, Calcutta.

## 29. Stenophora khagendræ Ray. (Fig. 31.)

†Stenophora khagendræ, Ray, 1933, pp. 343-51, figs. 1-9.

Young trophozoite intracellular, with a hyaline, conical or rounded epimerite. Sporonts solitary, "parrot-shaped." Epimerite disappears. Protomerite rounded at the anterior end and flattened at the septum, slightly broader than long.



Fig. 30.—Stenophora ellipsoida Chak. (After Chakravarti.) Fig. 31.—Stenophora khagendræ Ray. A young trophozoite. (After Ray.)

Deutomerite up to eight times as long as the protomerite and one and a half times as broad, broadest slightly behind the septum, and in full-grown individuals gracefully tapering towards the posterior end. Nucleus spherical. Gametocyst spherical, dehiscing by simple rupture to release spindle-shaped sporocysts.

Dimensions.—Trophozoite, young  $10-30\mu$  in length by  $6-16\mu$  in width; sporont,  $225\mu$  by  $56\mu$ ; gametocyst  $100-123\mu$  in diameter; sporocysts  $10\cdot25\mu$  by  $4\mu$ .

Habitat.—Intestine of a Millipede (belonging to a new species and probably to a new genus) related to Zikadesmus Chamb. : BENGAL, Calcutta.

## Genus HYALOSPORINA Chakravarti, 1935.

Hyalosporina, Chakravarti, 1935, pp. 211–18; Chakravarti & Mitra, 1936, p. 346.

Sporonts solitary. Epimerite a small tongue-like elevation bordered by a collar at its base. Gametes dissimilar; during fertilization only the nucleus of the male gamete transferred to the female gamete. Gametoeysts dehisce by simple rupture. Spores oval, with a surrounding hyaline membrane.

Remarks.—Characters such as intracellular development, the simple nature of the epimerite, solitary sporonts, and dehiscence of eysts by simple rupture clearly indicate the relationship of the genus with the Stenophorida. In the trophozoite the nucleus is tethered to the pellicle by myonemes, in this respect resembling Monoductus, but the spore has a hyaline membrane round it and is quite unlike that of either Monoductus or On this basis Chakravarti places the genus Stenophora. in a new family. Hyalosporinidæ, intermediate between Stenophoridæ and Monoductidæ. The genus has so many characters in common with the Stenophoridæ that I do not consider it necessary to place it in a new family on the basis of the character of the spore alone, especially when we remember that the character of the spore is not known for the majority of the species of Stenophora nor for both the other genera previously referred to the family.

## Key to Indian Species.

1 (2)	. Sporonts 800-1111 μ by 80-111 μ. Gam- etocysts oval, 292-390 μ by 263-375 μ.	p. 93.
	Sporocysts oval, 8 $\mu$ by 6 $\mu$	H. cambolopsisæ Chalk.,
2(1)	Trophozoites $130-173 \mu$ by $37-70 \mu$ .	
	Gametocysts spherical, 96–120 $\mu$ in diameter. Sporocysts oval 6.2 $\mu$ by	In 95
	$4\cdot 12\mu$	H. rayi Chak. & Mitra.

## 30. Hyalosporina cambolopsisæ Chakravarti. (Fig. 32.)

*†Hyalosporina cambolopsisæ*. Chakravarti, 1935, pp. 211-18, pl. vii, figs. 1-19.

Youngest forms penetrate the opithelial cells, pass beyond the nucleus, and develop intracellularly. Older forms, measuring 43-150  $\mu$  by 14-30  $\mu$ , are attached to the epithelial cells by an epimerite and grow extracellularly. Epimerite a simple structure, consisting of a darkly staining collar or ring, which grasps the host-cell, and a tongue-like process, which is inserted into the cell. In still older forms very fine rootlike processes are seen to arise from the ring and project over the tongue-like elevation. Protomerite small and conical in shape, with its orifice plugged by a darkly staining granule which disappears later, the pellicle becoming thickened in this region. Deutomerite the longest segment and circular in transverse section. Longitudinal epicytal striations more prominent on the deutomerite than on the protomerite; a very thin homogeneous layer of sarcocyte, and below that a layer of circular myonemes or myocyte to which the nuclear myonemes are tethered. Nucleus spherical, measuring  $55\mu$ 

by  $35\mu$ , and containing a spherical karyosome measuring  $15\mu$ in diameter. Two sets of myoneme fibres, arising from the nuclear membrane, run backwards and are tethered to the myocyte in the side-wall. Oval gametocysts are expelled



Fig. 32.- Hyalosporina cambolopsisæ Chakravarti. A, young trophozoite growing intracellularly; B, older trophozoite, with epimerite attached by the collar and the tongue-like process thrust into the host-cell; protomerite is at the breakingpoint, and shows the granule; C, adult trophozoite, with the nucleus tethered to the side-wall by the myonemes; D, mature sporocyst, showing the hyaline membrane, and eight sporozoites. (After Chakravarti.)

#### HYALOSPORINA.

with the host's excreta and complete development in five or six days if kept in a moist chamber. Gametes dissimilar; male gametes have one end pointed and female gametes are spherical. During fertilization only the nucleus of the male gamete is transferred to the female gamete. Gametocysts dehisce by simple rupture and release oval sporocysts. A hyaline membrane, more prominent at one pole than the other, surrounds each spore, and eight sporozoites are arranged superficially along the long axis of the spore.

Dimensions.—Trophozoites, young, attached,  $43-150\mu$  by  $14-30\mu$ ; sporonts  $800-1111\mu$  in length by  $80-111\mu$  in breadth; gametocysts  $292-390\mu$  by  $263-375\mu$ ; sporocysts  $8\mu$  by  $6\mu$ .

Habitat.—Alimentary canal of a Millipede, Cambolopsis sp. : BENGAL, Calcutta.

# 31. Hyalosporina rayi Chakravarti & Mitra. (Fig. 33.)

*†Hyalosporina rayi*, Chakravarti & Mitra, 1936, p. 346; Chakravarti, 1936, pp. 116-20, figs. 1-5.

Sporonts solitary, with early intracellular growth. Trophozoites elongate, the broadest part being slightly behind the



Fig. 33.—Hyalosporina rayi Chak. & Mit. A, trophozoite; B, sporocyst. (After Chakravarti.)

septum. Epimerite a rounded structure with a darkly staining area at its anterior extremity. Protomerite small

and triangular in shape, with a darkly staining dot or special type of granules. Deutomerite broader anteriorly, gradually tapering posteriorly, packed with granules. Nucleus spherical, with a central karyosome in the young forms, oval in the adult, tethered to the pellicle by two sets of myoneme fibres. Gametocysts spherical, rupturing by simple dehiscence. Sporocysts elongate oval, with an outer hyaline coat which is prominent at both poles. On one side of the spore there is a circular lidlike structure which stains deep brown with Lugol's solution.

Dimensions.—Trophozoites  $130-173 \mu$  by  $37-70 \mu$ ; gametocysts  $77-116\cdot 5 \mu$  in diameter; sporocysts  $6\cdot 2 \mu$  by  $4\cdot 12 \mu$ .

Habitat.—Alimentary canal of the Millipedes Polydesmus sp. and Strongylosoma contortipes Attems : BENGAL, Calcutta.

# 2. Family GREGARINIDÆ Labbé, 1899.

(Syn. CLEPSIDRINIDÆ Léger, 1892.)

Epimerite symmetrical, simple. Sporonts solitary or in associations up to 12. Development extracellular. Gametocysts with or without sporoducts, sporocysts symmetrical. Confined mostly to insects.

Kamm (1922) enumerates eleven genera as belonging to this family, and others have been described since. Only five genera are known from India.

#### Key to Indian Genera.

1 (3).	Sporonts solitary	2.
2.	Epimerite a simple, sessile knob. Dehis- conce by spore-ducts. Sporocysts	fp. 97.
	barrel-shaped, in chains	LEIDYANA Watson,
3 (1).	Sporonts in associations of 2 or more	
	individuals	4.
4 (7).	Sporonts associated in pairs	5.
5 (6).	Epimerite a simple globular or eylindrical	
• •	papilla. Dehisconce by spore-ducts.	
	Sporocysts barrel-shaped or cylindri-	[p. 98.
	cal, in chains	GREGARINA Dufour.
6 (5).	Satellite with a septum. Epimerite	,
• •	dilated anteriorly like a cauliflower	
	and narrower at the base. Dehiscence	
	by simple rupture. Sporocysts ovoidal	[& Set., p. 102.
	or spherical	CAULOCEPHALUS Bh.
7 (4).	Sporonts in associations of 2 to several	
	individuals	8.
8 (9).	Sporonts attenuate, often attached	
	laterally. Gametocysts not known.	
	Sporocysts barrel-shaped, with a spine	[Pinto, p. 103.
	at each corner	<b>PROTOMAGALHÆNSIA</b>
9 (8).	Sporonts in associations of 2 to 12 or	
	more. Epimerite a small cylindrical	
	papilla. Gametocysts spherical, de-	
	hisce by simple rupture. Sporocysts	[p. 105.
	ovoidal	HIRMOCYSTIS Léger,
		-

## Genus LEIDYANA Watson, 1915.

Leidyana, Watson, 1915, p. 35; 1916, p. 44; Kamm, 1922, pp. 10, 72; Reichenow, 1929, pp. 895-6; Kudo, 1931, pp. 296-7.

Sporonts solitary, epimerite a simple globular sessile knob. Gametocyst possesses spore-ducts. Sporocysts barrel-shaped, in chains.

## 32. Leidyana gryllorum (Cuénot) Watson. (Fig. 34.)

Clepsidrina gryllorum, Cuénot, 1897, pp. 52-4. Gregarina macrocephala, Labbé, 1899, p. 10. Gregarina gryllorum, Cuénot, 1901, pp. 594-5. Leidyana gryllorum, Watson, 1916, pp. 120-1, fig. 209. †Leidyana gryllorum, Bhatia & Setna, 1924, p. 288.

Sporonts solitary, never associative, cylindrical. Ratio of length of protomerite to total length 1:5; width of protomerite



Fig. 34.—Leidyana gryllorum (Cuénot). (After Cuénot.) Fig. 35.—Leidyana xylocopæ Bh. & Set. (After Bhatia and Setna.)

to width of deutomerite 1: 1.1. Epimerite globular and sessile. Protomerite subspherical, with deep constriction at septum. Deutomerite cylindrical, generally conical at the end. Nucleus spherical, generally with three karyosomes and numerous fine chromatin particles. Gametocysts spherical or ovoidal. Spore-ducts 3-8  $\mu$  in length; sporocysts barrel-shaped.

 $\hat{D}imensions.$ —Sporonts 420  $\mu$  in length ; gametocysts 190– 240  $\mu$  in diameter ; sporocysts 7  $\mu$  in longer axis. SPOR.

Remarks.--Watson (1916) has separated this species from L. erratica; but Bhatia and Setna (1924) are inclined to doubt if L. erratica and L. gryllorum are really distinct. Specimens having a broadly rounded protomerite may be rounded posteriorly, and conversely a specimen which might be referred to L. gryllorum (by virtue of a deep constriction at the septum and a conical posterior end) may possess a protomerite which is conical anteriorly. Several specimens show intermediate conditions in all these respects.

Habitat.-Gizzard and mid-gut of Gryllus sp.: PUNJAB, Lahore.

### 33. Leidyana xylocopæ Bhatia & Setna. (Fig. 35.)

†Leidyana xylocopæ, Bhatia & Setna, 1924, pp. 279-81, figs. 1-8.

Body elongate and eylindrical, rounded anteriorly and slightly tapering posteriorly, chalky-white in colour and filled with paraglyeogen granules. Epimerite a large, simple, sessile knob, narrowing but slightly near the base. Length of protomerite to total length as 1:10; width of protomerite to width of deutomerite as 1:1.4. Sporonts always solitary, varying in form and size. Length of protomerite to total length as 1:4.6-7; width of protomerite to width of deutomerite as 1:1.3-1.8. Protomerite rounded or dome-shaped, and there is a deep constriction at the septum. Deutomerite elongate, slightly broader in the middle and tapering posteriorly; posterior end always rounded. Nucleus varies in position in the deutomerite, and is a large rounded vesicle with a sharply defined membrane and a large spherical karyosome. Gametocysts and sporocysts not known.

Dimensions.—Fairly large cephalont  $90\mu$  in length; sporonts 23-174  $\mu$  in length.

Habitat.—Alimentary canal of the carpenter bee, Xylocopa æstuans (Linn.) : PUNJAB, Lahore.

#### Genus GREGARINA Dufour, 1828.

Gregarina, Dufour, 1828, p. 366.

Clepsidrina (corr. Clepsydrina), Hammerschmidt, 1838, p. 355.

Gregarina (part), Stein, 1848, p. 199. Gregarina, Schneider, 1873, pp. 515-33. Clepsydrina, Aimé Schneider, 1875, p. 572.

Gregarina, Labbé, 1899, p. 9; Watson, 1916, p. 43; Kamm, 1922, p. 11; Wenyon, 1926, p. 1117; Reichenow, 1929, pp. 894-5; Kudo, 1931, p. 296; Calkins, 1933, p. 561.

Sporonts associated in pairs. Epimerite a simple globular or cylindrical papilla. Gametocysts with spore-ducts; sporocysts barrel-shaped or cylindrical, extruded in chains.

# 34. Gregarina aciculata, sp. nov. (Fig. 36.)

†Gregarine parasite, "species B," Cornwall, 1915, pp. 130-1, pl. x, fig. 31; pl. xi, figs. 37, 38.

Young trophozoite intracellular in a swollen cell of the stomach. Full-grown trophozoite cordate, with a long acicular epimerite, penetrating between the epithelial cells right down to the basement membrane. Free individuals often assume peculiar shapes during syzygy. Sporonts coalesce, acquire a common capsule, and become small, white, shining gametocysts, only just visible with a lens. These cysts are passed out



Fig. 36.—Gregarina aciculata, sp. nov. A, adult trophozoite; B, gametocyst with spores coming out in a long chain. (After Cornwall.)

with the fæces, and often several are found in the same mass. Gametocysts possess a thick, smooth wall and a central sporal mass. In the course of seven days or so protrusions from the inside develop into curved or angular tubes about as long as the diameter of the cyst. Sporocysts are then thrust out through these tubes in long strings. Sporocysts very small, about  $4\mu$  by  $2\mu$ , symmetrically curved, and joined end to end. Dehiscence is along one border.

*Remarks.*—This species is more abundant than the larger one found in the same host.

Habitat.—Mid-gut of Lepisma saccharina Linn. (?): MADRAS, Coonoor.

## 35. Gregarina cornwalli, sp. nov. (Fig. 37.)

†Gregarine parasite, "species A," Cornwall, 1915, pp. 128-30, pl. xi, figs. 31 A-36.

Large; clearly visible to the naked eye when freed from the stomach of its host. In the fully developed trophozoite the epimerite contains a delicate transverse septum, and is divided off from the protomerite by another well-defined



Fig. 37.—Gregarina cornwalli, sp. nov. A, adult trophozoite; B, chain of sporocysts. (After Cornwall.)

septum. A trophozoite does not have its attachment in any particular cell; the globular epimerite appears to be held in a cavity, that it has formed, by the pressure of the neighbouring cells round its neck. Several cells seem to have been destroyed to make room for it. In dissections trophozoites readily free themselves from their attachments, either leaving the epimerite behind or soon casting it off. The remaining two segments wander about the field. Deutomerite contains an irregular-shaped nucleus in its anterior part. Nucleus contains a distinct, centrally placed karyosome, with a ring of chromatin material round it. Sporonts coalesce and surround themselves with a common capsule. Gametocyst rapidly becomes globular and is discharged with the fæces. After about

#### GREGARINA.

twelve days cysts rupture and allow the contained mass of sporocysts to escape. The contents of the eyst are long strings of sporocysts, interspersed with residual protoplasm. Each sporocyst has a thick protective covering and is roughly oval, with the curvature greater on one side than the other. A sporocyst contains eight shapeless sporozoites, grouped four and four at each end.

Remarks.—The form described above is sufficiently distinct from G. lagenoides (Léger), the only species previously known from Lepisma saccharina Linn.

Habitat.—Mid-gut of Lepisma saccharina Linn. (?): MADRAS, Cooncor.

# 36. Gregarina oviceps Diesing.

Gregarina achetæ abbreviatæ, Leidy, 1853, p. 238; 1856, p. 47. Gregarina oviceps, Diesing, 1859, p. 730. Gregarina achetæ, Lankester, 1863, p. 94. Gregarina achetæ abbreviatæ, Labbé, 1899, p. 35; Crawley, 1903a, p. 45; 1903 b, p. 639; 1907, pp. 220-1; Ellis, 1913 b, p. 266; Watson, 1915, p. 34. Gregarina oviceps, Watson, 1916, pp. 101-2, figs. 191-2.

†Gregarina oviceps, Bhatia & Setna, 1924, pp. 287-8.

Sporonts bi-associative, obese. Ratio of length of protomerite to total length of primite as 1:3 or more, width of protomerite to width of deutomerite as 1:1.1. Protomerite hemispherical to subglobose, width twice the length or even Slight constriction at septum. Deutomerite stoutmore. bodied, nearly as wide as long, up to twice as long as wide; widest at shoulder, where it is very little wider than protomerite. Posterior end truncate. Epimerite simple, and elongated like a nipple. Endocyte denser in deutomerite than in the protomerite, and distinctly marked off from the sarcocyte. Nucleus oval, with fine membrane and many fine chromatin granules. Gametocysts spherical. Spore-ducts 2 to 5, with maximum length of  $1000 \,\mu$ . Sporocysts barrelshaped.

Dimensions.—Sporonts, average,  $450\,\mu$  in length by  $225\,\mu$ in width; maximum length  $500 \mu$ ; gametoeysts  $250 \mu$  in average diameter ; sporocysts  $4.5 \mu$  by  $2.25 \mu$ .

Remarks -- The sporonts examined by Bhatia and Setna were shorter and narrower, the ratio of the length of the protomerite to the total length of the primite being as 1:4 or even  $1:5\cdot3$  instead of 1:3, as given by Watson. The width of protomerite was twice its length or even slightly more. The deutomerite was stout, and varied from nearly as wide as long up to twice as long as wide.

Habitat.-Gizzard and mid-gut of Gryllus sp.: PUNJAB, Lahore.

# Genus CAULOCEPHALUS Bhatia & Setna, 1924.

Caulocephalus, Bhatia & Setna, 1924, pp. 284-7; Ray & Chatterjee, 1936, p. 345.

Bi-associative; satellite with a septum. Epimerite dilated anteriorly like a cauliflower and narrowing at the base. Protomerite with a characteristic specialized zone anteriorly. Gametocysts dehisce by simple rupture. Sporocysts ovoidal or spherical.

# 37. Caulocephalus crenata Bhatia & Setna. (Fig. 38.)

†Caulocephalus crenata, Bhatia & Setna, 1924, pp. 284–7, figs. 12–23; Ray & Chatterjee, 1936, p. 345.

Body elongate, cylindrical, densely granular and opaque. Epimerite usually dilated anteriorly like a cauliflower and



Fig. 38.—*Caulocephalus crenata* Bh. & Set. A, adult trophozoites; B, enlarged view of the epimerite. (After Bhatia and Setna.)

narrow basally; its surface crenate. Protomerite elongate and conical and usually  $1\frac{1}{2}$  times as long as wide, usually widest about the middle, with a characteristic specialized zone at its anterior end. There is a distinct constriction at the septum. Deutomerite cylindrical,  $1\frac{1}{2}$  to 3 times as long as

broad, widest at its middle and gradually narrowing towards the rounded posterior end. Nucleus large and spherical or somewhat oval, and usually placed about the middle or a little in front of it, with a distinct nuclear membrane and one or Length of protomerite to total length as two karyosomes. 1:3·3-4·0; width of protomerite to width of deutomerite as 1: 1.2-1.5. In the sporonts the protomerite is considerably reduced in proportion to the total length of the body. Length of protomerite to total length as 1:6; width of protomerite to width of deutomerite as 1:1.6. Sporonts biassociative. Sporonts forming association are much more elongate than cephalonts. Gametocysts spherical. Sporocysts ovoid or nearly spherical.

Dimensions.—Sporonts 40-142  $\mu$  in total length; gametocysts about 90  $\mu$  in diameter; sporocysts 12  $\mu$  in diameter.

Remarks.—The epimerite, though presenting a crenated surface, cannot be regarded as complex; it varies in appearance and ultimately becomes simple and symmetrical. The protomerite resembles that of *Pyxinoides balani* and *P. cthamali* (vide Tregonboff, 1912), but the epimerite differs from that of *Pyxinoides*, which is described as a style dilated in the middle. Ray and Chatterjee (1936) have recently studied this parasite from the same host at Calcutta, and are of opinion that the cauliflower-like appearance of the epimerite is due to the action of the fixative or some other disturbing factor. Their observations have not yet been published in full.

Habitat.—Alimentary canal of the beetle Aulacophora foveicollis Kust. (=A. abdominalis G. et H.): PUNJAB, Lahore and Kalka; BENGAL, Calcutta.

# Genus PROTOMAGALHÆNSIA Pinto, 1918.

Protomagalhænsia, Pinto, 1918; 1923, pp. 90-91 (translation, pp. 25-6), pls. iii, fig. 48, & vi, fig. 93; Kamm, 1922, p. 11; Setna & Bhatia, 1934, pp. 38-42.

Sporonts in associations of two to several individuals, often attached laterally. Sporonts attenuate. Myonemes prominent. Epimerite unknown. Gametocysts unknown. Sporocysts barrel-shaped, with a spine at each corner.

**Remarks.**—In addition to the characters given above, Pinto (1923) mentions in the diagnosis of the genus that the development is always intracellular, and that in the syzygies the protomerite of the satellite embraces the deutomerite of the primite, like a pair of forceps. As he places the genus in the family Gregarinidæ Labbé, in which the development is extracellular, the word "intracellular" is probably a misprint for extracellular. Protomagalhænsia (?) attenuata Setna & Bhatia. (Fig. 39.)
 †Protomagalhænsia (?) attenuata, Setna & Bhatia, 1934, pp. 40-2, figs. 12-22.

Sporonts attenuate, forming early syzygies. Associations of



Fig. 39.—Protomagalhænsia (?) attenuata Set. & Bh. A, free cephalont; B, linear syzygy; C, lateral attachment of satellites to the primite; D, three satellites attached to a primite; E, four satellites attached to the posterior end of the first satellite; F, two primites followed by a single satellite. (After Setna and Bhatia.)

#### HIRMOCYSTIS.

two to several individuals, often attached laterally. Peculiar associations of two primites with a single satellite sometimes met with. Ratio of length of protomerite to total length as 1:13; width of protomerite to width of deutomerite as  $1:1\cdot1$ . Epimerite a simple rounded knob. Protomerite hemispherical; deutomerite cylindrical, rounded or flattened behind. Satellite usually longer than the primite, tapering posteriorly. Nucleus spherical or oval, with single karyosome. Gametocysts and sporocysts not known.

Dimensions.—Associations, maximum length  $473 \mu$ , maximum width  $22 \mu$ ; free sporonts  $20-70 \mu$ .

*Remarks.*—As in *Hirmocystis* (?) *parapeneopsisi* Set. & Bh., a few cases were seen of a unique type of association in which two primites were followed by a single satellite. Both the primites and the satellite were of a slender type and comparatively short in length.

In the lateral type of syzygy great diversity of form and attachment is met with. There may be a satellite in a line with the primite, and a second satellite arise from the side of the primite near its posterior end, or both the satellites may be attached laterally and diverge like two legs. Sometimes there are three satellites, all attached to the posterior end of the primite, and in still others several individuals may be seen attached to the posterior end of the first satellite.

The form resembles *Protomagalhænsia* in as much as the sporonts are attenuate and form associations of several individuals, while lateral attachment of sporonts is common. Spores in that genus are of a characteristic barrel-shape, with a spine at each corner. Until the spores are known the species cannot definitely be placed.

Habitat.—Intestine of the prawn, Parapeneopsis sculptilis (Heller): BOMBAY, Bombay.

# Genus HIRMOCYSTIS Léger, 1892.

Eirmocystis (corr. Hirmocystis), Léger, 1892, p. 110.

Hirmocystis, Labbé, 1899, pp. 12–13; Watson, 1916, pp. 167-9;
Kamm, 1922, pp. 11, 74–5; Reichenow, 1929, p. 896; Calkins, 1933, p. 562; Henry, 1933, pp. 102-9; Setna & Bhatia, 1934, p. 35.

Sporonts in associations of two to twelve (or more) in linear series, sometimes bifurcated or trifurcated. Epimerite a small conical or cylindrical knob, caducous. Gametocysts spherical: dehiscence by simple rupture. Sporocysts ovoidal, with two coats.

# Hirmocystis (?) parapeneopsisi Setna & Bhatia. (Fig. 40.) *†Hirmocystis* (?) parapeneopsisi, Setna & Bhatia, 1934, pp. 35-8, figs. 1-11.

Sporonts stoutly built, forming early syzygies. Associations of two or three individuals in linear chains, or unique associations of two primites with a single satellite. Ratio of length of protomerite to total length as 1:5-7; width of protomerite to width of deutomerite as  $1:1\cdot4$ . Epimerite a small rounded knob. Protomerite hemispherical or flattened, constricted at the septum; deutomerite cylindrical or barrelshaped, broadly rounded or flattened behind. Satellite longer than the primite, second satellite longer than the first. Nucleus spherical, with a single karyosome. Gametocysts





spherical or slightly elliptical. Dehiscence of gametocysts and sporocysts not observed.

Dimensions.—Associations, maximum length  $425\mu$ , maximum width  $44\mu$ ; free sporonts  $40-80\mu$ .

Remarks.—A few cases were met with in this species of the rare type of association in which two primites were followed by a single satellite. The primites were posteriorly rounded off and fitted into curved depressions in the anterior margin of the satellite. In other cases two satellites ran into one another and showed partial or complete fusion, the deutomerite containing one or two nuclei.

The form resembles *Hirmocystis* in as much as the sporonts form associations of two or more, and the epimerite is small,

STEININA.

knob-like, and caducous; but the species cannot be definitely placed in that genus until the dehiscence of the cyst and the form of the sporocysts has been observed.

Habitat.—Intestine of the prawn, Parapeneopsis sculptilis (Heller): BOMBAY, Bombay.

# 3. Family ACTINOCEPHALIDÆ Léger, 1892.

Sporonts always solitary. Epimerite symmetrical, simple or with appendages. Gametocysts dehisce by simple rupture. Sporocysts irregular, biconical or cylindrical, with conical extremities not provided with bristles.

# Genus STEININA Léger & Duboseq, 1904.

Steinina, Léger & Duboscq, 1904, p. 352; Watson, 1916, p. 256;
 Kamm, 1922, p. 79; Wenyon, 1926, p. 1151; Reichenow, 1929,
 p. 900; Kudo, 1931, p. 298; Calkins, 1933, p. 562.

Epimerite a short, mobile, digitiform process, changing into a flat crenulate disc. Sporocysts biconical.

# 40. Stelnina metaplaxi Pearse. (Fig. 41.)

†Steinina metaplaxi, Pearse, 1933, p. 293, fig. 3.

Body robust, somewhat flattened. Epimerite nearly spherical. Protomerite wider than long, rounded anteriorly. Deutomerite rectangular, with rounded angles. There is



Fig. 41.-Steinina metaplaxi Pearse. (After Pearse.)

a heavy pellicle around the protomerite and the deutomerite. Nucleus ellipsoidal, longest axis lying across body, situated usually at end of anterior third of deutomerite, but the position varies. Syzygy apparently does not occur.

Dimensions.—Sporont, length 40-60 $\mu$ , breadth 18-20 $\mu$ ; protomerite 16 $\mu$  by 10 $\mu$ ; deutomerite 25 $\mu$  by 18 $\mu$ ; nucleus 6 $\mu$  by 7 $\mu$ .

Remarks.-The genus has previously been reported from beetles and fleas only. I doubt if the species has been correctly referred to the genus Steinina, as in that genus the epimerite is a short, mobile, digitiform process changing into a flat crenulate disc.

Habitat.-In the intestine of the Indian crab, Metaplax dentipes Heller : BENGAL, Port Canning.

# 4. Family DACTYLOPHORIDÆ Léger, 1892.

Epimerite complex. Sporonts solitary. Gametocysts dehisce with lateral pseudocyst or by simple rupture. Sporocysts elongate, cylindrical or ellipsoidal. Parasites in Chilopoda.

# Genus GREBNECKIELLA, nom. nov.

(=NINA Grebnecki, 1873).

*Nina*, Grebnecki, 1873, p. 264.

Pterocephalus, Aimé Schneider, 1887, pp. 67-8; Labbé, 1899, p. 17; Léger, 1899, pp. 390-3; Minchin, 1903, pp. 171, 172, 190, 198; Léger & Duboseq, 1903, p. 333.

Nina, Léger & Duboscq, 1903, p. 353. Nina, Léger & Duboscq, 1909, pp. 33-68; Sokolow, 1911, pp. 281-2. Pterocephalus, Minchin, 1912, pp. 173, 327, 329, 330, 339. Nina, Watson, 1916, pp. 44, 82-4; Kamm, 1922, p. 15. Pterocephalus, Wenyon, 1926, pp. 74, 1141; Knewles, 1928, p. 500. Nina, Reichenow, 1929, pp. 904-5; Kndo, 1931, pp. 74, 298; Calkins, 1933, p. 562; Reichenow, 1935, p. 370.

Protomerite formed of two long, narrow, horizontal lobes, fused and upturned spirally at one end. Periphery shows many teeth, and long slender filaments project from it. Cyst dehisces by pseudocyst. Spores long-ovoidal, united in oblique chains. In the intestine of Myriopods. Remarks.—The name Nina is preoccupied for a molluscan

genus, Nina J. E. Gray, 1850. The second name, Pterocephalus, is also inadmissible, being preoccupied for an elasmobranch fish, Pterocephalus Swainson, 1838. It may incidentally be remarked that the name *Pterocephalus* has also been wrongly used for a Nematode (O. Linstow, 1899) and a Trilobite (F. Raw, 1907). I have, therefore, renamed the genus Grebneckiella, after the author of the type-species.

# 41. Grebneckiella navillæ (Mitra & Chakravarty).

Nina navillæ, Mitra & Chakravarty, 1937, p. 1.

Sporonts solitary. Gametocysts oval. Sporocysts spherical to oval, with two envelopes; liberated in chains.

MONODUCTUS.

Dimensions.—Sporonts  $819-975\mu$  by  $97-190\mu$ ; gametocysts  $125-175\mu$  by  $95-125\mu$ .

*Remarks.*—Full observations have not yet been published, and it is not certain that the form is distinct from other previously known species.

Habitat.—In the intestine of Scolopendra sp.: BENGAL, Calcutta.

# 5. Family MONODUCTIDÆ Ray & Chakravarty, 1933.

Sporonts solitary. Epimerite a small elevation with prongs attached to its base. Gametes dissimilar. Gametocysts dehiscing by a single elongated spore-duct. Sporocysts compressed, of broad spindle-shape, with median ridge on dorsal surface.

Genus MONODUCTUS Ray & Chakravarty, 1933.

Monoductus, Ray & Chakravarty, 1933, p. 359.

With the characters of the family.

# 42. Monoductus lunatus Ray & Chakravarty. (Fig. 42.)

†Monoductus lunatus, Ray & Chakravarty, 1933, pp. 352-60, pl. ii, text-figs. 1-5.

Young trophozoite intracellular. Full-grown trophozoite elongate, widest slightly behind the septum. Epimerite knob-like, with twelve to sixteen stiff, radiating processes attached to its neck. Protomerite more or less conical, relatively very small, with an orifice at its anterior extremity, through which longitudinal myonemes run connecting the base of the epimerite with a raised disc-like platform arising from the middle of the septum. There is a series of transverse myonemes also, just underlying the pellicle. Deutomerite elongate, circular in transverse section, with circular myonemes well developed, and with the cytoplasm usually packed with granules, which give the organism an opaque yellowish-white appearance. Nucleus contains a single spherical karyosome, and is shaped like a parachute, a number of fibrils running back from its posterior concave surface which vary the form and position of the nucleus. Prior to association, sporonts develop posterior pseudopodial processes by which coupling is effected. A thin transparent cyst is formed round the paired gametocytes. The spherical gametocysts are expelled with the host's excreta and complete their development in three

or four days if kept moist. There is no distinction between the gametocytes, but one gives rise to slightly drawn out and pointed (male) gametes and the other to perfectly elliptical (female) gametes. Sporocysts compressed, truncate at one pole, and with a median ridge. They are released through a single spore-duct in a long chain formed of pairs set obliquely.



Fig. 42.—Monoductus lunatus Ray & Chak. A, adult; B, same, showing the orifice in the protometerite and the longitudinal myonemes passing through it. (After Ray and Chakravarti.)

Dimensions.—Full-grown trophozoite  $225-445 \mu$  by  $33-47 \mu$ ; length of epimerite  $7\cdot 5-10\cdot 25 \mu$ ; nucleus  $30 \mu$  by  $12 \mu$ ; gametocysts  $225-230 \mu$ ; sporocysts  $10\cdot 25 \mu$  by  $4 \mu$ .

Habitat.—Alimentary canal of a Millipede, Strongylosoma contortipes Attems : BENGAL, Calcutta.

# II. Suborder SCHIZOGREGARINARIA Léger, 1900.

(Syn. AMœBOSPORIDIA A. Schneider, 1884.)

The Schizogregarines are parasites of the digestive tract and appended organs (e. g., Malpighian tubules) of Arthropods, Annelids, and Tunicates. The sporocyst gains entrance into the digestive tract of the specific host and the sporozoites are set free. These develop into trophozoites either in the lumen of the gut or within the cells of the host and undergo schizogony,



Fig. 43.—Life-cycle of a typical Schizogregarine, Schizocystis gregarinoides Léger. A, sporozoite escaping from the spore; B-E, growth of the sporozoite into the multinucleate schizont, of which there are two types, the vermiform schizont (a) which attaches itself to the epithelial cell by its anterior end, and the massive schizont (b) which lies free in the gut of the host; F, division of the schizont into a number of merozoites, which may grow into schizonts again ( $G^1$ ,  $G^2$ ), or grow into gamonts ( $G^3$ ); H, young gamonts; I, association of two full-grown gamonts; J, formation of a common cyst round the two; K, division of nuclei in each gametocyte; L, formation of gametes; M, fusion of gametes in pairs; N, each zygote becomes an oöcyst and develops eight sporozoites in it. (After Léger.) which may be binary or multiple fission or budding. As in EUGREGARINARIA, two full-grown trophozoites (sporonts) associate and give rise to gametes, which conjugate to produce zygotes. Each zygote becomes an oöcyst. The number of oöcysts produced by each pair of gametes varies from one to thirty. Each oöcyst, popularly called a spore, contains one to eight sporozoites.

Léger and Duboseq (1908) divided the suborder into Polysporidea and Monosporidea according to the number of oöcysts produced by a pair of associated Gregarines. Fantham (1908) on the other hand divided the group into Endoschiza and Ectoschiza, according as schizogony took place within a cell or was extracellular respectively. Keilin (1923) is of the opinion that neither of these classifications is a natural one, and that it is not impossible that the process of schizogony has been secondarily acquired by some members of the EUGREGARINARIA and that the various genera of SCHIZOGRE-GARINARIA will ultimately be distributed among the families of the EUGREGARINARIA. Wenyon (1926) and Ray (1930) have supported this view.

Reichenow (1929) does not group the genera into families, but Minchin (1912), Doflein (1916), and Calkins (1926) recognized a number of families, each based on one or two genera.

## Identification Table of Families.

1	(7).	Gametocyst contains a single oöcyst .	2.
2	(4).	Oöcyst contains a single sporozoite	3.
3		Gregarine spirally wound or cros-	[Dub.
		centic; schizogony intracellular	Spirocystidæ * Lég. &
4	(2).	Occyst contains eight sporozoites	5.
5	(6).	Gregarine ovoid, with an opimerite;	[Porter.
		schizogony intracellular	Merogregarinidæ *
6	(5).	Gregarine conical, with pseudopodial	
		processes along its base : schizogony	[& Dub.
		extracellular	Ophryocystidæ * Lég.
7	(1).	Gamotocyst contains many oöcysts	8.
8	(15).	Only one type of schizogony	9.
9	(12).	Schizogony intracellular	10.
10	(11).	Gregarine elongated, with longitudinal	
		striations, and chromatic bodies at	
		the anterior end. Gametocyst con-	
		tains many oöcysts, each containing	
		four or eight sporozoites	Seienidiidæ * Brazil †.
H	(10).	Grogarine elongate. Gametocyst con-	
		tains sixteen oöcysts, cach containing	
		eight sporozoites	Lipotrophldæ * Keilin.
12	(9).	Schizogony extracellular	13.
13	(14).	Gregarine vermicular. Gametocyst	
		contains large number (about thirty)	
		of oöcysts, each containing eight	[Dub.
		sporozoites	Schizocystidæ * Lég. &
~	+ Acc	cording to Ray (1930) certain species	of Selenidium do not

show any schizogony.

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14 (13). 15 (8).	Gregarine ovoid, with a process for attachment. Gametocyst contains eight oöcysts, each containing eight sporozoites	<b>Caulleryellidæ *</b> Keilin.
	cellular and the other extracellular. Gametocyst contains a large number of vöcents, each containing aicht	fwaylonghy
	or obeyses, each containing eight	yaviensky.
	sporozoites	Menzebleridæ * Bogo-

Up to the present time nobody has worked on the Schizogregarines of India.

# II. Order COCCIDIA Leuckart, 1879.

The COCCIDIA have a wide distribution, occurring as parasites in the Vertebrates as well as the higher Invertebrates. They are generally found as parasites of the epithelium of the digestive tract and associated glands. Alternation of generations is invariably present : asexual reproduction is by schizogony, and is followed by sexual reproduction by, in most cases, anisogamy. During the whole of their growth the male and the female gametocytes are apart and develop independently of one another. The male gametocyte generally produces a relatively large number (six or more) of male gametes. Both kinds of reproduction take place in the body of one and the same host (except in the Aggregatidæ and the Hæmogregarinidæ). The Coccidia are divided into two suborders, as follows :—

1.	Gametocytes dissimilar in size; associated	
	with each other during the later part of	[p. 113.
	trophic life; microgametes few	Adeleldea Léger.
2.	Gametocytes similar in size; independent,	
	each microgametocyte developing into	[p. 156.
	numerous microgametes	Eimeridea Léger,

# I. Suborder ADELEIDEA Léger, 1911.

The schizonts develop into micro- and macrogametocytes which become closely associated and develop in contact with one another. The microgametocyte produces a few (two or four) microgametes. The zygote divides into numerous sporoblasts, each of which develops into a sporocyst with two or four sporozoites. These sporozoites, which are small gregarinulæ, penetrate the epithelial cells of the host and grow into large rounded or oval schizonts (agamonts). The SPOB. nucleus undergoes repeated division and the schizont divides into a corresponding number of merozoites, which are set free within the lumen of the organ. Each merozoite infects another cell. After several such generations the merozoites develop within the cell into micro- and macrogametocytes.

Léger (1911) divided the suborder into three families, Hæmogregarinidæ, Legerellidæ, and Adeleidæ, on the basis of the number of sporocysts and contained sporozoites. Reichenow (1921 and 1929) grouped all the genera in a single family, Adeleidæ Léger, emend. Nöller (1928), after consideration of the various schemes proposed, is in favour of



Fig. 44.—Stages in the life-cycle of Adelina dimidata, a typical member of the suborder. (×1700.) A, association of macrogametocyte and microgametocyte; B, nuclear division in microgametocyte and formation of gametie nuclei; C, sporocyst with two sporozoites. (From Wenyon, after Schellack.)

employing Léger's system, as a matter of practical convenience, until a generally recognized natural system can be adopted.

Following Kudo (1931), the suborder is here divided into two families, Adeleidæ and Hæmogregarinidæ, which correspond to the two suborders of similar name of Wenyon (1926).

#### Identification Table of Families.

1	(2).	Parasitic in the epithelium of the digestive	
		Invortebrates. Zygote motionless ; be-	
		comes onclosed in a resistant occyst,	
		sexual and asexual cycles occur in one	[emend., p. 115
0	(1)	host	Adeieidæ Léger,
ц.	(1).	system of Vertebrates. Zygote motile :	
		forms an occyst which increases in size.	
		Alternation of hosts, asexual cycle in a Vertebrate and sexual cycle in an	Léger, p. 117.
		Invertebrate	Hæmogregarinidæ

# 1. Family ADELEIDÆ Léger, 1911, emend.

Two gametocytes, one of which is smaller in size, unite in a kind of pseudo-conjugation. The nucleus of the microgametocyte divides once or twice, and one of its products enters the macrogamete and fuses with its nucleus. The zygote is motionless and becomes enclosed in a resistant oöcyst, which does not increase in size. The sexual and asexual cycles occur in the same host. They are intestinal parasites of Invertebrates, and infection is contaminative.

Wenyon (1926) raised the group to the rank of a suborder, under the title ADELEIDEA, and divided it into four families, viz., Dobellidæ Ikeda, 1914; Legerellidæ Léger, 1911; Adeleidæ Mesnil, 1903; and Klossiellidæ Wenyon, 1926. Reichenow (1929) and Kudo (1931) place the family Dobellidæ in the suborder EIMERIDEA. The other genera so far known can conveniently be treated as belonging to a single family.

# Key to Indian Genera.

1 (2).	Oöcyst thin-walled, containing numerous	[p. 115.
	disc-shaped spores	ADELEA Schneider,
2 (1).	Oöcyst thick-walled, containing smaller	[p. 116.
	number of spherical spores	ADELINA Hesse,

## Genus ADELEA Aimé Schneider, 1875.

Adelea, Aimé Schneider, 1875, p. 598; Schaudinn & Siedlecki, 1897,
p. 193; Labbé, 1899, p. 56; Perez, 1899, pp. 694-6; Minchin,
1903, p. 233; Dobell, 1907, pp. 155-62; Jollos, 1909, pp. 249-62; Schellack & Reichenow, 1910, pp. 380-3; 1915, pp. 425-60;
Léger, 1911, pp. 71-88; Debaisieux, 1911, pp. 259-66; Minchin,
1912, p. 352; Greiner, 1918, pp. 522-9; Wenyon, 1926, p. 1069;
Nöller, 1928, p. 189; Reichenow, 1929, p. 912; Kudo, 1931,
pp. 277-8; Calkins, 1933, p. 544.

The zygote develops into a large thin-walled occyst which 12 eontains a variable number of flattened sporocysts, each with two sporozoites. Other characters as in the suborder and the family.

# 43. Adelea pachelabræ de Mello.

†Adelea pachelabræ, de Mello, 1921, p. 242. Adelea pachylabræ, Wenyon, 1926, p. 1069. Adelea pachelabræ, Reichenow, 1929, p. 912.

Schizonts show a sexual dimorphism. Macroschizonts rounded or oval, with the eytoplasm markedly alveolar, containing granules of reserve material, and the nucleus surrounded by a distinct membrane. Binary fission equal or unequal, leading to formation of up to sixteen nuclei. Microschizont has a feebly alveolar, non-granular cytoplasm, and the nucleus is compact and stains deeply. Macromerozoite is oval, with alveolar cytoplasm and spherical nucleus. Micromerozoite is oval or fusiform, and its nucleus contains a distinct karyosome. Fertilization is preceded by the association of gametes, one or several male gametes (micromerozoites) being attached to a female gamete (macromerozoite). Female gamete is rounded oval, and possesses a voluminous nucleus which does not show a karyosome. Male gamete is generally smaller and attaches itself to one of the poles of the female gamete. Zygote develops into an oöcyst with two binucleate sporoblasts, each nucleus representing a future sporozoite. Sporocysts dizoie.

Remarks.—Wenyon (1926) considers it doubtful if this parasite belongs to this genus.

Habitat.—Intestine and digestive glands of a Molluse, Pachelabra mæstra Reeve : PORTUGUESE INDIA, Nova Goa.

# Genus ADELINA Hesse, 1911.

Klossia (part), Aimé Schneider, 1885, p. 7.

Adelea (part), Labbé, 1896, p. 536; 1899, p. 56; Minchin, 1903, pp. 233, 235, 332.

Adelina, Hosse, 1911, pp. xv-xix; Wenyon, 1926, pp. 1070-2;
 Reichenow, 1929, pp. 912-4; Kudo, 1931, p. 278; Calkins, 1933, p. 566; Reichenow, 1935, pp. 371-2.

Occyst thick-walled. Sporocysts spherical and comparatively few in number.

# 44. Adelina schellacki Ray & Das-Gupta.

Adelina schellacki, Ray & Das-Gupta, 1937, p. 291.

Oöeyst oval or egg-shaped, without residue; eight spherical sporoblasts, with two sporozoites, developed in each within 10 to 15 days if the unsegmented oöcysts are kept in a moist chamber. The oöcystal membrane ruptures after being in the moist chamber for four or five days, and along with this the unused microgametes are discarded.

Habitat.—Intestine of the centipede, Cormocephalus dentipes Poc.: BENGAL, Calcutta.

# 2. Family HÆMOGREGARINIDÆ Léger, 1911.

The researches of Reichenow and others have shown the coccidial nature of the Hæmogregarines, long known to occur in the red or white blood-corpuscles of all classes of Vertebrates. They are in reality COCCIDIA, which have certain stages adapted to life within the circulating cells of the Vertebrate blood. Schizogony may or may not take place in the blood-corpuscles, but the gametocytes always enter the blood-corpuscles. The fertilization process is of the Adelea-type. The formation of oöeyst and sporozoites takes place in the body of the blood-sucking Invertebrate, which in turn transfers the sporozoites to a second Vertebrate host.

The group is also related to Lankesterellide, in which the life-cycle is typical of that of EIMERIDEA and takes place either in the intestine (the usual habitat of COCCIDIA) or in endothelial cells of the blood-vessels.

Wenyon (1926) raised the group to the rank of a suborder, under the title Hæmogregarinidea, and divided it into three families, viz., Hæmogregarinidæ Neveu-Lemaire, 1901; Hepatozoidæ Wenyon, 1926; and Karyolisidæ, Wenyon, 1926. Reichenow (1929) included the Hæmogregarines, as well as *Adelea* and related genera, in a single family, Adeleidæ Léger emend. I have, however, followed Kudo (1931) in placing the Hæmogregarines in a separate family from the Adeleidæ.

## Key to Indian Genera.

1 (3).	Schizogony in the red blood-corpuseles	
	of the internal organs of a Vertebrato .	2.
2.	Gametogony and fertilization in the	
	body of blood-sucking Invortebrate	
	(leech). Occyst small, directly pro-	
	ducing eight sporozoites, without form-	[]
	ing sporocysts	HÆ
3 (1).	Schizogony not in the red blood-cor-	
	puscles	4.
4 (5).	Schizogony in cells of the internal organs	
	(liver, spleen, bone-narrow, etc.) or	
	loucocytes of Vertebrates. Gameto-	
	gony and fertilization in the body of	
	a blood-sucking Invertebrate (tick,	
	mite, etc.). Oöcyst increases enor-	
	mously in size, producing sporoblasts,	
	sporocysts, and sporozoites in the	
	oöcvst	HEP
	·····	

[Danilewsky, p. 119. HÆmogregarina

p. 145. Hepatozoon Miller,



Fig. 45.-Life-cycle of Hæmogregarina stepanowi Danilewsky. The figures to the right of the dotted line represent the phases in the blood of the tortoise; those to the left the phases in the leech. A, sporozoite; B, C, early schizogony producing a large number of merozoites; D, merozoite penetrating a blood-corpuscle; E-H, later schizogony, in which few merozoites are produced; in F is seen the recurved vermicule within the corpuscle; I, free merozoite about to enter a corpuscle and repeat the stages D-H, or to initiate the next phase; J, K, final schizogonous generation which produces the gametocytes;  $L_1$ ,  $L_2$ , sexually differentiated merozoites which grow up into micro-  $(M_1)$  and macrogametocytes  $(M_2)$ respectively, and develop further in the leech when taken up by it; N, association of micro- and macrogametocytes in the gut of the leech: O, formation of four microgametes by the microgametocyte; P, one of the microgametes has penetrated the body of the macrogamete and fused with its nucleus; Q, zygote with synkaryon, the degenerating remains of the male gametocyte are attached to it, and are also seen in the next four stages ; R, S, T, successive divisions of the synkaryon; U, ripe cyst containing eight sporozoites. (From Minchin, after Reichenow.)

5 (4). Schizogony in the endothelial cells of blood-vessels of a Vertebrate. Gametogony and fertilization in the body of a mite. Oöcyst produces sporoblasts, which are liberated as motile vermicules and infect the host's eggs. The mite, hatched from the egg, has the sporo-cysts in its intestinal epithelium. These are cast off and voided with the fæces, which are eaten by the Vertebrate host. The sporozoites make their way to the ondothelial cells

{p. 154. KARYOLYSUS Labbé,

## Genus HÆMOGREGARINA Danilewsky, 1885.

Hæmogreyarina, Danilewsky, 1885, pp. 588-98.

Danilewskya, Labbó, 1894, p. 124.

Danilewskya+Laverania, Billet, 1895, p. 30.

 Hamogregarina, Labbé, 1899, pp. 76-7; Minchin, 1903, pp. 265, 266-7, fig. 77; 1912, pp. 372-6; Wenyon, 1926, pp. 1081-4; Roichenow, 1929, pp. 924-7; Thomas & Robertson, 1929, pp. 104-5, 109-10; Kudo, 1931, pp. 279-81; Reichenow, 1932, p. 45; Calkins, 1933, pp. 545, 567.

Schizogony takes place in the red blood-corpuscles or other cells of the body of Vertebrates, merozoites escape from the original host-cell and infect other corpuseles or cells. After several generations merozoites develop into gametocytes in the red blood-corpuscles. These are taken up with the blood by an Invertebrate (leech), in the gut of which association of the micro- and macrogametocytes occurs. The gametocytes escape from their host-cells, become more or less spherical, associate in pairs, and a cyst-wall is formed, enclosing both the macro- and microgametocyte. The microgametocyte produces two to four microgametes, and the macrogametocyte forms a single macrogamete. One of the microgametes fertilizes the adjacent macrogamete, and a small occyst is formed. The oöcyst directly produces eight sporozoites without the formation of sporocysts.

The genus was founded by Danilewsky for the Hæmogregarine H. stepanowi Danilewsky of the European tortoise. The life-history was described by Reichenow (1910) and may be followed from fig. 45.

45. Hæmogregarina berestneffi Castellani & Willey. (Fig. 46.)

- *Hæmogregarina* sp. (probably new), Berestneff, 1903, pp. 343-8, pl. viii, figs. 1-9.
- Hæmogregarina berestneffi, Castellani & Willey, 1905, p. 397.
- †Hæmogregarina sp., Patton, 1908, p. 319. †Hæmogregarina berestnefft, Dobell, 1910, p. 67, pl. ii, figs. 3-8.
- Hæmogregarina berestnieffi, de Mello, de Sá, de Sousa. Dias, & Noronha, 1917, p. 13.
- †Hæmogregarina sp., Donovan (first recorded in Wenyon, 1926, p. 1398).
  - Hæmogregarina berestneffi, Wenyon, 1926, p. 1398; Scott, 1926, p. 238.
  - Hæmogregarina encapsulæ, Wenyon, 1926, p. 1398.

Intra-corpuscular individuals of various forms and sizes, many of them showing a characteristic pink-staining sheath. Free gregariniform individuals actively motile in the blood plasma. Small forms enter by boring directly into the corpuscle. Occasionally the organism, on reaching the inside of the corpuscle, rests for a few minutes and then wriggles out again into the plasma.

Dimensions.—Length  $26-28\mu$ ; width  $4-5\cdot5\mu$ .



Fig. 46.—*Hæmogregarina berestneffi* Cast. & Will. *A*, intra-corpuscular form; *B*, free form. (After Dobell.)

*Remarks.*—According to Berestneff the parasite lies within a strongly refringent colourless capsule, both ends of which are rounded and curved, the whole embracing the displaced nucleus of the corpuscle after the manner of a *Halteridium*; it shows a very close resemblance to H. magna, found in the same hosts, but differs by the possession of a capsule. The parasite was named H. berestneffi by Castellani & Willey (1905), who, however, did not find the parasite in hosts of the same species from Ceylon. Dobell (1910) found the species in Rana tigrina from Ceylon. Patton (1908, p. 319) notes that he had the opportunity of studying no less than five Hæmogregarines in  $\hat{R}$ . tigrina and R. hexadactula and also in the leech which transmits them, but he does not mention the species encountered. Wenyon (1926, p. 1398) records H. encapsulæ Berestneff as a parasite of R. limnocharis and both H. encapsulæ Berestneff and H. berestneff Castellani & Willey from R. tigrina. The name H. encapsulæ Berestneff is an error, as Berestneff did not give this name to his form, and so it must be treated as a synonym.

Habitat.—Blood of Rana tigrina Daud. and R. limnocharis Wiegmann: BOMBAY, Bombay; Rana tigrina Daud.: CEYLON, Colombo; R. tigrina Daud. and R. hexadactyla Lesson, and in the leech which transmits them: MADRAS, Madras; R. tigrina Daud.: PORTUGUESE INDIA, Nova Goa; also R. tigrina Daud. from India, in the Zoological Gardens, London.

# 46. Hæmogregarina cantllei Sambon & Seligmann. (Fig. 47.)

*†Hæmogregarina* sp., Laveran, 1905 (as noted in Wenyon, 1926, p. 1392).

- *Hæmogregarina cantliei*, Sambon & Seligmann, 1907 a, p. 1650; 1907 b, p. 310.
- *†Hæmogregarina* sp., Patton, 1908, p. 318.
  - Hæmogregarina cantlici, Dobell, 1908, p. 293; Phisalix, 1913, pp. 304-7, 6 figs.; Castellani & Chalmers, 1919, p. 487; Wenyon, 1926, p. 1392.

Hæmogregarina sp., Wenyon, 1926, p. 1392.

Adult sporonts club-shaped, the anterior third rounded or globular and the rest of body tapering to a slender tail; enclosed within thick sausage-shaped capsules. Nucleus median or nearer posterior extremity, oval or irregular in outline and with chromatin frequently arranged in cross-strands.



Fig. 47.—Hæmogregarina cantliei Samb. & Selig. A. uninfected corpuscle; B. intra-corpuscular form surrounded by a capsule; C. D. same without capsule; E. free forms. (After Phisalix.)

Dimensions.—Free sporonts,  $16-18\mu$  in length by  $3\cdot5\mu$  in breadth; tail  $2-3\mu$  in length; nucleus  $4\mu$  long by  $3\mu$  broad.

Habitat.—Blood of the snake, Eryx conicus Schneid. (=Gongylophis conicus): MADRAS, Madras.

#### 47. Hæmogregarina hankini Simond. (Fig. 48.)

- *†Hæmogregarina hankini*, Simond, 1901 b, pp. 183-5; 1901 e, pp. 331-8, pl. vii.
- *†Hæmogregarina* sp. (? *H. hankini* Simond), Dobell, 1910, pp. 68, 79, pl. ii, figs. 9, 10.
- Hæmogregarina hankini, Castellani & Chalmers, 1919, p. 486; Wenyon, 1926, p. 1396.

The parasite has two forms, vermicular and oval. The vermicular forms are large, doubled-up in the red bloodcorpuscles in the circulating blood. Sometimes the two limbs approximately equal in thickness, sometimes one considerably thicker than the other. Nucleus a compact mass of deep, purple granules (Giemsa-stained). The oval

#### SPOROZOA.

forms also contain a nucleus which may be compact or fragmented. Certain large forms are schizonts.

Dimensions.—Vermicular form, length  $12-15\mu$ ; sehizonts,  $20\mu$  in diameter.



Fig. 48.—Hæmogregarina hankini Simond. A, large form with compact nucleus; B, parasite with two nuclei, due to two parasites adhering together; C, oval form, with a number of nuclei arranged on the periphery; D, young parasite with dispersed nucleus; E, larger form with dispersed nucleus (n, nucleus of corpuscle; p, parasite). (After Simond.)

*Remarks.*—According to Simond two forms of the parasite are met with, viz., vermicular and oval. In the young vermicular form the nucleus is compact or fragmented into a number of granules arranged in each limb of the parasite. The parasite is always doubled upon itself, the two limbs being parallel and sometimes in contact with one another. In the oval form also the nucleus may be compact or fragmented. Cells with incompletely developed merozoites were observed in the lung. These schizonts were spherical, and contained 30 to 40 fusiform merozoites without any residue of segmentation. If it be confirmed that schizogony takes place in the lung, the species will have to be transferred to the genus *Hepatozoon*.

Habitat.—Blood of Gavialis gangeticus Gmelin: UNITED PROVINCES, Jumna River; Crocodilus porosus Schneider (?): INDIA (exaet locality not eited); and Crocodilus porosus Schneider: CEYLON, Daduganoya, Veyangoda.

#### 48. Hæmogregarina laverani Simond. (Fig. 49.)

†Hæmogregarina laverani, Simond, 1901 e, pp. 327-31, fig. 2.

- Hæmogregarina laverani, Castellani & Chalmers, 1919, p. 487; Wenyon, 1926, p. 1395.
- *Hæmogregarina laverani*, de Mello, 1932, pp. 1412-4, pl. i, figs. S 1-S 14.

Young stages amœboid, measuring barely  $3\mu$  in diameter, provided with a small nucleus and many small cyanophil granules disposed either in the form of a coil or a horseshoe. Frequently the young stages are in the form of a minute vermicule bent upon itself, the limbs adhering in such a manner that it is difficult to distinguish this form from the amœboid. Full-grown stage large, reniform, containing, besides the nucleus, two large refringent ovoid granules, and occupying a considerable extent of the surface of the corpuscle. The vermicular stage is much smaller than the reniform stage and does not extend beyond half the length of the corpuscle; it possesses a bulging end and a short drawn-out tail, bent round on the swollen portion.



Fig. 49.—Hæmogregarina laverani Simond. A, young amæboid stage with a nucleus; B, young stage in which the cyanophil grains are dispersed in the form of a coil; C, young stage in which the cyanophil nuclear substance is in the form of a horseshoe; D, young vermicular stage; E, full-grown stage folded upon itself and showing two refringent granules. (From do Mello, after Simond.)

*Remarks.*—The chief characteristic of the species is the possession in most of the stages of two refringent granules, which serve to distinguish this species from all others.

Habitat.—Blood of Lissemys punctata granosa (Schoepff): UNITED PROVINCES, Agra.

# 49. Hæmogregarina leschenaultii Robertson. (Fig. 50.)

†Hæmogregarina leschenaultii, Robertson, 1908, pp. 182, 184, figs. 4–7. Hæmogregarina leschenaultii, Wenyon, 1926, pp. 1388.

Hæmogregarine with two free motile forms, only present in the blood: (1) slender free form with dense nucleus; no granules in the cytoplasm; rather actively motile: (2) broad, massive, granular form; less active, period of movement succeeded by period of rest. Two intra-corpuscular forms also present: (1) long recurved form corresponding exactly with the slender free form; causes hypertrophy of the corpuscle; this is always the prevailing type in an infection: (2) broad form with reticulate nucleus, growing to a somewhat larger size; never very numerous even in a good infection.
Dimensions.—Free motile forms, length  $26-28\mu$ ; broad, intra-corpuscular form,  $30\mu$ .

Habitat -Blood of the lizard, Hemidactylus leschenaulti Dum. & Bibr. : CEYLON, Trincomalee.



Fig. 50.-Hæmogregarina leschenaultii Robertson. A, broad intracorpuscular form; B, broad free form; C, slender recurved intra-corpuscular form: D, slender free form. (After Robertson.)

50. Hæmogregarina magna (Grassi & Feletti). (Fig. 51.)

Hæmogregarina ranarum (part), Kruse, 1890, p. 541; Celli & Sanfelice, 1891, p. 504, pl. v. figs. 16-18.

Drepanidium magnum, Grassi & Feletti, 1891, p. 82: 1892, pl. i, fig. 15.

Drepanidium krusci, Labbé, 1892, p. 617.

Danilewskya krusei, Labbé, 1894, p. 127.

Hæmogregarina magna, Labbé, 1899, p. 76. †Danilewskya krusei, Berestneff, 1903, p. 347, pl. viii, figs. 9–13.

Hæmogregarina magna, Patton, 1908, p. 319 (?); Wenyon, 1926. p. 1398.

Adult form oval, very large, may be folded upon itself in the corpuscie.

Habitat.-Blood of Rana tigrina Daud., R. limmocharis Wiegmann: BOMBAY, Bombay; R. tigrina Daud.: MADRAS, Madras.

## 51. Hæmogregarina malabarica de Mello. (Fig. 52.)

†Hæmogregarina malabarica, de Mello, 1932, pp. 1411-25, pl. ii, figs. A-D.

Two forms are present: (1) The vermicular form consists

of a sausage-shaped body with a caudal appendage which is always applied to the body along its concave border. The convex border is bounded by a thin band of protoplasm which



Fig. 51.—Hæmogregarina magna (Grassi & Foletti). A, young intracorpuscular form; B, adult oval form; C, adult form folded upon itself; D, uninfected corpuscle. Uninfected corpuscles are also shown with A, B, and C for the sake of comparison. (After Berestneff.)



Fig. 52.—Hæmogregarina malabarica do Mello. A, free vormicular form; B, intra-corpuscular vermicular form; C, intracorpuscular bean-shaped form; D, cyst, containing eight merozoites; E, F, larger merozoites found in the liver cell. A, B, C as seen in the blood; D, as seen in smears from the liver; E, F, as seen in sections of the liver. (After de Mollo.)

stains a clear blue or sometimes violet with Romanowsky's stain, and is thicker at the pole at which the nucleus is situated, becoming narrower to form the tail; the tail does not extend

to the other pole of the body. The nucleus is nearly always found near the zone of separation of the two parts, and contains irregularly dispersed chromatin particles. The body is more or less filled with metachromatic granules. The parasitized corpuscles become hypertrophied, measuring 20- $25\,\mu$ , whereas the normal corpuscles measure  $16-18\,\mu$ . (2) The reniform or bean-shaped forms are found in very much smaller numbers than the vermicular ones. The nucleus is rounded and central, but may sometimes be situated close to one of the poles; it contains chromatin granules or rods. The cytoplasmic constitution is also different from that of the vermicular form. In one of the poles there are two or more vacuoles; there may be vacuoles, but fewer, in the other pole also. The parasitized corpuscle is hypertrophied, but is enlarged in its width rather than in its length, measuring  $17-18\mu$  by  $14-16\mu$ . Schizogony takes place in the liver, and is of two types, resulting in large and small merozoites. Cysts are found in the liver of the host; they are rounded, with a thick membrane, and contain 8 rounded or falciform merozoites. The free merozoites are generally found in pairs. In sections of the liver the cysts containing 8 merozoites are not encountered, but larger merozoites,  $12-16\mu$  in size, occur singly, or in groups in 2, 4, 8 or larger numbers.

Dimensions.—Vermicular form  $17-22 \mu$ , reniform type  $8-12\mu$ ; cysts, diameter  $18-20\mu$ ; merozoites  $6-8\mu$  or  $12-16\mu$ .

Remarks.—This species is recognizable from H. laverani by the young stages not being ameboid and the vacuoles being inconstant in number and occurring at one or both poles of the reniform stages; the vernicular stage also differs in being larger than the reniform stages, in occupying nearly the whole of the corpuscle, and in having a tail nearly as long as the body of the parasite. The species resembles H. vittatæ in possessing cysts with 8 merozoites, but in H. malabarica the cysts are regularly spherical, whereas in H. vittatæ they are navicular and the reniform stages of that species contain so-called plastids. Schizogony in the lungs or in the blood does not occur in this species, though it is known to take place in others.

The species also somewhat resembles H. nicoriæ, but is clearly marked off from it by the non-existence of a clear pole, the chromatic granules filling completely the opposite pole, the tail being closely applied to the body, the vermicular forms being larger and more abundant than the bean-shaped forms, and by the chromatoid granules filling up the cytoplasm.

Habitat.—Blood, liver, bone-marrow and other viscera of the Indian tortoise, *Lissemys punctata granosa* (Schoepff): PORTUGUESE INDIA, Nova Goa.

## 52. Hæmogregarina mesnili Simond. (Fig. 53.)

†Hæmogregarina mesnili, Simond. 1901 a. pp. 150-2; 1901 e, pp. 322-7, fig. 1. Hæmogregarina mesnili, Castellani & Chalmers, 1919, p. 487; Wenyon, 1926, p. 1395.

The young stages are amæboid. The older stages may be either vermicular with a distinct horn-like prolongation bent upon the main body, oval or reniform, slender and doubled, or folded into three parts. The nucleus may be fragmented or compact.

Dimensions.—In the folded forms the total length of the parasite may exceed  $30\mu$ .



Fig. 53.—Hæmogregarina mesnili Simond. A, young amœboid stage; B, horned vormicular stage, showing two nuclei; C, adult roniform stage with colourless granules; D, vermicular stage, with two equal limbs; E, very long vermicular stage, folded twice over, with three limbs interlacing together: F, fragmentation of a schizont, with free morozoites. (After Simond.)

Remarks.—The young amœboid forms measure  $3-6\mu$  in length and contain a fragmented nucleus. The horned vermicule stage recalls the appearance of *Hæmoproteus metchnikowi*, described by the same author from *Chitra indica*. The reniform individuals are often seen to be filled with refringent colourless granules. The elongate vermicular forms are uniform in diameter throughout the greater part of their length, and are folded into two equal limbs, or in the largest individuals folded twice over, the three limbs interlacing like a figure of 8, a characteristic not met with in other species.

Habitat.—Blood of Kachuga tectum (Gray): UNITED PROVINCES, Jumna River.

53. Hæmogregarina mirabilis Castellani & Willey. (Fig. 54.)

- *†Hæmogregarina mirabilis*, Castellani & Willey, 1904, pp. 86–90, figs. 28–38; Patton, 1908, p. 318.
- Hæmogregarina mirabilis, Dobell, 1908, p. 294.
- †Hæmogregarina mirabilis, Plimmer, 1913, p. 149.
- Hæmogregarina mirabilis, Castellani & Chalmers, 1919, p. 487, fig. 153; Wenyon, 1926, p. 1394.

+Hæmogregarina mirabilis, Scott, 1926, p. 236.

Form elongate, thick, Gregarine-like and bent; cytoplasm

(with Romanowsky's stain) stains uniform blue, leaving no clear pole; the nucleus is stained reddish-blue and is near the anterior pole.



Fig. 54.—Hæmogregarina mirabilis Cast. & Will. A, intra-corpuscular form bent double; B, parasite within the capsule inside a corpuscle : U, D, parasito emerging from the cyst and the corpuscle : E, F, free forms. (After Castellani & Willey.)

Dimensions.—Length about  $12\mu$ .

Remarks.-Castellani and Willey (1904) described the occurrence within the corpuscles of parasites of relatively large size and slightly crescentic or reniform shape, and consisting of a membrane (cytocyst) inside which developed an elongate body (monozoite) with a well-defined nucleus. This organism escaped from the cytocyst and the corpuscle and became a freely motile organism in the blood. The exact stages cannot be determined from their description, but probably they were dealing with the development of the gametes from the gametocytes.

Habitat.-Blood of Tropidonotus asperrimus Bouleng.: CEYLON; and Tropidonotus piscator Schneider: CEYLON (also in specimens from Ceylon in the Zoological Gardens, London): MADRAS, Madras.

#### 54. Hæmogregarina najæ Laveran. (Fig. 55.)

Hæmogregarina najæ, Laveran, 1902, p. 1037, figs. 1-3.

†Hæmogregarina najæ, Patton, 1908, p. 318.

Hæmogregarina najæ, Dobell, 1908, p. 293. †Hæmogregarina najæ, Plimmer, 1912, p. 413; 1913, p. 149; 1914, p. 188; 1916, p. 85.

Hæmogregarina najæ, Castellani & Chalmors, 1919, p. 487; Wenyon, 1926, p. 1110, pl. xix, figs. 8-10; p. 1393.

†Hæmogregarina najæ, Scott, 1926, p. 236; 1927, p. 189.

Intra-corpuscular form elongate, vermicular, rounded at one end and drawn out at the other. When fully developed, bent upon itself. Nucleus oval, more or less elongate, situated about the middle.

Dimensions.—Vermicular form  $14\mu$  in length; fully developed form  $21-22\mu$  by  $3\mu$  at the rounded end.

Remarks.—Schizonts of this species were discovered by Wenyon (1909) in the lungs of Naja hajæ in the Sudan.



Fig. 55.—Hæmogregarina najæ Laveran. A, elengated form; B, recurved form; C, form liberated from the corpuscle. (After Laveran.)

Habitat.—Blood of Naja naja (Linn.): MADRAS, Madras (also in specimens from India in the Zoological Gardens, London).

55. Hæmogregarina nicoriæ Castellani & Willey. (Fig. 56.)

*Hæmogregarina nicoriæ*, Castellani & Willey, 1904, pp. 85-6, figs. 17-27;
 Patton, 1908, p. 319: Robortson, 1908, p. 178; 1910, pp. 741-62, pls. xxxii-xli; Dobell, 1910, p. 68; Alexeioff, 1912, p. 97.
 *Hæmogregarina nicoriæ*, Castellani & Chalmers, 1919, p. 486;

Wenyon, 1926, pp. 1084, 1105, 1395; Reichenew, 1919, p. 486;

Form elongate, Gregarine-like, with one end granular and the other end clear, the central nucleus being a more or less diffuse aggregation of chromatin granules. Sometimes the organism is bent round upon itself. Schizogony takes place in the blood-vessels of the lung and produces nearly 70 merozoites, or in the circulating red blood-corpuseles producing six to eight smaller merozoites which grow into gametocytes.

When the Hæmogregarines are taken together with the blood of the tortoise into the crop of the leech, some of them pass into the intestine and are there found as motile vermicules. They penetrate into the intestinal wall, where the differentiation of the hitherto indistinguishable gametes takes place, culminating in a process suggesting anisogamous conjugation. The zygote breaks up to form eight sporozoites, which pass through the intestinal wall into the blood-spaces. The Hæmogregarine is probably passed into the blood of the tortoise through contamination of the wound by the leech while feeding.

Dimensions.—Length  $12\mu$ .

Remarks.—The life-history of the organism, as worked out by Robertson, confirms in all essential respects that described by Reichenow (1910) for *H. stepanowi*. In *H. nicoriæ* SPOR. K schizogony, producing the large merozoites, occurs free in the blood-vessels of the lung, and that producing the smaller gametocytes in the circulating red blood-corpuscles. In H. stepanowi, on the other hand, schizogony always takes place in the bone-marrow, always inside the blood-corpuscles, and the number of merozoites does not exceed twenty-four.

Christophers once showed Patton some bodies from a leech off *Lissemys punctata granosa* which suggested developmental forms of the Hæmogregarine of the tortoise, but Patton eame to the conclusion that they probably represented some stage in the life-cycle of a *Coccidium* parasitic in the leech.



Fig. 56.—Hæmogregarina nicoriæ Castellani & Willey. A, bean-shaped Hæmogregarine, with circular type of nucleus; B, early stage of schizogony in the blood-stream; C, late stage of schizogony in the lung; D, fully-formed merozoites, only a very few of the total number formed shown in the section; E, free motile stage in the lumen of the intestine of the leech; F, microgametocyte lying closely applied to macrogametocyte; G, the microgametocyte giving rise to the microgamete nuclei, one of which fuses with the nucleus of the macrogamete; H, sporocyst with eight nuclei; I, sporocyst showing sporozoites. (After Robertson.)

Habitat.—Blood of the tortoise Geoemyda trijuga (Schweiger) and in the leech Ozobranchus shipleyi Harding: CEVLON, Colombo; blood of the tortoise Lissemys punctata granosa (Schoepff) and in the transmitting leech: MADRAS, Madras.

#### 56. Hæmogregarina nucleobisecans Shortt. (Fig. 57.)

- *†Hæmogregarina nucleobisecans*, Shortt, 1917, pp. 408–12, pl. xxxi, figs. 1–17.
- Hæmogregarina bisecans, Wenyon, 1926, p. 1397.

*†Hæmogregarina* sp., Donovan (first recorded in Wenyon, 1926, p. 1397).

†Hæmogregarina sp., Wenyon, 1926, p. 1397; Scott, 1926, p. 236.



Fig. 57. -Hæmogregarina nucleobisecans Shortt. A, mature undivided schizont; B, division of schizont; C, cyst, containing merozoites; D, E, single infection of an erythrocyte;
F, double infection of an erythrocyte; G, treble infection of an erythrocyte; I, parasite passing out of ruptured erythrocyte; I, parasite free in plasma. (After Shortt.)

Schizogony only in the liver. The mature schizont broadly oval and lying in a thin-walled cyst closely surrounded by hepatic cells. The cyst grows larger, large number of nuclei are produced, and a correspondingly large number (sometimes as many as 115) of very small merozoites are produced. Each merozoite has a pink-staining, hyaline protoplasm and a well-marked nucleus. Gametocytes intra-corpuscular. sausage-shaped, slightly curved, and lying with concave border applied to the nucleus of the host-cell. Sometimes a gametocyte may lie with its convex border towards the nucleus, or may occupy one end of the corpuscle. Sometimes one or both extremities are effilated and slightly recurved. The gametocytes may vary considerably in size : each is enclosed in a thick eapsule; the cytoplasm stains (with Leishman's stain) an azure-blue or faint pink colour, and contains a large ovoid nucleus, staining a dull crimson. Two or three gametocytes may be found in the same corpuscle. Free forms are found in the plasma, having escaped by rupture of the corpuscle, leaving the empty capsule behind. Sporogony not known. The secondary host may perhaps be a blood-sucking worm, Angiostoma sp., found in the lung of the toad, or a sand-fly.

Dimensions.—Mature schiznot  $16\mu$  by  $7\mu$ ; fully developed cyst 28-30  $\mu$  by 24.5-26  $\mu$ ; merozoite 3  $\mu$  by 1.3  $\mu$ ; gametocytes from  $9.5\mu$  by  $4\mu$  to  $21.8\mu$  by  $4.8\mu$ .

Habitat.-Blood of Bufo melanosticus Schneider: PUNJAB, Ambala ; Delhi ; UNITED PROVINCES, Cawnpore ; also from the same host from India in the Zoological Gardens, London.

#### 57. Hæmogregarina pythonis (Billet).

Danilewskya pythonis, Billet, 1895, p. 30, figs. 1-3.

Hæmogregarina pythonis, Labbé, 1899, p. 76.

+Hæmogregarina pococki, Sambon, 1907, p. 283.

+Hæmogregarina sp., Patton, 1908, p. 318.

Hæmogregarina pococki, Dobell, 1908, p. 293.

Hæmogregarina pythonis, Johnstone, 1912, p. 235. †Hæmogregarina pococki, Plimmer, 1912, p. 412; 1913, p. 148: 1914, p. 189; 1916, p. 85; 1917, p. 32.

†Hamogregarina sp., Phisalix, 1913, pp. 1052-4.

Hæmogregarina pythonis, Castellani & Chalmers, 1919, p. 487.

Hæmogregarina sp., Wenyon, 1926, p. 1393.

†Hæmogregarina pococki, Wenyon, 1926, p. 1393; Scott, 1926, p. 236.

Intra-corpuscular. Body club-shaped, often folded in the red blood-corpuscle. Anterior extremity broader and rounded. Posterior extremity attenuated and recurved. Cytoplasm more or less granular. Nucleus median or nearer posterior extremity, large, oval, and with coarse, deeply staining chromatin granules. Parasite lies parallel or obliquely to long axis of host-cell, of which it occupies about two-thirds. without causing much alteration beyond displacement of nucleus.

Dimensions.-14-16  $\mu$  in length.

Remarks.—Plimmer found the host-cells often deformed and generally diminished in size. Johnstone (1912) regards *H. pococki* Sambon as identical and synonymous with *H. pythonis* (Billet).

Habitat.—Blood of Python molurus (Linn.): MADRAS, Madras; also in specimens from India in the Zoological Gardens, London.

## 58. Hæmogregarina rara Laveran & Mesnil. (Fig. 58.)

\*Hicmogregarina rara, Laveran & Mesnil, 1902, p. 611. Hicmogregarina rara, Castellani & Chahners, 1919, p. 487 : Wenyon, 1926, p. 1395.

Intra-corpuscular or free. The intra-corpuscular form is elongate or curved into the shape of an arc, with one of the extremities rounded and the other more or less attenuate. Sometimes the parasite lies in one part of the corpuscle without displacing its nucleus, at other times it is strongly recurved, and the nucleus of the corpuscle is pushed to one end. When the organism is liberated from the corpuscle it is fusiform,



Fig. 58.– Hæmogregarina rara Laveran & Mesnil. A, elongated; B, curved form; C, form leaving the corpuscle; D, E, free in the plasma. (After Laveran and Mesnil.)

transparent, and motile, and possesses a very elongate and elear nucleus. In the stained preparation (with Leishman's stain) the cytoplasm is of a light blue colour and is finely granular, with large chromatoid bodies. The nucleus is elongate, cylindrical, nearly always swollen at the ends, and is stained deep violet. It occupies about two-thirds of the length of the body. Multiplication appears to be by binary fission. Other stages of development have not as yet been discovered.

Dimensions.—Average length  $15 \mu$  by  $2-3 \mu$  in width.

Habitat.-Blood of the tortoise, Chinemys reevesii (Gray): CEYLON.

- 59. Hæmogregarina rodriguesi de Mello, de Sá, de Sousa, Dias, & Noronha. (Fig. 59.)
  - †Hæmogregarina rodriguesi, de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, pp. 13, 14, pl. i, figs. 8–14; de Mello, 1934 a, p. 1786.
  - †Hæmogregarina proceece, de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, pp. 14, 15, pl. i, figs. 15–17; de Mello, 1934 a. p. 1786.

The parasite occurs in two forms, elongate or shorter and stumpy. In the extra-corpuscular form it is gregariniform and without a capsule. Cytostome stains blue with Leishman's stain, and the nucleus is elongate, rectangular, generally compact, rarely vesicular, occupying a large part of the parasite and situated near one or other extremity. The intra-corpuscular stage has the same appearance, but possesses a large clear capsule containing sometimes one or two vacuoles in the centre. The short and stumpy form is equally broad at both poles, and has a less extensive capsule.



Fig. 59.—Hæmogregarina rodriguesi de Mello. de Sá, de Sousa, Dias, & Noronha. A-D, intra-corpuscular forms; E, free form. (After de Mello and others.)

Remarks.—The parasite is very common, 50 per cent. of the lizards examined being infected. It could not be determined whether schizogony takes place in the blood or in the internal organs. The short stumpy form was originally described as a separate species under the name of H. proence, but de Mello (1934 a) has amalgamated the two.

Habitat.—Blood of Hemidactylus brooki Gray : PORTUGUESE INDIA, Nova Goa.

- 60. Hæmogregarina stepanowiana Laveran & Mesnil. (Fig. 60.)
  - †Hæmogregarina stepanowiana, Laveran & Mesnil. 1902, pp. 609-10. figs. 1-5.

Hæmogregarina stepanowiana, Castellani & Chalmers, 1919, p. 487; Wenyon, 1926, p. 1395.

Intra-corpuscular form oval or reniform, displacing the nucleus when fully developed; cytoplasm finely granular and containing numerous chromatoid particles; nucleus central rounded or oval, placed at right angles to the long axis of the organism, and containing chromatin granules of various sizes. When the parasite doubles upon itself the two portions are not equal, and the nucleus is always situated in the longer part near the level of the bend. Extra-corpuscular form like a vermicule, with one extremity much wider than the other. The intra-corpuscular form, when about to multiply, assumes an oval form, and its nucleus divides repeatedly into eight nuclei ; the protoplasm then divides into as many parts,



Fig. 60.—Hæmogregarina stepanowiana Laveran & Mosnil. A, B. elongated corpuscular forms : C, full-grown parasite doubled upon itself : D, free vermicule-like form : E, form showing nuclear multiplication. (After Laveran and Mesnil.)

the corpuscle breaking up at the same time. Multiplication forms were not found in the blood, but were numerous in smears from the liver.

Dimensions.—Extra-corpuscular form  $18-20 \mu$  by  $5 \mu$ .

Remarks.—According to the authors of the species the young form shows greatest resemblance to the young form of *H. stepanowi* Danilewsky; but the mature form differs markedly from that species. In *H. stepanowi* the parasite becomes doubled exactly about its middle, its nucleus is very elongate, and the nucleus of the corpuscle is displaced, but not hypertrophied. The liberated Hæmogregarine is a vermicule, measuring  $30-40 \mu$  by  $3-4 \mu$ . In *H. stepanowiana* the doubling of the intra-corpuscular parasite is not exact, and the extracorpuscular vermicule is much shorter and wider than in *H. stepanowi*.

Habitat.—Blood of the tortoise, Chinemys reevesii (Gray) CEYLON.

## 61. Hæmogregarina testudinis Laveran & Nattan-Larrier.

*Hamogregarina testudinis*, Laveran & Nattan-Larrier, 1912, pp. 134-6, 10 figs. (Abstract in Wiegmann's Archiv, 1913, p. 411).
 *Hæmocystidium testudinis*, Castellani & Chalmers, 1919, p. 516.
 *Hæmogregarina testudinis*, Wenyon, 1926, p. 1396.

Small, medium, and large intra-corpuscular forms. Free

and multiplication forms also present. Cysts  $12-15\mu$  in length and  $12-14\mu$  in width. Number of merozoites in each cyst 4 to 8 macro- or 12 to 24 microgametes.

Habitat.—Blood of the tortoise, Testudo emys Schleg. & Müll. : INUIA (exact locality not cited).

## 62. Hæmogregarina thomsoni Minchin. (Fig. 61.)

*†Harmogregarina thomsoni*, Minchin, 1907, pp. 1098–1104, pls. lv-lvi, tigs. 1–23.

Hamogregarina sp., Shortt, 1917, pp. 402-8, pl. xxx, figs. 1-21.

Hæmogregarina thomsoni, Castellani & Chalmers, 1919, p. 487; Wenyon, 1926, p. 1388.

Both intra-corpuscular and free forms (so-called vermicules) are met with. Intra-corpuscular parasite always distinctly sausage-shaped, and slightly bowed in the plane of the corpuscle, this normal curvature being due to the parasite being situated to the side of the corpuscular nucleus. In a few cases the curve of the parasite is reversed, its convexity being towards the corpuscular nucleus, which is then much more displaced, and the parasite resembles its free form very closely.

Young intra-corpuscular forms vary in length from about a half to two-thirds of the blood-corpuscle: cytoplasm very clear, and a delicate nucleus, consisting of faintly staining granules and strands of chromatin, forms a band round the waist of the parasite. Full-grown intra-corpuscular parasites are at least three-fourths the length of the blood-corpuscle; cytoplasm finely granular, nucleus exceedingly rich in chromatin, forming a deeply staining mass of irregularly spongy texture, occupying the middle region of the body for its whole width and nearly one-third of its length. The free vermicules closely resemble the full-grown intracorpuscular forms, long, medium, and stumpy forms being distinguishable.

Sometimes most of the intra-corpuscular parasites have the form of an elongated sausage, slender and drawn out ; cytoplasm clear and free from coarse granulations ; nucleus forming a band or zone at the middle of the body, equal in width to nearly half the length of the body, and with chromatin arranged in the form of transverse strands.

Small forms are generally situated at one end of the erythrocyte; larger forms lie in the long axis of the host-cell, and displace the nucleus laterally. The parasite is usually situated with its concave border embracing the convexity of the erythrocyte nucleus, but its position may be reversed. Invasion by two or more forms is not uncommon, the parasites lying parallel to one another and both on the same side, or the two embracing the nucleus between them. Infected cells, especially those with two parasites, are increased in size.

Schizogony takes place in the endothelial cells of the capillaries of the lung, and is of two forms. One form of schizont produces a smaller cyst, with larger and less numerous merozoites, and the other form produces a larger cyst, with smaller and more numerous merozoites. In the former case the schizont in the endothelial cell is broadly oval, and usually forms a smaller cyst giving rise to 16 merozoites. The larger cysts are more rare, and may contain as many as 40 merozoites. What the two kinds of merozoites develop into has not been determined. The process probably differs from that described



Fig. 61.--Hæmogregarina thomsoni Minchin. A, small intra-corpuscular form; B, large intra-corpuscular form showing the so-called "capsule"; C, a large and an intermediate-sized form in the samo corpuscle; D, two large individuals lying side by side, both on ono side of the nucleus; E, two large individuals embracing the nucleus between them; F, the free form, (After Minchin.)

by Reichenow and Robertson for the Hæmogregarines of cold-blooded Vertebrates.

Dimensions.—Intra-corpuscular forms, young 9–11  $\mu$  in length, full-grown 15–17  $\mu$  by 5  $\mu$ ; schizonts 15  $\mu$  by 6  $\mu$ ; cysts, small 21.5 $\mu$  by 14.5 $\mu$ , large 25  $\mu$  by 15  $\mu$ .

Habitat.-Blood of the lizard, Agama tuberculata Gray: PUNJAB, Kasauli.

#### SPOROZOA.

### 63. Hæmogregarina thyrsoideæ de Mello & Vales. (Fig. 62.)

*†Hæmogregarina thyrsoidea*, de Mello & Vales, 1936, pp. 403-4, pl. xxxv.

Youngest form ovoid, with conspicuous nucleus and cytoplasm stained light blue with Romanowsky's stain. The fullgrown parasite shows irregular vacuolization of the cytoplasm. The nucleus is generally central, often divided into two more or less irregular masses, with indistinct nuclear membrane,



Fig. 62.—Hæmogregarina thyrsoideæ de Mello & Vales. A, young form; B, normal full-grown form; C, form with contral nucleus, containing two chromatin granules; D, form with one nuclear mass at each pole. (After de Mello and Vales.)

and minute volutin granules. Sometimes the chromatin is reduced to two very minute centrally situated granules : or the nucleus is situated at one of the poles and composed of two masses, or the two nuclear masses are situated one at each pole.

Habitat.—Blood of the freshwater eel, Thyrsoidea macrurus (Bleeker) : PORTUGUESE INDIA, Nova Goa.

#### 64. Hæmogregarina triedri Robertson. (Fig. 63.)

*†Hæmogregarina triedri*, Robertson, 1908, pp. 181, 184, figs. 10, 11. *Hæmogregarina tiedri*, Wenyon, 1926, p. 1388.



Fig. 63.—Hæmogregarina triedri Robertson. A, broader bean-shaped form; B, long form. (After Robertson.)

Hæmogregarine with double capsule, a delicate inner capsule and a loose outer one, with tendency to stain very deeply with Giemsa's stain. The inner capsule has an opercular lid at one end. Two forms present: (1) broad, rather bean-shaped form; (2) long, slightly recurved specimens, with elongate nuclei. Both forms frequently show at one end an irregularly shaped body which stains a bright red with Giemsa's stain. The young forms have no capsule. Transmitting host not known.

Dimensions.—13–15  $\mu$  in length.

Habitat.—Blood of the lizard. Hemidactylus triedrus (Daudin): CEYLON, Trincomalee.

## 65. Hæmogregarina vlttatæ Robertson. (Fig. 64.)

- †Hæmogregarina vittatæ, Robertson, 1908, pp. 179, 180, 183, figs. 2–3. Hæmogregarina vittatæ, Castellani & Chalmers, 1919, p. 487, figs. 149, 157, 158.
- *†Harmoyregarina vittata*, Donovan (first recorded in Wenyon, 1926, p. 1395).
- *Harmogregarina vittata*, de Mello, 1932, pp. 1417–18, pl. i, figs. R1, R2.

Hæmogregarine infection associated almost invariably with Trypanosoma vittatæ. Two forms present: (1) broad massive form; (2) recurved form with pale cytoplasm, the two limbs



Fig. 64.—Hamogregarina vittatæ Robertson. A, elongate, recurved form; B, broad massive form. (After de Mello.)

being equally long; nucleus dense. Broad form shows reticu late dense cytoplasm and rather delicate loose nucleus; the larger forms have two red-staining plastid (?)-like bodies at one end. Schizogony occurs in the spleen and liver; eight merozoites are formed; these are enclosed in pairs in a delicate boat-shaped capsule. Transmitting host probably *Glossiphonia*.

Dimensions.—Recurved form  $22-26 \mu$  in total length.

Habitat.—Blood of the tortoise, Lissemys punctata granosa (Schoepff.): CEYLON; PORTUGUESE INDIA, NOVA GOA.

## 66. Hæmogregarina xavieri de Mello. (Fig. 65.)

†Hæmogregarina xavieri, de Mello, 1932, pp. 1426-27, pls. iii & iv.

Two forms present: (1) Broadly oval or sometimes reniform, occupying, according to the state of its development, a greater or less extent of the corpuscle. Cytoplasm alveolar, containing generally two, sometimes more, rounded vacuoles, usually confined to the polar regions. Nucleus central, containing granules or rods of chromatin in compact masses or variously dispersed. The parasitized corpuscles are generally not altered, sometimes reduced or enlarged in size, and the nucleus of the corpuscle is displaced to one side. (2) Elongate or vermicular forms, sometimes enclosed in a fine, membranous capsule or entirely without a capsule, provided with a tail which stands out from the body, and does not exceed onequarter or one-third of its length. These tailed forms are much less numerous than the oval forms. One pole sometimes presents one or two vacuoles, but generally the cytoplasm alveolar, and the vacuoles not clearly indicated. Nucleus central, and the chromatin showing the same disposition as in the oval forms. The metachromatic granules generally abundantly distributed in the tail, and rarely in the anterior pole as well. Forms intermediate between the oval and the vermicular are also met with, and sometimes two individuals are seen in the same corpuscie.

Schizogony takes place in the lungs, spleen, liver, etc. Cysts found in smears from the lungs are oval, and contain 2 or 3 schizonts; those found in smears from the spleen are rounded or oval, and contain 6 or 7 schizonts; while those in smears from the liver are circular or more or less oval, and contain 2 or 3 schizonts. In sections of parasitized organs oval cysts are found to contain 6 or 7 schizonts (merozoites) similar to those met with in the smears. Fusiform merozoites are seen in the blood-stream and in the intercellular spaces.

Dimensions.—Oval form  $7.5-13.5\mu$  in length by  $2.5-6.2\mu$ in width; vermicular form  $9-10\mu$  in length by  $4\mu$  in width; cysts from lungs  $8-10\mu$  by  $4-5\mu$ , from liver, spherical,  $15\mu$  in diameter, or oval,  $8.5\mu$  by  $5\mu$  or  $10\mu$  by  $7.5\mu$ .

Remarks.—The species is morphologically distinct from H. laverani Simond, described from the same host at Agra. Both H. malabarica and H. xavieri do not show any amœboid young stages, nor do they show the two refringent granules so characteristic of H. laverani. H. xavieri differs from H. malabarica in the tail being considerably shorter, and not being closely applied to the body, the nucleus being central in the elongate forms, and in the dimensions being considerably smaller in both elongate and oval forms than in the other species.







Fig. 65.—Hæmogregarina xavieri de Mello. A. In blood. A. B. oval forms; C. kidney-shaped form; D. elongate form with a short tail; E. elongate form within a capusle.
B. A. cyst containing few schizonts; B. cyst containing

B. A. cyst containing few schizonts; B. cyst containing six schizonts; C, cyst containing six schizonts (merozoites); D, E, fusiform merozoites in the blood; A. B, C, in smears from lungs, spleen, and liver respectively; D, E, in section of a blood-vessel. (After de Mello.)

Α.

#### SPOROZOA.

Habitat .- Blood, lungs, spleen, and liver of the tortoise, Lissemys punctata granosa (Schoepff): PORTUGUESE INDIA, Nova Goa.

In addition to the species that are recorded above, a number of Hæmogregarines have been observed in a variety of hosts; but, unfortunately, the published descriptions are so meagre that it is impossible to refer the forms to any known species. For the sake of completeness I have given these records below.

#### 67. Hæmogregarina sp.

*†Hæmogregarina* sp., Plimmer, 1914, p. 189. Hæmogregarina sp., Wenyon, 1926, p. 1403.

Habitat.—Blood of the fish, Trichogaster fasciatus Schneider: from INDIA, in the Zoological Gardens, London.

### 68. Hæmogregarina sp.

†Hæmogregarina sp., Simond, 1901 e, p. 320; Plimmer, 1912, p. 412: 1914, p. 189.

Hæmogregarina sp., Wenyon, 1926, p. 1390.

Large; of ordinary type. Nucleus of parasitized cell divided by the parasite into two parts with a connecting thread. Schizogony in lungs.

Remarks.-This form may be the same as H. varani Laveran, 1905. described from Varanus niloticus.

Habitat.-Blood of the lizard, Varanus monitor (Linn.): INDIA; also from INDIA, in the Zoological Gardens, London.

#### 69. Hæmogregarina sp.

Hæmogregarina sp., Dobell, 1908, p. 292. †Hæmogregarina sp., Patton, 1908, p. 318; Plimmer, 1913, p. 149. Hæmogregarina sp., Wenyon, 1926, p. 1391.

Both short and long forms present.

Habitat.-Blood of Bungarus cæruleus (Schneid.) (=Bungarus candidus): MADRAS, Madras; also from INDIA, in the Zoological Gardens, London.

## 70. Hæmogregarina sp.

*†Hæmogregarina* sp., Robertson, 1908, p. 182, fig. 12.

Hæmogregarina sp., Pobertson, 1908, p. 182, hg. 12. Hæmogregarina sp., Dobell, 1908, p. 294. †Hæmogregarina sp., Patton, 1909, pp. 149, 152; Dobell, 1910, p. 70; Plimmer, 1912, p. 413; 1913, p. 149; 1914, p. 188. Hæmogregarina sp., Wenyon, 1926, pp. 1391, 1394.

Young intra-corpuscular form without a capsule. Full-grown form, with a very marked capsule thickened at both ends, and showing a deep red staining area at either end and a delicate nucleus. Free motile form also without a capsule.

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*Remarks.*—Five species are known from various species of *Zamenis* from other parts of the world, and the forms described by Robertson and Dobell and noted by Patton and Plimmer may belong to one of them.

Habitat.—Blood of the snakes, Chrysopelea ornata (Shaw): CEYLON; and Zaocys mucosus (Linn.): CEYLON, Colombo, Paradeniya; MADRAS, Madras; also in same host from INDIA, in the Zoological Gardens, London; in the linguatilid, Porocephalus pattoni Stephens, from the lung of Zaocys mucosus (Linn.): MADRAS, Madras.

#### 71. Hæmogregarina sp.

<sup>†</sup>*Hixmogregarina* sp., Plimmer, 1914, p. 189. *Hxmogregarina* sp., Wenyon, 1926, p. 1391.

Habitat.—Blood of the snake, Coluber helena Daud., from CEVLON, in the Zoological Gardens, London.

#### 72. Hæmogregarina sp.

*†Hæmogregarine*, Simond, 1901 e, p. 320. *Hæmogregarina* sp., Dobell, 1908, p. 293; Wenyon, 1926, p. 1391.

Habitat.—Blood of a snake, Coluber sp. : INDIA.

## 73. Hæmogregarina sp.

†Hæmogregarina sp., Dobell, 1910, p. 70.

Habitat.-Blood of the snake, Dipsadomorphus forstenii (Duméril & Bibron): CEVLON, Colombo.

#### 74. Hæmogregarina sp.

†Hæmogregarina sp., Dobell, 1910, p. 70.

Habitat.—Blood of the snake, Dipsadomorphus ceylonensis Gunther: CEYLON, Paradeniya.

#### 75. Hæmogregarina sp.

†Hæmogregarina sp., Patton, 1908, p. 318.

Hæmogregarina sp., Dobell, 1908, p. 293; Wenyon, 1926, p. 1391.

Habitat.—Blood of the snake, Dendrophis pictus Gmelin: MADRAS, Madras.

#### 76. Hæmogregarina sp.

*†Hæmogregarina* sp., Patton, 1908, p. 318. *Hæmogregarina* sp., Dobell, 1908, p. 293. *†Hæmogregarina* sp., Dobell, 1910, p. 70. *Hæmogregarina* sp., Wenyon, 1926, p. 1392.

Habitat.—Blood of the snake, Dryophis mycterizans (Daud.): MADRAS, Madras; CEYLON, Colombo, Paradeniya.

#### 77. Hæmogregarina sp.

*†Hæmogregarina* sp., Patton, 1908, p. 318. *Hæmogregarina* sp., Dobell, 1908, p. 293. *†Hæmogregarina* sp., Plimmer, 1912, p. 414; 1913, p. 149; Scott. 1926, p. 236. *Hæmogregarina* sp., Wenyon, 1926, p. 1392.

Habitat.—Blood of the snake, Eryx johnii (Russ.): MADRAS, Madras; also from INDIA, in the Zoological Gardens, London.

#### 78. Hæmogregarina sp.

*†Hxmogregarina* sp., Plimmer, 1913, p. 149 : Scott, 1926, p. 236. *H:emogregarina* sp., Wenyon, 1926, p. 1393.

Habitat.—Blood of the snake, Naja bungarus Schleg., from INDIA, in the Zoological Gardens, London.

#### 79. Hæmogregarina sp.

*Haemogregarina* sp., Simond. 1901 c, p. 320; Robertson, 1908, p. 182.

Hæmogregarina sp., Doboll, 1908, p. 293.

*†Hiemogregarina* sp., Dobell, 1910, p. 70. *Hæmogregarina* sp., Wenyon, 1926, p. 1393.

Remarks.—The form probably belongs to H. najæ Laveran.

described from the same host. Habitat.—Blood of the snake, Naja naja var. atra (Cantor):

INDIA; blood of Naja naja (Linn.): CEYLON, Paradeniya.

#### 80. Hæmogregarina sp.

*†Hæmogregarina* sp., Simond, 1901 e, p. 320. Hæmogregarina sp., Wenyon, 1926, p. 1393.

Habitat.-Blood of a snake, Naja sp. : INDIA.

### 81. Hæmogregarina sp.

†Hæmogregarina sp., Robertson, 1908, pp. 182, 183. Hæmogregarine sp., Wenyon, 1926, p. 1393.

Hæmogregarine showed extraordinary activity. The free form moved with a rapid swimming motion, and entered a blood-corpuscle by simply piercing it, swimming round between the nucleus and the corpuscle wall, and bursting the corpuscle by curling and then suddenly straightening itself. The process takes a few seconds. It also injures the corpuscles, which it touches in passing, the corpuscle losing all its hæmoglobin immediately. The intra-corpuscular stage very closely resembles the free form.

*Remarks.*—Possibly the form examined by Robertson may have been the same as *H. pythonis* (Billet).

Habitat.-Blood of a snake, Python sp.: CEYLON.

#### 82. Hæmogregarina sp.

Laverania sp., Billet, 1895, p. 31. Hæmogregarina sp., Labbé, 1899, p. 77. †Hæmogregarina sp., Patton, 1908, p. 318. "Danilewskya"=Hæmogregarina sp., Dobell, 1908, p. 294. +Hæmogregarina sp., Dobell, 1910, p. 70. Hæmogregarina sp., Wenyon, 1926, p. 1394.

Remarks.—Dobell (1910) thinks that it is probably the same as "Danilewskya" described in this host from Tong-king by Billet (1895) (=Hæmogregarina sp., Labbé).

Habitat.-Blood of Tropidonotus stolatus (Linn.): MADRAS, Madras : CEYLON, Paradeniva.

#### 83. Hæmogregarina sp.

†Hæmogregarina sp., Patton, 1908, p. 318.

Hæmagregarina sp., Dobell, 1908, p. 294.

*†Hæmogregarina* sp., Plimmer, 1912, p. 414; 1913, p. 149; 1915, p. 130.

Hæmogregarina sp., Wenyon, 1926, p. 1394.

Habitat.-Blood of Russell's Viper, Vipera russellii (Shaw) : MADRAS, Madras; also from India in the Zoological Gardens. London.

### Genus HEPATOZOON Miller, 1908.

Leucocytozoon, James, 1905 a, pp. 1-12; 1905 b, p. 1361; Bentley, 1905, p. 988.

Hæmogregarina, Balfour, 1905 a, p. 240; 1905 b, p. 1330.

Hæmogregarine, Laveran, 1905, p. 295.

Leucocytozoa, Patton, 1906, pp. 1-13; 1908, p. 319.

Leucocytozoon, J. R. Adie, 1906, p. 325; Christophers, 1906, pp. 1-16; 1907, pp. 1-12.

Hepatozoon, Miller, 1908, pp. 1-48,

Leucocytozoon, Porter, 1908, pp. 703-16.

Leucocytogregarina, Porter, 1909, p. 264

Hæmogregarina, Wenyon, 1910, pp. 70-1; 1911, pp. 324-32. Leucocytozoon, Christophers, 1912, pp. 37-44. Hepatozoon, Minchin, 1912, pp. 372, 376.

Leucocytogregarina, Sangiorgi, 1912, pp. 287–93; de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, p. 13.

Hepatozoon, Wenyon, 1926, pp. 1085-6; Reichenow, 1929, pp. 920-3; Kudo, 1931, p. 281; Calkins, 1933, pp. 361, 527.

Schizogony takes place in cells of the internal organs (bonemarrow, liver, spleen, kidney) of Vertebrates. After several generations of merozoites have been produced some of them enter erythrocytes or leucocytes and develop into gametocytes. These are taken into the body of a blood-sucking Arthropod (tick, mite, louse, etc.), and the micro- and macrogametes develop and unite in pairs. The zygote becomes encysted in the occyst, which increases enormously in size, eventually producing sporoblasts, sporocysts, and sporozoites. Each large occyst SPOR. L

contains numerous sporocysts, all of which produce numerous sporozoites.

The genus was founded by Miller for a parasite of the leucocytes of rats, which had been previously referred to the genera *Hæmogregarina* or *Leucocytozoon*.

## 84. Hepatozoon adiei Hoare. (Fig. 66.)

†Hepatozoon adiei, Hoare, 1924. pp. 63-6, 1 pl.

Hepatozoon adiei, Wenyon, 1926, pp. 1086, 1095, fig. 456; Roichenow, 1929, p. 923.

Schizonts more or less regularly ovoid. Nuclear multiplication gives rise to 12 to 24 nuclei. Merozoites elongate and verniform. Younger schizonts enclosed in definite cells,



Fig. 66.—Hepatozoon adiei Hoare. A, B, ovoid schizonts; C, schizont showing formation of merozoites; D, rod-like gametocyte enclosed in a leucocyte. A-C, in the smears of the lung; D, in the peripheral blood. (After Hoare.)

probably endothelial cells of the capillaries. In later stages, host-cell reduced to a thin membrane with the flattened nucleus at one side of it. Gametocytes rod-like, rounded at both extremities; nucleus usually terminal in position. The gametocyte either displaces or is wedged in the leucocyte nucleus.

Dimensions.—Schizonts from  $8\mu$  by  $4\cdot 8\mu$  to  $20\cdot 8\mu$  by  $11\cdot 2\mu$ ; gametocytes  $8\cdot 5\mu$  by  $4\cdot 5\mu$ .

*Remarks.*—Only dried films of the blood and smears of the internal organs of the bird were available for study. The schizonts were found in smears of the lung, and the gametocytes in the leucocytes of the peripheral blood. The schizonts and

the gametocytes closely resemble H. can is and H. muris. Nothing is known of the sexual cycle in the Invertebrate.

Habitat.—Blood and smears from the lung of an Indian eagle (not identified) : PUNJAB, Kasauli.

### 85. Hepatozoon canis (James). (Fig. 67.)

*Leucocytozoon canis*, James, 1905, pp. 1–12, 1 pl.; Bentley, 1905,
 p. 988; Christophers, 1906, pp. 1–16, 1 pl.; 1907, pp. 1–12, 1 pl.

†Leucocytozoon bentleyi, James, 1905 a, p. 1361.

Leucocytogregarina rotundata, Patton, 1910, p. 280.

Hæmogregarina canis adusti, Nuttal, 1910, p. 108.

- Hæmogregarina canis, Wenyon, 1911, pp. 324-32, text-figs. 4-8, pl. xvi, figs. 1-17.
- Leucocytozoon canis, Christophers, 1912, pp. 37-44, 1 diagram, text-figs. 4-8.

Hepatozoon canis. Minchin, 1912, p. 377.

Hæmogregarina canis, Castellani & Chalmers, 1919, p. 483.

†Hepatozoon canis, Rau, 1925, p. 293.

†Hiemogregarina canis, Rau, 1926, p. 244.

Hepatozoon canis, Wenyon. 1926, pp. 1091, 1092, fig. 455; pl. xix, figs. 3, 4; pp. 1355-7; Reichenow, 1929, pp. 921, 922, figs. 885, 886.

Schizogony occurs in the spleen, bone-marrow or liver. Schizonts of several types; some produce a small number (generally three) of large merozoites, which become schizonts again, whilst others produce a large number of small merozoites. These latter enter the mononuclear or polymorphonuclear leucocytes and become gametocytes. Free vermicules found in the gut of the infected ticks.

The gametocytes undergo development in the common dog-tick, *Rhipicephalus sanguineus* Koch, in the tissues of which are eventually produced very large oöcysts, containing from 30 to 50 sporocysts, each containing about 16 sporozoites and a residual body. After the sporocysts are fully developed, the oöcyst appears to break up and sporozoites are found scattered among the tissues of the tick. Infection of the dog is probably produced by the ticks being eaten.

Dimensions.—Gametocytes  $8-10 \mu$  by  $4-5 \mu$ , nucleus  $2-3 \mu$ ; oöcysts up to  $100 \mu$ ; sporocysts  $15-16 \mu$  in length; sporozoites  $14-15 \mu$  in length.

*Remarks.*—Rau (1925) infected dogs by injection of spleen material containing schizonts, as also by the injection of the tissues of infected ticks. Parasites appeared in the blood two or three weeks after inoculation and infection progressed in intensity, bringing about the death of some of the animals. Wenyon, however, doubts if other causes of death were excluded.

Habitat.—Internal organs and leucocytes of the Indian wild dog, Cyon dukhunensis (Sykes): ASSAM, Borjulie, Gauhati,

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Fig. 67.—Hepatozoon canis (James). A, parasite in a leucocyte of the dog; B, free vermicule in the blood of the dog; C, D, schizogony in the bone-marrow of the dog; E, gametocytes lying side by side; F, G, formation of the microgametes; H, zygote showing a number of nuclei; I, section of oöcyst in which sporoblast formation is taking place; J, section of fully developed oöcyst in which the sporocytes, have formed. A-G in the dog; H-J in the tissues of the tick. (A-I, after Christophers; J, after Wenyon.)

Tejpur, Nowgong; the dog, Canis familiaris (Shaw): MADRAS, Madras; the jackal, Canis aureus Linn.: MADRAS, Madras; the fox, Vulpes bengalensis (Shaw): MADRAS, Madras. Also the tick, Rhipicephalus sanguineus Koch: MADRAS, Madras.

### 86. Hepatozoon felis domestici (Patton).

†Leucocytozoon felis domestici, Patton, 1908, p. 319. Hæmogregarina felis, Castellani & Chalmers, 1919, p. 486. Hepatozoon felis, Wenyon, 1926, pp. 1085, 1355.

Remarks.—Patton (1908) merely recorded this as a new species, without giving any description. It is, therefore, not certain that this is morphologically distinct from H. canis.

Habitat.-Leucocytes of the cat, Felis sp. : INDIA.

## 87. Hepatozoon funambuli (Patton). (Fig. 68.)

Leucocytozoon funambuli, Patton, 1906, pp. 1-13, pl. i; 1908, p. 319. Hepatozoon funambuli, Minchin, 1912, p. 377. Hamogregarina funambuli, Castellani & Chalmers, 1919, p. 486. Hepatozoon funambuli, Wenyon, 1926, pp. 1085, 1358.

The gametocyte has an elongate oval body, with one end larger and rounder than the other. It lies in the large mononuclear leucocyte, and shows slow vermicular movements. The narrow end shows a distinct bend upwards, simulating a tail. In escaping from the host-cell the more rounded end protrudes first. The free vermicules are elongated and spindle-shaped; they are very active, performing rapid serpentine movements, twisting and curling about. In preparations stained with Romanowsky's stain cytoplasm stains light blue, somewhat darker at the two ends. There is no capsule round the parasite. but by prolonged staining a faint pink outline is seen round it. Nucleus large, irregularly quadrilateral, lying about the centre, staining more deeply but not uniformly. A number of large chromatic dots are seen in the cytoplasm, more in one extremity than in the other. Sometimes two parasites are seen in the same leucocyte, lying elose to each other, between the two detached parts of the nucleus, or one on either side of the nucleus. The free vermicule contains a rounded or oval nucleus.

Dimensions.—Intracorpuscular forms  $10\mu$  by  $5\mu$  in the widest part; free vermicules  $13-14\mu$  by  $3-4\mu$ .

*Remarks.*—The parasites were present in the peripheral blood and in smears from the spleen and liver. Vermicules were also seen from the mid-gut and the body-cavity of the louse, *Hæmatopinus*, but no developmental forms were met with. Habitat.—Leucocytes of the squirrel, Funambulus pennantii Wroughton, and the body of the louse, Hæmatopinus sp.: BOMBAY, Kathiawar.



Fig. 68.—Hepatozoon funambuli (Patton). A, two leucocytes, showing two parasites in each; B, three free forms, showing chromatic particles; C, four free parasitos from spleen-smears; D, free attonuate form from peripheral blood; E, free vernicule from the peripheral blood; F, vernicule from the mid-gut of the squirrel-louse; G, large vernicule from the body-cavity of the louse. (After Patton.)

#### 88. Hepatozoon gerbilli (Christophers). (Fig. 69.)

*Hæmogregarina gerbilli*, Christophers, 1905, pp. 1-15, 1 pl. *Hæmogregarina gerbilli*, Patton, 1906, p. 3. *Hepatozoon gerbilli*, Minchin, 1912, pp. 376, 377. *Hæmogregarina gerbilli*, Castellani & Chalmers, 1919, p. 485. *Hepatozoon gerbilli*, Wenyon, 1926, pp. 1085, 1358; Reichenow, 1929, p. 922.

Stages of the schizogonous multiplication are not known. Gametocytes are found in the red blood-corpuscles and are marked by a short hook-shaped projection at their hinder end. Nucleus close to the bent end. Active vermicule stage free in the plasma. Further development takes place in the louse, *Hæmatopinus stephensi*. In the alimentary canal of the louse free worm-like parasites are recognized. In the body-cavity are found large oöcysts in various stages of development. The fully developed oöcysts contain numerous oval sporocysts; each sporocyst contains 6 to 8 sporozoites and a residual body.



Fig. 69.—Hepatozoon gerbilli (Christophers). A, forms in the red blood-corpuscles; B, fully matured oöcyst; C, sporocyst containing sporozoites. (After Christophers.)

Dimensions.—Oöcysts  $350 \mu$  in diameter.

*Remarks.*—In imperfectly stained films the general appearance of the parasite is very like that of the malarial crescents, except that the pigment is absent. This is due to the "tail" not being differentiated from the body.

Habitat.—Red blood-corpuscles of the Indian gerbil, Tatera indica (Hardw.) (=Gerbillus indicus), and in the mid-gut and the pyloric ampulla of the louse, Hæmatopinus stephensi Christophers & Newstead : INDIA (locality not cited).



Fig. 70.—Life-cycle of Hepatozoon muris (Balfour). A, sporozoites escaping from sporocysts in the intestine of rat; B, sporozoites penetrating through intestinal epithelium and entering into the blood-vessels of the villi; C, sporozoites passing from blood-vessels into liver-cells; D-G, schizogony in livercells; H, merozoites escaping from liver-cells and entering other liver-cells to repeat schizogony; I, merozoites (gametocytes) leaving liver-cells to enter blood-vessels; J, gametocytes in mononuclear cells of blood; K, gametocytes

#### 89. Hepatozoon leporis (Patton).

†Leucocytozoon leporis, Patton, 1908, p. 319. Hepatozoon leporis, Wenyon, 1926, p. 1359.

Patton (1908) merely recorded this as a new species, without giving any description.

Habitat.—Blood of the black-naped hare, Lepus nigricollis (Cuvier): MADRAS, Madras.

### 90. Hepatozoon muris (Balfour). (Fig. 70.)

Leucocytozoon muris, Balfour, 1905, pp. 110-11, pl. xi.

†Leucocytozoon ratti, Adie, 1906, pp. 325-6, text-figs.

Hepatozoon perniciosum, Miller, 1908, pp. 1-48, pls.

Hepatozoon muris, Minchin, 1912, pp. 376, 377.

Hæmogregarine ratti, Castellani & Chalmers, 1919, p. 486.

Hepatozoon muris, Wenyon, 1926, pp. 1085, 1086-90, fig. 453, pl. xix, figs. 1, 2.

+Hepatozoon muris, Donovan (recorded in Wonyon, 1926, p. 1360).

Hepatozoon muris, Reichenow, 1929, pp. 920-1; Kudo, 1931, p. 282, fig. 119, k.

Schizogony takes place in the liver-cells of a rat. Smallest schizont is a spherical, uninucleate body. The schizont increases in size, and the nuclei multiply by repeated division till there are 12 to 20. The full-grown schizont is surrounded by a delicate cyst-wall. Merozoites are budded off, enter other cells, and repeat schizogony. After a time the merozoites become young gametocytes, enter the blood-vessels, invade the mononuclear leucocytes, and appear as Hæmogregarines. When the blood is sucked by the mite, Lælaps echidninus, the Hæmogregarines are liberated from the leucocytes and escape from the enclosing cysts. The gametocytes associate in pairs, each becoming flattened to produce an elongated body, with pointed extremities. Complete fusion of the two gametocytes is said to take place. Before actual union takes place the macrogametocyte increases somewhat in size and encloses the smaller microgametocyte. After fertilization the zygote elongates and becomes a motile oökinete; this moves about in the stomach-contents, increasing in size. Later it penetrates through the intestinal wall and settles down in the surrounding tissue, becoming spherical and growing,

escaping from mononuclear cells in the stomach of the mite; L-O, syngamy and penetration of intestinal wall by zygote ( $\ddot{o}$  kinete); P-S, growth of zygote (sporont) in  $\ddot{o}$  cover in tissues of mite; T, surface of sporont, showing sporoblast formation by budding; U, portion of occyst containing sporoblasts V, portion of occyst containing sporoblasts; V, portion of occyst containing sporoblasts with multiplying nuclei; W, portion of occyst containing sporocysts in each of which are a number of sporozoites. The mite is eaten by the rat, in the intestine of which the sporozoites escape from the sporocysts. (From Wenyon, after Miller.)

It acquires a cyst-wall and grows still further to become the ripe oöcyst. Sporogony takes place in the body-cavity of the mite. Nuclear multiplication and formation of sporoblasts and sporocysts take place, each spore containing 12 to 24 sporozoites. Re-infection takes place by a rat devouring infected mites.

Dimensions.—Schizont, young  $10 \mu$ , full-grown  $30-35 \mu$  by  $25-28 \mu$ : oökinete, young  $25 \mu$  by  $10 \mu$ , full-grown  $50 \mu$  by  $25 \mu$ ; oöcyst  $200-250 \mu$ .

*Remarks.*—According to Miller (1908) and the Japanese workers Kusama, Kasai, and Kobayashi (1919), complete fusion of the two gametocytes takes place, and no formation of micro- or macrogametes is described. Wenyon (1926), however, from what is known of other Hæmogregarines, such as *H. stepanowi*, thinks it probable that the microgametocyte produces microgametes, and that a fertilization of the *Adelea* type occurs instead of the complete fusion of the two gametocytes.

Miller succeeded in infecting rats by placing on them infected mites, and also by contaminating food with crushed nuites, and as injection of crushed infected mites into the peritoneal cavity did not produce infection, it is obvious that infection takes place by way of the alimentary canal.

Habitat.—Leucocytes of the brown rat, Rattus norvegicus (Berkenhout) (=Mus decumanus=Mus norvegicus): PUNJAB; Rattus rufescens (Gray) (=Mus rufescens); and the rat-mite. Lælaps echidninus Berlese: INDIA.

#### 91. Hepatozoon sp.

*†Hepatozoon* sp., Donovan (recorded in Wenyon, 1926, p. 1362).

Habitat.—Blood of the flying squirrel, Pteromys petaurista Pallas : INDIA.

#### Genus KARYOLYSUS Labbé, 1894.

Hæmogregarina (part), Danilewsky, 1886.

Karyolysus, Labbé. 1894, p. 109; 1899, p. 75; Minchin. 1903, pp. 261, 265, 266; 1912, p. 372; Reichenow, 1913, pp. 317-63; 1921, pp. 179-291; Wenyon, 1926, pp. 1095-101; Reichenow, 1929, pp. 865, 916-19; de Mello & de Meyrelles, 1937, pp. 119-41.

Sporogony in the epithelial cells of the intestine of the Invertebrate host, a mite, produces an oöcyst with a number of sporoblasts, which escape from the oöcyst as motile vermicules (sporokinetes) and enter the egg of the mite, where they secrete sporocysts within which sporozoites are developed. The mite hatched from the egg has the sporocysts in its intestinal epithelium. The sporocysts enter the intestinal

#### KARYOLYSUS.

epithelium of the Vertebrate host, a lizard, through the ingestion of the fæces of the nymph or of the nymph itself. The sporozoites pass to the blood-vessels and penetrate the endothelial cells, where schizogony takes place. Certain merozoites enter the red blood-corpuscles as gametocytes and appear as Hæmogregarines. The gametocytes are taken up by the mite, in the gut of which gametogony takes place and the oöcyst is formed.

*Remarks.*—The genus owes its names to the fact that the nucleus of the host-cell is often karyolysed and fragmented, but this character cannot be regarded as of generic value.

## 92. Karyolysus jorgei de Mello & de Meyrelles. (Fig. 71.)

†*Karyolysus jorgei*, de Mello & de Meyrelles, 1937, pp. 119–41, pl. vi, 4 text-figs.

Gametocytes, occurring in the red blood-corpuscles in the peripheral circulation of the lizard, are elongate oval, marked



Fig. 71.—Life-cycle of Karyolysus jorgei de Mello & de Meyrelles. A, male and female gametocytes in peripheral blood; B, endoglobular cycle in capillaries producing macrocysts with four merozoites; C, endothelial cycle in liver and lungs producing microcysts with eight leishmaniform merozoites; D, free merozoites which may invade either the red cells or endothelial cells. (After de Mello and de Meyrelles.)

by a strong limiting membrane, and contain a large central nucleus, a smaller roundish paranuclear body, and near the opposite pole a more or less irregular zone named as a polar eapsule. The gametocytes are sexually differentiated. In

the male the nucleus is central, oval or quadrangular in form, and always compact. In the female the nucleus may often be polar, and always contains a definite karyosome in its centre. In the capillary circulation occurs the endoglobular cycle. A merozoite invades the red corpuscle, grows into a schizont, and by twice repeated binary division forms a macroeyst containing four large gregariniform merozoites. These merozoites invade the endothelial cells of the lungs and liver and produce microcysts, each containing eight leishmaniform merozoites. The microcysts liberate the merozoites either in the interstices of the tissues or in the protoplasm of the endothelial cell itself. This second schizogonic cycle has been designated as the endothelial cycle. The leishmaniform merozoites may invade either the red corpuseles, where they undergo binary divisions again, or the endothelial cells, where they again become microcysts. It is not known from what particular forms the gametocysts are derived. Sporogony completely unknown.

Dimensions.—Gametocytes measure 5-11  $\mu$  by 1.5-2.5  $\mu$ . Macrocysts measure 13-15 $\mu$  by 8-10 $\mu$ ; macromerozoites measure 8-11 $\mu$  by 2.5-3 $\mu$ . Leishmaniform merozoites: roundish 1.5-3 $\mu$  in diameter, fusiform 3-5 $\mu$  by 1-1.5 $\mu$ .

*Remarks.*—The lack of karyolitic action on the part of the parasite and the occurrence of endoglobular schizogony makes the organism a remarkable transitional form between *Hæmo-gregarina* and the previously known species of *Karyolysus*. The differentiation of the gametocytes and the occurrence of a paranuclear body and specially differentiated polar area in the gametocytes and the peculiar form and structure of the micromerozoites are other characteristic features of the species.

Habitat.—Red blood-corpuscles and endothelial cells of the liver and lungs of *Calotes versicolor* Daud. subsp. major Blyth : PORTUGUESE INDIA, Nova Goa.

# 11. Suborder EIMERIDEA Léger, 1911.

The schizonts develop into micro- and macrogametocytes which are similar in size and develop independently of one another. The microgametocyte produces a relatively large number (six or more) of microgametes. The motionless zygote secretes a resistent oöcyst, which does not increase in size. The asexual and the sexual cycles occur in the same host.

Various schemes of classification of the suborder have been proposed. It is generally admitted that the composition of the mature oöcyst, viz., the number of sporocysts and sporozoites

#### EIMERIDEA.

it contains, provides the most convenient diagnostic characters for the differentiation of the genera : but different opinions have been held regarding the basis upon which the genera should be united into subfamilies and families. Following Lühe (1906), Minchin (1912), Reichenow (1921) and Wenyon (1926) arrange the genera into higher groups, mainly according to the type of schizogony. Schneider (1881) and Léger (1911) adopted the characters of the ripe oöcyst as the basis for classification : but while Schneider and his followers, Bütschli (1882), Labbé (1899) and Minchin (1903), arranged them according to the number of sporocysts within the oöcyst, Léger (1900), followed by Mesnil (1903), Poche (1913), Doflein (1916), Pinto (1928), and Nöller (1928), classified them according to the total number of sporozoites in the oöcyst.

Wenyon (1926) divides the suborder into six families, viz., Selenococcidiidæ, Cryptosporidiidæ, Eimeriidæ, Caryotrophidæ, Aggregatidæ, and Lankesterellidæ. Reichenow (1929) reshuffled the genera among three families, and brought over Dobellidæ from the ADELEIDEA and Leucocytozoidæ from the HÆMOSPORIDIA. Thus he classified the EIMERIDEA into five families, viz., Selenococcidiidæ, Aggregatidæ, Dobellidæ. Eimeriidæ, and Leucocytozoidæ.

Hoare (1933), leaving out of consideration the families Dobellidæ and Leucocytozoidæ (which might as well be included, as in Wenyon's classification), has proposed a modification of Léger's classification, in which the subfamilies are distinguished from each other by the number of sporocysts within the oöcyst, while the genera within each subfamily differ from one another in the number of sporozoites within each sporocyst. The suborder is divided into two families : (1) Selenococcidiidæ and (2) Eimeriidæ. The family Eimeriidæ is divided into six subfamilies. I have followed this classification in this work.

## Identification Table of Families.

[Léger & Duboscq. Selenococcidildæ \* Eimerildæ Léger, em. [Hoare, p. 158.

vermiform..... Eimerildæ Lég

#### SPOROZOA.

# 1. Family EIMERIIDÆ Léger, emend. Hoare, 1933.

Schizogony and sporogony are very uniform in character throughout the family, but variations occur in the number of the sporocysts and the sporozoites developed within the oöcyst. The family is divided into six subfamilies on this latter basis.

## Key to Subfamilies and Genera.

1. Schizogony and sporogony in the same host.

1.	Oöcyst asporocystid	Sublam. Cryptosporidiinæ
		[Poche, emond, Hoare, p. 159.
	Očevst tetrazoie	Cruptosporidium * Tyzzer.
	Očevst oetozoie	Pfeifferinella * Wasielewski.
	Očevst octozoje	Schellackia * Reichonow.
	Oöcyst polyzoic	Lankesterella Labbé, p. 159.
	v 1 v	
2.	Oöcyst monosporocystid	Subfam. CARYOSPORINÆ * (Wopyon.
	Sporocyst tetrazoic	Mantonella * Vincent
	Sporocyst cotrazole	Curuosporg * Loger
	Sporocyst detozote	Cargonporta Hogor.
3.	Oöcyst disporocystid	Subfam. CyclosporinÆ
		[Wonyon, p. 162.
	Sporocysts dizoic	Cyclospora * A. Schneider.
	Sporocysts tetrazoic	Isospora A. Schneider, p. 162.
	Sporocysts octozoic	Dorisiella * Ray.
4.	Oöcyst tetrasporocystid	Subfam. EIMERIINÆ Wenyon.
		[p. 173.
	Sporocysts dizoic	Eimeria A. Schneidor, p. 173.
	Sporocysts tetrazoic	Wenyonella Hoare, p. 197.
	Sporocysts polyzoic	Angeiocystis * Brasil.
5.	Oöcysts octosporocystid	Subfam. YAKIMOVELLINÆ *
	v 1 v	Gouseff,
	Sporocysts polycystid	Yakimovella * Gouseff.
		Q LC Demonstration
6.	Obcyst polysporocystia	Sublam. DARROUSSINÆ
	~	[(Wenyon), p. 199.
	Sporocysts monozoic	Barroussia * A. Schneider.
	Sporocysts monozoic	Echinospora * Léger.
	Sporocysts tetrazoic	Pythonella Ray & Das-
		[Gupta, p. 199.

II. Schizogony and sporogony in different hosts.

Occysts polysporocystid	Subfam.AGGREGATINÆReiche- [now, emend. Hoare, p. 199.
Sporocysts dizoic	Merocystis * Dakin.
Sporocysts dizoic	Pseudoklossia * Léger &
- •	[Duboscq.
Sporocysts trizoie	Aggregata Frenzel, p. 200.
Sporocysts dodecazoic	Caryotropha * Siedlecki.
Sporocysts dodecazoic	Ovivora * Mackinnon & Ray.
Sporocysts polyzoic	Myriospora * Lermantoff.

## Subfamily CRYPTOSPORIDIINÆ Poche, emend. Hoare, 1933.

Oöcyst contains four, eight, or many sporozoites developed without the formation of sporocysts.

### Genus LANKESTERELLA Labbé, 1899.

Drepanidium, Lankester, 1882, pp. 53-65; Labbé, 1894, p. 76.
Lankesterella, Labbé, 1899, pp. 73-5; Minchin, 1912, pp. 372, 378;
Nöller, 1913b, p. 231; 1920b, p. 176; Wenyon, 1926, pp. 878-80,
1105; Knowles, 1928, p. 471; Reichenow, 1929, pp. 964-5; Kudo,
1931, p. 275; Calkins, 1933, p. 566; Hoare, 1933, pp. 383, 385.

The whole of the development takes place in the endothelial cells of the blood-vessels. The oöcyst contains numerous (32 or more) sporozoites developed without the formation of sporocysts. The sporozoites finally enter the bloodcorpuscles and are mechanically transferred to another host by a blood-sucking animal.

Remarks.—This genus includes certain types which were originally considered as Hæmogregarines of cold-blooded animals. Nöller (1913 a, b and 1920 b) showed that the forms within the blood-corpuscles are sporozoites, and that the rest of the cycle takes place in the endothelial cells of the bloodvessels (instead of the intestine as in the typical coecidian) and is of the *Eimeria* type. Reichenow (1919) described another genus, *Schellackia*, from lizards, in which the development is on similar lines, but takes place in the intestine. He (1921 a) placed the two genera in a new family, Lankesterellidæ, and Wenyon (1926) followed this arrangement; but Reichenow (1929) has since placed both genera in the family Eimeriidæ, and Hoare (1933) has put them in a new subfamily, Cryptosporidiinæ, of the same.

## Key to Indian Species.

Sporozoite like a vornicule, with its anterior	[p. 159.
extremity tapering, $10-15 \mu$ in length	L. minima (Chaussat),
Sporozoite usually constricted into 3 segments,	
the middle containing the nucleus;	[p. 162.
$15-16\mu$ in length	L. monilis (Labbé),

## 93. Lankesterella minima (Chaussat). (Fig. 72.)

Anguillula minima, Chaussat, 1850.

Drepanidium ranarum, Lankester, 1871, pp. 387-9, figs. 3, 4; 1882, pp. 53-65; Labbé, 1891, p. 479; 1892, p. 617; 1893, p. 1207.
Drepanidium princeps, Labbé, 1894, p. 76.

Lankesterella ranarum, Labbé, 1899, p. 74; Minchin, 1903, pp. 239, 253-7, 260, 261, 265, 267, 270, 345; 1912, pp. 372-8.

Lankesterella minima Minchin 1903 p. 265. †Lankesterella minima, Patton, 1908, p. 319; 1909, pp. 146-7.

Hæmogregarina minima Mathis & Léger, 1911, pp. 448-9.

- Lankesterella minima, Minchin, 1912, p. 372; Nöller, 1912, pp. 201-8, pl. xx : 1913 a. pp. 313-16 : 1913 b. pp. 222-32, pl. xiv, figs. 55-68; pl. xv, figs. 69-72; 1920 a, pp. 169-89, pls. iv-vi; Wenyon, 1926, pp. 878-80, fig. 380; p. 1105.
- †Lankesterella minima, Scott, 1926, pp. 237-8; 1927, pp. 190-1.
- Lankesterella minima, Reichenow, 1929, pp. 964-5, fig. 930; Kudo, 1931, p. 276, fig. 15, g.

Lankesterella ranarum, Calkins, 1933, p. 545, fig. 218, B, C.

Infection of the frog is brought about by a leech, which introduces the sporozoites. Sporozoites make their way into the blood-capillaries of various organs, and apparently enter the endothelial cells, where the entire development takes place. Each sporozoite becomes rounded, grows into a schizont. and produces a large number of merozoites. The merozoites escape into the blood and infect other endothelial cells. Merozoites of a special kind are finally produced, and these, after entering the endothelial cells, develop into micro- and macrogametocytes. Microgametocyte produces a large number of microgametes, and fertilization of the macrogamete results. An occyst is formed round the zygote, which breaks up directly. without the formation of sporoblasts and sporocysts, into a number of sporozoites. The latter, by rupture of the oöcyst. escape into the blood and enter red blood-corpuscles. Here the sporozoite is seen as a small vermicule, which may attain half the length, but no more, of the corpuscle. The leech sucks up sporozoites with the blood and transfers them mechanically to another frog (tadpole).

Dimensions.—Sporozoite  $10-15\mu$  in length.

Remarks.-The cycle as described above is based on the researches of Nöller (1912, 1913, 1920). As Wenvon has remarked: "if this cycle of development is confirmed, it is a remarkable one in that the whole development, up to the formation of sporozoites, takes place in the endothelial cells of the blood-vessels, and is an illustration of a coccidium. originally transferred from host to host in the occyst stage, as in the more typical forms, having become adapted to life in the blood-stream.... The possibility of the escape of the occysts to the exterior having been lost by this change of habit, the difficulty is overcome by the leech transferring from host to host."

Some observers have described the vermicule as becoming spherical and undergoing schizogony in the red blood-corpuseles. but Nöller believes that such schizonts belong to Dactylosoma ranarum, another parasite of the frog's corpuscles.

Habitat.—Blood of Rana tigrina Daud., and in the leech which transmits it : MADRAS, Madras; also from the same host from India in the Zoological Gardens, London.



Fig. 72.—Lankesterella minima (Chaussat). A, young schizont;
B, schizogony in an endothelial cell; C, young forms in endothelial cell; D, microgametocyte; E, formation of microgametes in the endothelial cells of blood-vessels;
F, fertilization of a female gamete in an endothelial cell cell; C, occyst formed in an endothelial cell; H, mature occyst containing sporozoites in endothelial cell. (From Wenyon, after Nöller.)

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## 94. Lankesterella monilis (Labbé). (Fig. 73.)

Hæmogregarina ranarum (part). Celli & Sanfelice, 1891, p. 504, pl. v, figs. 2, 3, 4, 12-15; Kruse, 1890, p. 541.
Drepanidium monile, Labbé, 1894, p. 76, pl. iii.
Lankesterella monilis, Labbé, 1899, pp. 74-5, fig. 140.
†Lankesterella monilis, Berestneff, 1903, pl. viii, fig. 7.
Lankesterella monilis, Minchin, 1903, pp. 265, 267, 345.

Free stages very mobile and showing successive undulations in locomotion. Body shows constriction into three segments, the middle one containing the nucleus. Nucleus vesicular,



Fig. 73.—Lankesterella monilis (Labbé). Free forms. (After Labbé.)

containing a karyosome and numerous chromatoid granules. No vacuole. Cysts as in *L. ranarum*.

Habitat.—Blood of Rana tigrina Daud. and R. limnocharis Wiegmann : BOMBAY, Bombay.

# Subfamily CYCLOSPORINÆ Wenyon, 1926.

Occysts contain two sporocysts, each containing two, four, or eight sporozoites.

## Genus ISOSPORA Aimé Schneider, 1881.

Isospora, Aimé Schneider, 1881, p. 401.
Coccidium, Grassi, 1881, p. 135, pl. iii, figs. 37-40; p. 192.
Diplospora, Labbé, 1893, p. 407.
Hyaloklossia, Labbé, 1896, p. 535; 1899, p. 55.
Isospora, Labbé, 1899, p. 72.
Diplospora, Labbé, 1899, p. 71.
Isospora, Laveran & Mesnil, 1902 c, p. 83; Castellani & Chalmers, 1919, pp. 473, 475; Dobell, 1922, pp. 1497-8; Craig, 1926, pp. 349-53; Wenyon, 1926, pp. 806-28; Knowles, 1928, pp. 351-8; Reichenow, 1929, pp. 955-61; Kudo, 1931, pp. 274-5; Calkins, 1933, p. 565; Hoare, 1933, pp. 364-9, 383, 385.

Oöcyst develops two sporocysts, each containing four sporozoites.

Remarks.—Species belonging to this genus are known from man, cats and dogs, birds, lizards, and frogs. Cats and dogs

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harbour three distinct species. One, or probably two, species are known from man. Becker (1934) gives the names and hosts of 42 named species and the hosts of 4 unnamed species of Isospora.

## Key to Indian Species.

1(12).	Infection limited to epithelial cells	2.
2 (4).	Oöcysts spherical	3.
3.	Oöcysts $18-23 \mu$ in diameter ; sporo-	
	cvsts $12-15 \mu$ by $8-10 \mu$ . Intra-	[Das-Gupta, p. 170.
	nuclear	1. knowlesi Rav &
4 (2).	Oöcysts not spherical	ő.
5 (10).	Oöcysts egg-shaped	6-9.
6.	Occysts $25-33 \mu$ by $12\cdot 5-16 \mu$ : sporo-	
	$evsts 12-14 \mu bv 7-9 \mu$ . In man	L. belli Wenvon, p. 163.
7.	Obeyets $35-45 \mu$ by $25-35 \mu$ ; spore-	
••	$evets 18.4 \mu$ by 11.4 $\mu$ In cats	L felis Wenvon n 168
8	Measurements of occusts and spore.	In 167
0.	cysts not recorded In lizards	I calotesi en nov
9	Obevets 15, by 7.5, sporocysts	Des Cupte n 170
01	7.5 u in diameter In cobra	L. minuta Mitra &
10 (5)	Occysta subcylindrical	1)
11 (0).	Obeyets $16-20 \mu$ by $11-14 \mu$ spore.	Das.Gunta n 17?
	evets $8_{\mu}$ by $4_{\mu}$	I menuoni Roy &
19 (1)	Infaction may extend to the sub-	1. wengoni Hay a
1 - (1).	anitholial tissue also	19
12 (14)	Larger of outs 18 $20 \mu$ by $14 - 16 \mu$ (in	15.
13 (14).	Larger obcysts $18-20\mu$ by $14-10\mu$ (m	
	$(\log s)$ ; smaller occysts $10-14\mu$ by	Em. 1.07
	$75-9\mu$ (in cats and dogs); sporo-	$[\mathbf{p}, 100,$
14 / 19	$e_{y_{858}} = 15^{\circ}0^{-10^{\circ}}0^{\circ}\mu \text{ Dy } 9^{-10^{\circ}}\mu \dots \dots$	1. orgemina (Stiles),
14 (13).	Uncertain $20-25 \mu$ by $15-22 \mu$ ; sporo-	p. 171.
	cysts 10 $\mu$ Dy 10 $\mu$	1. rivena (Grassi),

#### 95. Isospora belli Wenyon. (Fig. 74.)

Isospora hominis, Dobell, 1922, pp. 1497-8, fig. 533 A. Isospora belli, Wenyon, 1923 a, p. 269.

- *Hospira belli*, Knowles, 1924, p. 64. *Isospora belli*, Wenyon, 1926, pp. 820-4 fig. 350, 7-10; 1926 a, pp. 253-66.

Isospora hominis, Dobell, 1926, pp. 74-85; Craig, 1926, pp. 349-53, fig. 62.

†*Isospora belli*, Knowles, 1928, pp. 356-8, fig. 81, 4, 17-21.

Isospora belli, Reichenow, 1929, p. 958, fig. 923; Kudo, 1931, p. 274, fig. 114, a-c; Calkins, 1933, p. 405. †Isospora belli, Knowles, 1933, p. 53; Das-Gupta, 1934. pp. 133-4,

pl. ii.

Only the occysts and sporocysts are known. The former are elongate, egg-shaped, with one end more constricted than the other, forming a kind of neck. They are transparent, colourless bodies, with a wall consisting of two layers, the outer thick and porcellanous, the inner thin and membranous. At the narrow end there is an indication of a micropyle. In fresh stools are found unripe occysts with cytoplasmic

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contents contracted into a spherical body with highly refractile granules, and a single nucleus seen as a clear pale area. The oöcysts complete their development in one to four days, according to the temperature. The cytoplasm divides into two sporoblasts, which become elongated and covered with cyst-walls. In each sporocyst are developed four sporozoites and a large spherical residual body. Sporozoites are elongate structures, rounded at the anterior and tapering at the posterior end, and with a nucleus lying at the junction of the anterior and the middle third.

The organism is probably a parasite of the epithelial cells of the small intestine, where schizogony and the development of the gametocytes will be found to occur.



Fig. 74.—Isospora belli Wenyon.

Dimensions.—Oöcysts  $25-33 \mu$  in length by  $12 \cdot 5-16 \mu$  in width; sporocysts  $12-14 \mu$  by  $7-9 \mu$ .

Pathogenicity.—Wenyon (1926) reports an observation by Conrad (1922) on a laboratory worker who accidentally ingested material containing developed occysts. Six days later diarrhœa set in and persisted for thirty days. The occysts were found three weeks after the onset, and persisted in the patient's stools for 12 days, after which they disappeared, and recovery was complete.

Similar symptoms and similar cysts have been found by a number of workers in different countries, but it is not certain if they belong to one or more species.

A, immature oöcyst; B, mature oöcyst. (From Reichenow, after Dobell.)

Remarks .- The parasite named Isospora hominis (Railliet & Lucet, 1901) was first discovered by Virchow in 1860. Human Coccidia were observed, among others, by Woodcock (1915), Wenyon (1915), Cragg (1917), Dobell and O'Connor (1921), and Reichenow (1925), and referred to I. hominis, but according to Wenyon (1926) all these later findings of Isospora occysts refer to I. belli and not to I. hominis, which he regards as a species with small occysts. Dobell (1926) considers that the small subepithelial form is identical with the larger form in the epithelium. He holds that the case for two species is not proven, and that the name Isospora hominis should be adhered to. Wenyon (1926 a) has replied vigorously and adheres to his former opinion. Reichenow (1929) recognizes the two species as distinct.

Cragg (1917) reported four cases from Bombay, but all these patients are believed to have contracted the infection in the Mediterranean war area. Knowles (1928) reported having observed the infection in man five times during the six preceding years, and reported another case in 1933. Das-Gupta (1934) recorded his observations on the case of a Bengali Brahman from Calcutta who had never been abroad.

Habitat.—Fæces of man: BOMBAY, Bombay; BENGAL, Calcutta.

#### 96. Isospora bigemina (Stiles). (Fig. 75.)

Corpuscles géminés, Finck, 1854.

Cytospermium villorum intestinalium canis et felis, Rivolta, 1874, p. 1; 1877, pp. 42-6, 85-8.

Coccidium bigeminum, Stiles, 1891, p. 163; 1892, pp. 517-26;

 Railliet & Lucet, 1891, p. 250; Labbé, 1896, p. 545; 1899, p. 67.
 *Hospora bigemina*, Castellani & Chalmers, 1919, p. 473.
 *Isospora bigemina*, Wenyon, 1923 a, p. 257, pl. xiii, figs. 1-11;
 Wenyon & Sheather, 1925, p. 10; Wenyon, 1926, pp. 809-13,
 figs. 343, 344; 1926, pp. 253-66; Knowles, 1928, pp. 351-5,
 figs. 343, Dicker and State an fig. 81, 1; Reichenow, 1929, p. 957; Kudo, 1931, p. 274, fig. 114 d.

Sporogony not confined to the epithelial cells; it usually takes place and is completed in the subepithelial tissue of the villi. In the acute stages of infection reproduction occurs in the epithelium, and immature occysts are passed in the fæces. Ööcysts of two types. The smaller ones occur in both cats and dogs, while the larger ones have hitherto been The occysts ripen in the gut-tissue rather seen only in dogs. than in the fæces: they have thin walls and the sporocysts may escape from them and be present in the fæces and carry the infection from one host to another. The sporocyst contains four sporozoites and a little residual substance in the form of a small clump or as dispersed granules.

Dimensions.—Occysts, smaller  $8-14\mu$  by  $7-9\mu$ , larger  $18-20 \mu$  by  $14-16 \mu$ ; sporocysts  $13\cdot 5-15\cdot 5 \mu$  by  $9-10 \mu$  (in dog). *Remarks.*—This parasite is usually found in the subepithelial tissue of the villi, where it completes its sporogony, so that immature occysts are not passed out in the fæces. Wenyon and Sheather (1925) had an opportunity of studying the intestine of a dog which had been killed during an acute phase



Fig. 75.—Section of villas of cat, showing *Isospora bigemina* in the subepithelial tissues and *Isospora felis* in the epithelium  $(\times 1000)$ . Six mature occysts of *I. bigemina* are seen in the subepithelial tissues. In the epithelium are seen a microgametocyte with microgametes, and one partially grown and two mature macrogametocytes. (After Wenyon.)

of infection. The whole of the epithelium of the small intestine was found to be crowded with reproducing parasites; but the subepithelial tissue was not invaded. The schizonts were small, measuring up to  $5\mu$  and giving rise to eight minute merozoites. There were numerous macrogametocytes measuring about  $7.5\mu$  in diameter. During life this dog passed in its fæces numerous immature oöcysts measuring  $10-14\mu$  by  $7.5-9\mu$ , the sporocysts measuring  $7.5-9\mu$  by  $5-7\mu$ . In another puppy a large number of oöcysts of the same dimensions were passed, and completed their development outside the body in the usual manner, forming two sporocysts, each with four sporozoites and a residual body.

Wenyon (1926) is of the opinion that there are three species of *Isospora* in eats and dogs, and gives the dimensions of the oöcysts as follows :—

- I. felis, 39-48  $\mu$  by 26-37  $\mu$ .
- I. rivolta, 20-24  $\mu$  by 15-20  $\mu$ .
- I. bigemina (large),  $18-20 \mu$  by  $14-16 \mu$ .
- I. bigemina (small),  $10-16 \mu$  by  $7.5-10 \mu$ .

Dobell (1926) criticizes this view, but Wenyon (1926) adheres to it. Reichenow (1929) recognizes these species as distinct. According to Yakimoff, Matikaschwili, Rastegaïeff, and Lewkowitsch (1930) the domestic cat harbours three species, viz., *I. bigemina* var. cati (Stiles), *I. rivolta* (Grassi), and *I. felis* Wenyon. Sen (1932) noted an unidentified coccidian from a dog at Muktesar.

Habitat.—Subepithelial tissue of the intestine in dogs: CEYLON, Colombo.

## 97. Isospora calotesi, sp. nov.

†Isospora sp., Setna, 1933, p. 97.

Schizonts spherical. Multiple fragmentation gives rise to a number of small ovoid merozoites which gradually become vermiform. Merozoites vary in size : there may be four large merozoites clustered round a central cytoplasmic residue, or a large number, up to about 100, of slender fusiform merozoites. Male and female gametocytes may develop in the same epithelial cell or in separate ones, and in the earlier stages are difficult to distinguish from the schizonts and from one another.

*Remarks.*—The developmental stages are clearly marked off, and as many as 80 per cent of the lizards examined were infected. Infection was limited to the epithelium; the subepithelial tissue was not affected.

Dimensions.—Schizonts up to  $22 \mu$  by  $17 \mu$ ; merozoites, small and slender  $9 \mu$  by  $1 \cdot 2 \mu$ , large  $8 \mu$  by  $2 \cdot 8 \mu$ .

Habitat.—Intestine of the lizard, Calotes versicolor (Daudin) : BOMBAY, Bombay.

## 98. Isospora felis Wenyon. (Figs. 75 & 76.)

Isospora felis, Wenyon, 1923 a, p. 248, pls. ix-xii; 1926, pp. 808, 814, figs. 342, 344-8; 1926 a, pp. 253-66.

†Isospora sp., Knowles, 1928, p. 354.

Isospora felis, Reichenow, 1929, p. 956, figs. 921 A, 922; Kudo, 1931, pp. 274, fig. 114 f.

†*Isospira* sp. (" type B "), Knowles & Das-Gupta, 1934, pp. 387–90, pl. vii, figs. 1-4, 6.

Development takes place in the epithelial cells only of the small intestine and not in the deeper layers of the villi. Oöcysts egg-shaped, with one pole somewhat narrowed. The oöcysts



Fig. 76.—Mature oöcyst of *Isospora felis* Wenyon. (From Reichenow, after Dobell.)

drop out of the epithelial cells into the lumen of the gut and pass out with the fæces, the formation of sporocysts and sporozoites taking place in the oöcysts while outside the body of the host. The macrogametocyte is easily distinguished in slightly advanced stages, as it has a single nucleus with a characteristic appearance. In the microgametocyte the nucleus divides repeatedly and hundreds of microgametes develop in each. Each microgamete is a flexible rod-like body with two flagella. Fertilization takes place while the macrogamete is within an epithelial cell. The oöcyst develops

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a thick chitinous wall after it has passed into the lumen of the intestine. Oöcysts containing two sporoblasts may be found in the gut, and in this condition are passed out in the fæces. Outside the body the sporoblasts in the oöcyst elongate, acquire a double contour, and become sporocysts. In each sporocyst are developed four elub-shaped sporozoites and a large rounded, granular, residual body.

Dimensions.—Oöcysts, egg-shaped type  $35-45\,\mu$  by  $25-35\,\mu$ (39-48 $\mu$  by 26-37 $\mu$  according to Wenyon), rounded type  $25-37\,\mu$  by 21-37 $\mu$ ; microgamete  $3\cdot1\,\mu$ ; sporocysts  $18\cdot4\,\mu$  by 11·4 $\mu$ .

Remarks.-Knowles (1928) stated that Isospora infection was not uncommon in cats in Calcutta, and that infection was absolutely limited to the epithelial cells, but did not mention the name of the species or give the dimensions of the occvsts. On my request for further information, Knowles and Das-Gupta (1934) examined the stools of 13 cats and 8 kittens, and by the employment of concentration method found 14 out of 21 animals examined to be infected. In all they measured 353 oöcysts. After discussing the views of Wenyon and of Dobell they are of opinion that the evidence collected by them seems to point to two "types" being present, though they are not in a position to assert that they are different species. The smaller, oval type of oöcyst encountered measured from  $20.4 \mu$  by  $15.3 \mu$  to  $47.6 \mu$  by  $40.8 \mu$ , and the larger, egg-shaped type of oöcysts measure  $38-45 \mu$  by  $27 - 36 \mu$ .

Their "type A" or small oöcysts are always perfectly oval and resemble I. rivolta, and the "type B" or large oöcysts are more egg-shaped, pyriform or ovoid, and resemble I. felis. I am inclined to think that the form of the oöcyst and the measurements given show that the two "types" are distinct species, as is held by Wenyon and Reichenow.

The vast majority of the oöcysts seen by Knowles and Das-Gupta in freshly passed fæces were in the unsegmented state, with the protoplasmic contents present as a single spherical mass within the thick oöcyst wall, while in a few the first nuclear division had taken place and two sporoblasts were forming, although the sporocysts had not yet formed. They also noted the occurrence of a fair number of motile sporozoites in the eontents of the jejunum and ileum, both in the fresh preparations and in fixed and stained films, from which they conclude that an occasional oöcyst may develop to maturity within the lumen of the gut, although the vast majority are passed in the unsegmented state.

Habitat.—Epithelial cells of the small intestine of cat, Felis domesticus Linn.: BENGAL, Calcutta.

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### 99. Isospora knowlesi Ray & Das-Gupta. (Fig. 77.)

†*Isospora* sp., Knowles & Das Gupta, 1935, pp. 703, 705, pl. xxx, figs. 14, 20.

Isospora knowlesi. Ray & Das-Gupta, 1937 c, pp. 269-74, pl. vii.

Young gametocyte intranuclear. Male gametocyte gives rise to biflagellate gametes which drop into the lumen of the intestine or penetrate the neighbouring epithelial cells. Oöcysts thick-walled, spherical. Sporoeysts ellipsoidal, with a knob-like structure at one pole. Sporozoites arranged regularly, with large sporoeystic residue. Unsegmented or segmented but immature oöcysts discharged from the host. Sporulation takes place in 4 to 5 days.



Fig. 77.—Isospora knowlesi Ray & Das-Gupta. A, oöcyst within the nucleus of a host-cell; B, oöcyst from the fæcal matter; C, mature oöcyst. (After Ray & Das-Gupta.)

Dimensions.—Oöcysts  $18-23\,\mu$  in diameter; sporocysts  $12-15\,\mu$  by  $8-10\,\mu$ .

Remarks.—The occysts resemble those of Isospora mesnili Sergent (1902) in being intranuclear in habitat, and approach those of I. camilleri Hagenmuller (1898) in size.

Habitat.—Nuclei of epithelial cells of the small intestine of Hemidactylus flaviviridis (Rüppell): BENGAL, Calcutta.

## 100. Isospora minuta Mitra & Das-Gupta.

Isospora minuta, Mitra & Das-Gupta, 1937 a, p. 291.

Oöcysts disporceystid, final stages of sporceyony taking place outside the body of the host. Sporceysts spherical. Oöcystic residue absent.

Dimensions.—Maximum size of oöcyst  $15 \mu$  by  $7.5 \mu$ ; sporocyst  $7.5 \mu$  in diameter.

Remarks.-Full description of the species has not yet been

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published. The form is said to differ from I. dirumpens Hoare in that development inside the oöcyst takes place outside the body of the host. The disporocystid condition of the oöcyst was evidenced after being kept in I per cent. chromic acid for three days. It also differed from I. naix Fantham in that there was no oöcystic residuum.

Habitat.—In the fæcal matter of Naja naja (Linn.) : BENGAL, Calcutta.

## 101. Isospora rivolta (Grassi). (Fig. 78.)

- Coccidium rivolta, Grassi, 1879, p. 135, pl. xxxiii, figs. 41-4; 1881 a, p. 632.
- Coccidium bigeminum, Labbé, 1899, p. 67.
- *Isospora rivolta*. Brown & Stammers, 1922, p. 1165; Wenyon, 1923 a, p. 260, figs. 12–15; 1926, pp. 808, 813, figs. 342, 345, 346, 350.
- †Isospora sp., ("type A"), Knowles, 1928, p. 354, fig. 81, 2.
- Isospora rivolta, Reichenow, 1929, pp. 956-7, fig. 921 b; Kudo, 1931, p. 274, fig. 114 e.
- †Isospora rivolta, Knowles & Das-Gupta, 1931, pp. 175-6, pl. ix.
- †*Isospora* sp. ("type A"), Knowles & Das-Gupta, 1934, pp. 387-90, pl. vii, figs. 5, 6.
- †Isospora rivolta, Aiyar, 1937, p. 402.

Occysts ovoid, with a double contour. In each are developed two sporocysts, and a small residual body may be present. The sporocysts are elongate bodies with rounded ends.



Fig. 78.—Mature oöcyst of *Isospora rivolta* (Grassi). (After Reichenow.)

Each sporocyst contains four sporozoites and a large residual body filled with globules of a refractile material. The infection is usually confined to the epithelial layer, but the form may also reproduce in subepithelial tissues of the alimentary canal of the host and produce male and female gametocytes.

Dimensions.—Oöcysts  $20-25\mu$  in length by  $15-22\mu$  in breadth; sporocysts  $16\mu$  by  $10\mu$ .

**Remarks.**—This is a common parasite of cats and dogs. Knowles and Das-Gupta (1934) have recorded the measurements of 253 oöcysts and consider the smaller oöcysts, which are perfectly oval and measure  $23-28 \mu$  by  $18-23 \mu$ , as belonging to "type A," which corresponds to *I. rivolta* (Grassi).

Knowles and Das-Gupta (1931) also found in the cæcal contents of a small Indian mongoose immature oöcysts of a coccidium containing a single rounded unsegmented mass of protoplasm, and studied its development in a moist-chamber. The average dimensions of the oöcyst were  $20.6 \mu$  by  $17.2 \mu$ .

Habitat.—Epithelial cells of the small intestine of cat, Felis domesticus Linn. : BENGAL, Calcutta ; also in the cæcal contents of the mongoose, Herpestes auropunctatus (Hodgs.) : BENGAL, Calcutta.

## 102. Isospora wenyoni Ray & Das-Gupta. (Fig. 79.)

†Isospora wenyoni, Ray & Das-Gupta, 1935 a, pp. 219-24, pl. viii.

Young intracellular trophozoites are found in the epithelial



Fig. 79.—Isospora wenyoni Ray & Das-Gupta. A, merozoite entering an epithelial cell; B, merozoites escaping into the humen of the intestine; C, microgametocyte; D, microgametes clustered round a central mass of cytoplasm; E, macrogametocyte showing spherical nucleus and the karyosome; F, macrogamete showing a recurved tail, an elongated nucleus, and two microgametes near the posterior end; G, mature očcyst. (After Ray and Das-Gupta.)

cells of the small intestine. Young schizonts possess eight to twelve nuclei, and schizogony results in producing the same number of merozoites. Each merozoite is spindle-shaped, and carries a pair of hyaline blades or laminæ at its anterior end, which help it to enter the epithelial cell. The male gametocyte shows a large number of nuclei situated towards its periphery, and the mature microgametes are found to cluster round a residual mass of cytoplasm. The macrogametocyte is distinguished by its size and by the presence of darkly staining granules in its cytoplasm; its posterior end shows a short recurved tail; the nucleus at an early stage is spherical and contains a karyosome, but when mature becomes elongated. and the karyosome breaks up into a number of small, irregularly scattered granules. Occysts are subcylindrical, disporocystid, and tetrazoic, and develop outside the host. There is no oöcystic residuum, and the oöcystic membrane has a double contour. Sporocysts have their long axis directed at right angles to the long axis of the occysts; a sporocystic residuum is present.

Dimensions.—Trophozoites, young  $10\mu$  by  $3\mu$ ; schizonts, young  $20-25\mu$ ; merozoites  $12\mu$  by  $5\mu$ ; microgametes  $2\cdot4\mu$  by  $1\cdot5\mu$ ; macrogametes  $16-20\mu$  by  $11-14\mu$ ; oöcysts  $16-20\mu$  by  $11-14\mu$ ; sporocysts  $8\mu$  by  $4\mu$ .

Habitat.—In the small intestine of the toad, Bufo melanostictus Schneider : BENGAL, Calcutta.

## 103. Isospora sp.

†Isospora sp., Cooper & Gulati, 1926, pp. 191-2.

Oöcysts of an unidentified species of *Isospora* were found in the fæces of a heifer calf at Dhupatal, of a bull calf and two cows at Tocklai, and of a heifer at Shillong, in Assam.

Oöcysts were quite round, and within twenty-four hours two fully formed sporoblasts were seen in all cases.

Habitat.—Alimentary canal of cows: Assam, Dhupatal, Tocklai, and Shillong.

## Subfamily EIMERIINÆ Wenyon, 1926.

Oöcysts contain four sporocysts, each containing two, four, or many sporozoites.

## Genus EIMERIA Aimé Schneider, 1875.

Gregarina, Eimer, 1870, p. 4. Eimeria, Aimé Schneider, 1875 a, pp. xl-xlv. Psorospermium, Rivolta, 1878. Coccidium, Leuckart, 1879, p. 254. Orthospora, Aimé Schneider, 1881, p. 389. Crystallospora, Labbé, 1896, p. 554; 1899, p. 63.

Goussia, Labbé, 1896, p. 551; 1899, pp. 63-5.

Eimeria, Labbé, 1899, pp. 58-9.

Paracoccidium, Laveran & Mesnil, 1902 b, p. 858.

 Eimeria, Castellani & Chalmers, 1919, pp. 475-6; Dobell, 1919 a, pp. 147-97; Snijders, 1920, pp. 427-32; Dobell, 1922, pp. 1498-9.
 Jarrina, Léger & Hesse, 1922, pp. 74-7.

Eimeria, Craig, 1926, pp. 353-8; Thomson & Robertson, 1926, pp. 282-3; 1926 a, pp. 420-1; Wenyon, 1926, pp. 828-62; Knowles, 1928, pp. 358-66; Reichenow, 1929, pp. 937-55; Kudo, 1931, pp. 271-4; Stiles, 1929, p. 881; Levine & Becker, 1933, pp. 83-106; Calkins, 1933, pp. 403, 565; Hoare, 1933, pp. 374-7, 378, 381, 383, 385; Wenrich, 1935, pp. 7-9; Ray & Das-Gupta, 1936, p. 345-6; 1937 d, pp. 275-7.

Oöcyst develops four sporocysts, each containing two sporozoites.

*Remarks.*—The typical life-history is as described by Schaudinn for E. schubergi, of which a brief summary is given for ready reference. Occysts gain entrance into the host through the mouth. Sporozoites escape from the sporocysts and, passing through the micropyle of the occyst, move about in the lumen of the gut till they enter the epithelial cells of the gut-wall, where they grow into schizonts. Schizonts are large rounded bodies, which give rise to merozoites. The latter escape into the lumen of the gut and enter new host-cells and repeat the process. Some merozoites develop into macro- and microgametocytes. The macrogametocyte produces a single macrogamete after extruding part of its nuclear material. The microgametocyte produces a number of biflagellate microgametes. Syngamy takes place and the zvgote secretes a membrane around itself, forming an occyst. The nucleus divides twice and four sporoblasts are developed inside the oöcyst. Each sporoblast secretes a membrane and becomes a sporocyst, and two sporozoites are developed inside each sporocyst. Oöcysts pass out in the fæcal matter of the host and infect other hosts by being ingested.

The genus has a very wide distribution, numerous species having been described from all classes of Vertebrates, terrestrial, freshwater, and marine, and a few Invertebrates (myriopods, etc.). They have been found to occur as intestinal parasites in man, horses, cattle, pigs, sheep and goats, rats and rabbits ; domestic birds such as fowls, ducks, pigeons, pheasants, etc. ; a few lizards and tortoises ; in frogs, newts, and salamanders ; in fish ; in centipedes, etc. According to Levine and Becker (1933) no less than 220 species of this genus have been recorded from 183 species of hosts of widely different groups. These hosts include Annelids, 2 species ; Myriapods, 6 species ; Insects, 3 species ; Enteropneusta, 1 species ; Fishes, 43 species ; Amphibia, 10 species ; Reptiles, 26 species ; Birds, 24 species ; and Mammals, 60 species. The shape, dimensions,

and colour of the oöcyst, the appearance of the oöcyst wall, the character of the micropyle, and the presence or absence of a residual mass when the sporoblasts separate, as also the shape and size of the sporocyst, are taken into account in identifying the species. Allen (1934) gives a key to twentyfour known species of *Eimeria* in birds.

According to Dobell (1932), Leeuwenhoek probably first saw the occysts of rabbit Coccidia as far back as 1674, but, as remarked by Wenrich (1935), it has only recently been demonstrated beyond a doubt that rabbit Coccidia are peculiar to



Fig. 80.—Life-cycle of *Eimeria schubergi* (Schaudinn). ( $\times$ 400.) *A*, entrance of a sporozoite in tho gut epithelium of the host and growth of schizont; *B*, three stages in schizogony to form merozoites, which repeat schizogony; or *C*, become macro- and microgametocyte; *D*, *E*, formation of macrogameto; *F-H*, formation of microgametos; *I*, inature gametes and fertilization; *J*, secretion of a membrane round the zygote; *K-N*, stages in sporocyst formation; *O*, oöcyst containing four sporozysts, each with two sporozoites; *P*, escape of the sporozoites. (From Kudo, after Schaudinn.)

rabbits and different from those of cattle and poultry; and further, that rabbits may harbour five or six different species of *Eimeria*, of which *E. stiedæ*, occurring in the liver, is the one most often causing disease and death; the other species live in the intestine.

## SPOROZOA.

# Key to Indian Species.

1(15)	. Oöcysts spherical	2.
2 (8)	. Sporocysts with rounded ends	3.
3.	Oöcysts $18-21 \mu$ in diameter, without	
	residium; sporocysts $10\mu$ by $7\mu$ ,	(han), p. 177.
	with residuum. In fish and man	E. clupearum (Thélo-
4.	Oöcysts almost spherical (sometimes	
	oval), $16-20 \mu$ by $14-18 \mu$ , without	
	residuum; sporocysts spherical or	[p. 184.
	oval. In lizards	E. knowlesi Bhatia,
5.	Oöcysts 16–18 $\mu$ in diameter, without	
	residuum; sporocysts oval, with	[p. 186.
	residuum. In tortoises	E. legeri (Simond),
6.	Obcysts $20-22 \mu$ , without residuum;	
	sporocysts oval, with residuum. In	[(Simond), p. 183.
	gharials	E. kermorganti
7.	Occvsts spherical or almost spherical,	v
	$12-25\mu$ in diameter; sporocysts	
	pear-shaped, 9.9-11 $\mu$ by $5.3-5.7 \mu$ ;	[p. 194.
	without residuum. In cattle	E zürni (Rivolta).
8 (2)	. Sporocysts fusiform or with pro-	• •
,	jections	9.
9 (11).	Sporocysts olliptical, narrow end pro-	
. ,	jecting as a neck and bearing an	
	inverted V-shaped appendage	10.
10.	Oöcysts 9-13 $\mu$ in diameter; sporo-	[Bana, p. 18].
	cysts $9 \mu$ by $4 \mu$ . In fish	E. harpodoni Setna &
11 (9).	Sporocysts fusiform	12.
12. `´	Obcysts 36-52 $\mu$ in diameter, with	
	residuum; sporocysts fusiform,	
	$30-32 \mu$ by 7.5 $\mu$ , with residuum. In	[p. 189.
	fish and man	E. sardinæ (Thélohan),
13.	Oöcysts $14\mu$ in diameter, without resi-	
	duum; sporocysts spindle-shaped,	[p. 184.
	with residuum. In tortoise	E. koormæ Das-Gupta,
14.	Oöcysts 8–11 $\mu$ in diameter, no resi-	•
	duum; sporocysts $4.5-6\mu$ by $3\mu$ ;	
	schizonts bear laminæ at anterior	[p. 185.
	end. In toads	E. laminata Ray,
15 (1).	Oöcysts not spherical	16.
16 (18).	Oöcysts mitro-shaped	17.
17.	Occysts with 4 or 5 projections, 10-	
	$15\mu$ in diameter, without residuum ;	[Mesnil), p. 187.
	sporocysts oval. In tortoises	E. mitraria (Laveran &
18 (16).	Oöcysts subspherical, ovoid, oval or	
	cylindrical	19.
19 (24).	Oöcysts subspherical, ovoid or lemon-	
	shaped	20.
20 (21).	Occysts subspherical, $16.4 \mu$ by $14.35 \mu$ .	
	Oöcystic residuum present. In	[Das-Gupta, p. 178.
	pigeons	E. columbæ Mitra &
21 (20).	Oöcysts ovoid or lemon-shaped	22.
22 (2 <b>3</b> ).	Oöcysts lemon-shaped, $17-20.4 \mu$ by	[& Das-Gupta, p. 182.
	13.6–17 $\mu$ . In lizards	E. hemidactyli Knowles
23 (22).	Occysts usually ovoid, $20-40\mu$ by	
	$17-20 \mu$ , sometimes spherical, $18 \mu$ in	
	diameter ; sporocysts $13 \mu$ by $6 \mu$ ,	[Marotel), p. 179.

24 (19).	Oöcysts oval or cylindrical	25.
25 (34).	Oöcysts oval	26.
26 (31).	Sporocysts ovoid or oval	27.
27.	Occysts $17-34 \mu$ by $11-15 \mu$ , without residuum; sporocysts ovoid, $7-9 \mu$ by $5-7 \mu$ , with residuum, In lizards	[Bana, p. 179. E. flaviviridis Setna &
28.	Oöcysts egg-shaped, $25-35 \mu$ , without residuum; sporocysts pear-shaped, $10.8-14.4 \mu$ by $7-9 \mu$ , with residuum. In cattle	[& Galouzo, p. 191.
29.	Očcysts $32.4-41.4 \mu$ by $21.6-28.8 \mu$ , without residuum; sporocysts oval, $13-19 \mu$ by $3-5.4 \mu$ , without resi-	[gaïeff, p. 194.
	duum. In nilgau	E. yakimovi Raste-
30.	Oöcysts egg-shapod, $18\mu$ by $14\cdot4\mu$ . Sporocysts not known In deer	[gaïeff, p. 194.
31 (26)	Sporocysts fusiform	39
32 (33).	Occysts $18:8-27:04 \mu$ by $18:8-20:8 \mu$ , without residuum; sporocysts $14:6 \mu$ by $6 \mu$ . In snakos	[Gupta, p. 188. E. najæ Ray & Das-
33 (32).	Obcysts 29-31 $\mu$ by 22.5-24.5 $\mu$ , with residuum; sporocysts 14 $\mu$ by 4-6 $\mu$ . In snakes	[Cupta, p. 189. E. piscatori Ray & Das-
34 (25).	Oöcysts cylindrical	35.
35 (36).	Occysts $36 \mu$ by $18 \mu$ , with residuum. In snakes	[Das-Gupta, p. 179. E. cylindrica Ray &
36 (35).	Obcysts $25 \mu$ by $12 \mu$ , with residnum; sporocysts oval, $10-12 \mu$ in length. In fish	[p. 193. E. southwelli Halawani,

## 104. Eimeria clupearum (Thélohan). (Fig. 81.)

Coccidium sp., Thélohan, 1892, p. 158, pl. xii, figs. 13, 14.

Coccidium clupearum, Thélohan, 1894, p. 565, pl. xxii, figs. 19, 20. Goussia clupearum, Labbé, 1896, p. 552, pl. xviii, figs. 24, 25; 1899, p. 64.

Eimeria sp., Wenyon, 1915 b, p. 1404.

*Eimeria wenyoni*, Dobell, 1919 *a*, pp. 187-8, pl. viii, fig. 2; Castellani & Chalmers, 1919, pp. 475-6; Doboll & O'Connor, 1921, p. 100, pl. vi, fig. 104.

†Eimeria clupearum, Knowles, 1924, p. 24; 1928, p. 362, fig. 83.

*Eimeria clupearum*, Wenyon, 1926, pp. 851-2, 861, fig. 350, 11; Thomson & Robertson, 1928, pp. 360-1, 363; fig. 83, 2; Reichenow, 1929, pp. 951, 952-3, fig. 918 b; Kudo, 1931, p. 274, fig. 113 n.

The oöcyst is spherical, is of a light brown colour, and possesses a fairly thick wall, roughened on the outer surface and lined by a delicate membrane on the inner surface. It contains four sporocysts, each having rounded ends. Each sporocyst contains two sporozoites and some residual substance in the form of one or two masses of refractile material. Each sporozoite, with rounded anterior and pointed posterior end, contains an ovoid refractile body in its anterior portion.

Dimensions.—Oöcyst 18-21  $\mu$  in diameter; sporocyst 10 $\mu$  by  $7\mu$ .

SPOR.

Remarks.—This form was originally described as a parasite of the liver of herrings, mackerel and sprats. Wenyon (1915 b) found the ripe occysts in the fæces of a patient returned to England from Gallipoli. Dobell (1919 a), considering it to be a human parasite, named it *E. wenyoni*.



Fig. 81.—Oöcyst of Eimeria clupearum (Thélohan). (After Dobell and O'Connor.)

Fig. 82.—Eimeria faurei (Moussu & Marotel). A, immature oöcyst; B, mature oöcyst. (From Wenyon, after Nöller, Schurjohann, and Verbrodt.)

Knowles (1924) recorded it from the faces of a man at Calcutta. Thomson and Robertson (1926) have shown that the occysts of E. clupearum occur in large numbers in the liver of herrings, mackerel and sprats, and that this is the source of the occysts in human faces.

Habitat.---Fæces of man : BENGAL, Calcutta.

### 105. Elmeria columbæ Mitra & Das-Gupta.

†Eimeria columbæ, Mitra & Das-Gupta, 1937 b, p. 291.

Oöcysts subspherical, tetrasporocystid. Oöcystic residuum present. Sporocyst dizoic.

Dimensions.—Maximum size of occyst  $16.4 \mu$  by  $14.35 \mu$ .

Remarks.—Full description of the species has not yet been published. Tetrasporocystid dizoic condition was observed after keeping the oöcysts in 1 per cent. chromic acid for four days. It is said to differ from E. avium (Silvestr. & Rivolta) in the oöcysts being subspherical, and from E. pfeifferi Labbé in oöcystic residuum being present.

Habitat.—Intestine of the pigeon, Columba livia intermedia Strickland : BENGAL, Calcutta.

### 106. Eimeria cylindrica Ray & Das-Gupta.

†Eimeria cylindrica, Ray & Das-Gupta, 1936 a, p. 345.

Oöcysts cylindrical, measuring  $36 \mu$  by  $18 \mu$ . Oöcystic residuum present.

Habitat.—Rectum of snake, Natrix piscator (Schneid.): BENGAL, Calcutta.

## 107. Eimeria faurei (Moussu & Marotel). (Fig. 82.)

Coccidium sp., Moussu & Marotel, 1901, pp. 1087-9.

Coccidium faurei, Moussu & Marotel, 1902, pp. 82-98, 10 figs.

†Coccidium faurei, Baldrey, 1906, p. 387.

Eimeria faurei, Nöller, Schürjohann & Vorbrodt, 1922, p. 993,
 7 figs.: Wenyon, 1926, pp. 843-4, fig. 363; Reichenow, 1929,
 pp. 945-6; Rastegaïeff, 1930, pp. 379, 399; Kudo, 1931, p. 272.

Oöcysts usually ovoid, but sometimes spherical, and possess a definite micropyle closed by a cap. Four sporoblasts are developed in the oöcyst, and a residual body may or may not be present. Sporocysts possess a micropyle at the more pointed end; a residual body is always left in the sporocyst.

Dimensions.—Oöcyst, ovoid 20-40 $\mu$  by 17-26 $\mu$ ; spherical 18 $\mu$  in diameter; sporocysts 13 $\mu$  by 6 $\mu$ 

*Remarks.*—The parasite is said to cause progressive pernicious anæmia that sooner or later ends fatally.

Habitat.—Intestine of sheep : INDIA (locality not noted).

#### 108. Eimeria flaviviridis Setna & Bana. (Fig. 83.)

†Eimeria sp., Setna, 1933, p. 97, fig. 2; Ray & Das-Gupta, 1935 b. p. 315.

*†Eimeria* ("species B"), Knowles & Das-Gupta, 1935, pp. 703, 705, pl. xxx, figs. 6-9, 17.

†Eimeria flaviviridis, Setna & Bana, 1935 b, pp. 256–60, pls. i–iii; Ray & Das-Gupta, 1937 c, p. 270.

Various developmental stages occur either in the cells of the epithelial lining of the gall-bladder or attached to them, or floating freely in the fluid in the gall-bladder. Schizonts rounded, oval, or irregular in shape and give rise to from 16 to Merozoites are elongate curved bodies, 140 merozoites. with pointed extremities. Microgametocyte is a large ovoid body. Microgametes simple, rod-shaped. Macrogametocyte characterized by a crescentic mass of granules surrounding its nucleus. Occysts colourless, elliptical. Four sporoblasts develop as roundish masses, without the formation of any residual body. Sporocysts ovoid, without a cap: each consists of two valves, applied to one another like two watchglasses, and contains two sporozoites, which are elongated structures slightly longer than the sporocyst, with their ends coiled in. The sporocystal residue is represented by a mass of granules lying in the middle. Development of the sporo-

N 2



Fig. 83.—Eimeria flaviviridis Setna & Bana. A, section of a portion of the gall-bladder showing developmental stages in the epithelial cells (a, schizont; b, merozoites; c, macrogametocyte);
B, schizont, showing repeated division of the nucleus;
C, formation of merozoites, with a large residual body in the middle; D, macrogametocyte; E, microgametocyte;
F, oöcyst with four sporoblasts; G, mature oöcyst containing sporocysts, each with two sporozoites and a residual body. (After Setna and Bana.)

zoites takes place while the oöcysts and sporocysts are floating in the fluid in the gall-bladder. Ööcysts pass into the alimentary canal and are expelled with the fæces, fresh infection taking place by these being ingested.

Dimensions —Schizonts  $12-21 \mu$ ; merozoites  $8-10 \mu$  by  $1\cdot 3-1\cdot 5 \mu$ ; microgametocytes  $16-17 \mu$ ; oöcyst  $17-34 \mu$  in length by  $11-15 \mu$  in width; sporocysts  $7-9 \mu$  by  $5-7 \mu$ ; sporozoites  $9\cdot 13 \mu$  by  $1\cdot 04 \mu$ 

Remarks.—Setna and Bana (1935 b) described this parasite as a new species under the name of E. flaviviridis, and observed both schizogony and sporogony taking place within the gallbladder. Development of the oöcyst is completed in 62 hours within the gall-bladder, or within 54 to 61 hours outside the body of the host under experimental conditions.

Rarely oöcysts containing three sporoblasts and three sporocysts were encountered, but one of these was double the size of the others and developed four sporozoites, the total number of sporozoites being thus the normal number eight.

Knowles and Das-Gupta (1935) have recorded three species from the same host. In their "species B" the oöcysts range in size from  $17-22 \mu$  in length by  $12-15 \mu$  in breadth. They also met with occasional giant forms measuring over  $30 \mu$ in length, and remarked that they possibly corresponded with the species observed by Bana and reported by Setna (1933). The oöcysts reported by Setna and Bana measured  $25-34 \mu$  in length and  $11-14 \mu$  in breadth. It is thus highly probable that *E. flaviviridis* described by Setna and Bana from Bombay and "species B" described by Knowles and Das-Gupta from Calcutta are identical.

The species shows a very close resemblance to *E. agamæ* Laveran & Pettit, 1910, as regards the occurrence of both schizogony and sporogony in the gall-bladder and bile-ducts and in the form and dimensions of the occysts, but differs as regards the form of the sporocysts. In *E. agamæ* the occysts are more or less elongated oval, measuring  $20-25\mu$  by  $11-14\mu$ , and the sporocysts fusiform, measuring  $8\mu$  by  $4\mu$ . In *E. flaviviridis* the sporocysts are ovoid, and measure  $7-9\mu$ by  $5-7\mu$ .

Habitat.—Gall-bladder and intestine of the lizard, Hemidactylus flaviviridis (Rüppell): BENGAL, Calcutta; BOMBAY, Bombay.

109. Eimeria harpodoni Setna & Bana. (Fig. 84.)

†*Eimeria* sp., Setna, 1933, p. 97, fig. 1.

†Eimeria harpodoni, Setna & Bana, 1935 a, pp. 165-9, figs. 1-4.

Oöcysts almost spherical, with four sporoblasts and a large honeycomb-like residual body. Sporocyst elliptical in outline, with the narrow end of the oval projecting in the form of a neck, and bearing a broad, inverted, V-shaped appendage, clearly visible in both living and stained preparations. Each sporocyst contains two sporozoites, and a sporocyst residue is usually present at one end. Sporozoites usually slightly curved, with one end broader than the other.



Fig. 84.—*Eimeria harpodoni* Setna & Bana. *A*, spherical oöcyst with four sporoblasts, from the lumen of the gut; *B*, mature oöcyst with four peculiar-shaped sporocysts each containing two sporozoites and residue, after development eutside the body. (After Setna and Bana.)

Dimensions.—Oöcysts  $9-13 \mu$  in diameter; sporocysts  $9 \mu$  by  $4 \mu$ .

Habitat.—Alimentary canal of the fish popularly known as "Bombay Duck," Harpodon nehereus (Ham.-Buch.): BOMBAY, Bombay.

110. Eimeria hemidactyli Knowles & Das-Gupta. (Fig. 85.)

†Eimeria hemidactyli, Knowles & Das-Gupta, 1935, pp. 703, 705, pl. xxx, figs. 10-13, 18, 19; Ray & Das-Gupta, 1937 c, p. 270.

Oöcysts lemon-shaped. No micropyle could be seen, but the wall of the oöcyst was definitely thinner at the narrow anterior pole than at the posterior end.



Fig. 85.—Eimeria hemidactyli Knowles & Das-Gupta. Lemon-shaped occysts. (After Knowles and Das-Gupta.)

Dimensions.—Oöcysts 17-20·4 µ by 13·6-17 µ. Habitat.—Gut-contents of Hemidactylus flaviviridis (Rüppell): BENGAL, Calcutta.

#### 111. Eimeria kermorganti (Simond). (Fig. 86.)

†Coccidium kermorganti, Simond, 1901 c, pp. 483-5, figs. 1-6. Eimeria kermorganti, Wenyon, 1926, p. 860; Reichenow, 1929, p. 949.

In the young condition the macrogamete is a small, naked, granular sphere, provided with a nucleus and situated in a hostcell. The sphere grows till it reaches a diameter of  $20-22 \mu$ . After undergoing syngamy it secretes a double membrane round itself. The granular mass contracts and divides into four sporoblasts. Oöcyst spherical; contains four oval



Fig. 86.—Eimeria kermorganti (Simond). A, growing macrogamete; B, encysted condition; C, oöcyst containing sporoblasts; D, oöcyst containing four sporocysts; E, fully developed sporocyst; F, schizont dividing into merozoites. (After Simond.)

sporocysts without any residuum. Sporocyst oval; contains two comma-shaped sporozoites, the larger ends of which are situated at opposite poles, and there is a small mass of sporocystal residuum. Schizogony was found in some cells, resulting in a large number of merozoites.

*Remarks.*—It is rare to find a Coccidium in an organ from which there is no exit for the ripe oöcysts. Simond believed that the parasite must be occurring also in other organs connected with the intestine, but could not find them there.

Habitat.—Spleen of the gharial, Gavialis gangeticus (Gmelin), of the River Ganges.

## 112. Eimerla knowlesl Bhatia. (Fig. 87.)

†Eimeria ("species A"), Knowles & Das-Gupta, 1935, p. 701.
 pl. xxx, figs. 1-5, 15, 16.
 Eimeria knowlesi, Bhatia, 1936, p. 177.
 †Eimeria ("species A"), Ray & Das-Gupta, 1937 c, p. 270.

Oöcysts almost spherical, sometimes oval. The mature oöcyst contains four sporocysts, each containing two sporozoites, and does not show any micropyle. Sporocysts show no cap.

Dimensions.—Oöcysts from  $16-20 \mu$  by  $14-18 \mu$ .



Fig. 87.—Eimeria knowlesi Bhatia. A, spherical oöcyst; B, oval oöcyst. (After Knowles and Das-Gupta.)

Remarks.—Knowles and Das-Gupta (1935) remark that the form may be Eimeria raillieti Léger, but it was not possible for them to make certain as they were unable to obtain Léger's paper in India, nor could it be procured even in London. This was due to the reference having been wrongly cited by Wenyon. Coccidium raillicti was originally described by Léger (C. R. Soc. Biol. x1, i (1899), pp. 309-11) from Anguis fragilis Linn., and an abstract of the paper is given in Wiegmann's 'Archiv' (1904, pt. 3, pp. 115-16). The obcysts of E. knowlesi correspond to those of  $\hat{E}$ . raillieti in size, but those of the latter are oval and show a small button-like protuberance at one pole. As, however, the occysts in E. knowlesi are sometimes oval, and the size-range is not markedly different from that of E. flaviviridis (="species B" of Knowles and Das-Gupta, 1935), it is not certain that it is a distinct species.

Habitat.—Gut-contents of Hemidactylus flaviviridis (Rüppell): BENGAL, Calcutta.

## 113. Eimeria koormæ Das-Gupta.

†Eimeria koormæ, Das-Gupta, 1938, p. 155.

Oöcyst spherical, thick-walled, possessing a pseudo-micropyle when mature; without oöcystal residuum. Sporoblasts spindle-shaped; sporocystal residuum present.

<sup>1</sup> Dimensions.—Oöcysts  $14 \mu$  in diameter ; sporoblasts  $10 \mu$  by  $4.5 \mu$ .

*Remarks.*—Full description of the form has not yet been published. It differs from *E. legeri* (Simond), described from the same host-species, in the form of the sporoblasts.

Habitat.—Rectum of the Indian tortoise, Lissemys punctata Bonnaterre : BENGAL, Calcutta.

## 114. Eimeria laminata Ray. (Fig. 88.)

†Eimeria laminata, Ray, 1935, pp. 369-73, pl. x, figs. 1-11.

Young schizonts occur in the intestinal epithelial cells; liberated merozoites are active "grogarinulæ," and show two



Fig. 88.—Eimeria laminata Ray. A, a portion of the transverse section of the small intestine (a, merozoite free in the lumen; b, merozoite entering a cell; c, macroschizogony; d, microgamete formation; e, macrogamete; f. microschizont; g, three merozoites in a cell); B, C, macro- and microschizonts showing laminæ and a siderophilous area containing contractile elements; D, oöcyst showing sporoblasts. E, mature oöcyst. (After Ray.)

kinds of movements, viz., a bending movement and a screwing movement, in which the organism turns rapidly on its long axis and advances with the more pointed end forwards. This anterior end of the body bears on either side a hyaline bladelike structure or lamina. Schizogony is initiated at a very early stage and there are two types of schizonts: (a) macroschizonts, releasing twenty to thirty macromerozoites, which develop into macrogametes, and (b) microschizonts, producing six to eight micromerozoites, which become microgametocytes and give rise to numerous uniflagellate microgametes. The microschizonts are distinguished by the absence of darkly staining granules from their cytoplasm, while the macroschizonts show the granules scattered through the cytoplasm from a very early stage. Sporogony is strictly intracellular. The oöcyst occurs in the intestinal epithelial cell, is spherical, and develops four sporoblasts; there is no oöcystic residuum. The sporocysts are spindle-shaped, and each contains two sporozoites and a residuum. The mature oöcysts are dropped into the lumen of the intestine and pass out in the fæcal matter.

Dimensions.—Young schizont and merozoite  $3\mu$  by  $1.25\mu$  to  $12\mu$  by  $6\mu$ ; očcyst 8–11 $\mu$  in diameter; sporocyst 4.5– $6\mu$  by slightly less than  $3\mu$ .

*Remarks.*—Of the 200 specimens of *Bufo melanostictus* examined only two were found to be infected, and schizogony and sporogony were occurring simultaneously in the same individual.

Habitat.—Small intestine of Bufo melanostictus Schneider: BENGAL, Calcutta.

## 115. Eimeria legeri (Simond). (Fig. 89.)

†Coccidium legeri, Simond, 1901 d. pp. 485-6, figs. 1-6.

Eimeria legeri, Wenyon, 1926, p. 860; Reichenow, 1929, p. 950.

Young macrogametes enter the cells of the liver and grow till they reach a diameter of  $16-18\,\mu$ , when they acquire a thin



Fig. 89.—Eimeria legeri (Simond). A, growing macrogamete; B, encysted condition; C, oöcyst containing sporoblasts; D, oöcyst containing sporocysts; E, fully developed sporocyst; F, sporozoite. (After Simond.)

membrane. Oöcyst spherical, the cytoplasm dividing into four sporoblasts without leaving any residuum. The four sporocysts have clear contents. Each shows one smaller and two larger spheres; the two larger ones represent the larger end of each of the contained comma-shaped sporozoites, and the smaller one a residual body which is granular to start with, but soon becomes transparent and refringent like the sporozoites.

*Remarks.*—The parasite differs from *E. kermorganti* in the smaller dimensions of the oöcysts and in the appearance of the sporocysts. The oöcyst wall is very thin and all the developmental stages are passed within the tissues of the host. The sporocysts fall into the bile ducts and pass out through the alimentary canal.

Habitat.—Gall-bladder and bile-ducts of the tortoise, Lissemys punctata granosa (Schoepff): INDIA.

## 116. Elmeria mitraria (Laveran & Mesnil). (Fig. 90.)

Coccidium mitrarium, Laveran & Mesnil, 1902, p. 613, figs. 12-14.
 Eimeria mitraria, Wenyon, 1926, p. 859, 860, fig. 369, 3-5;
 Reichenow, 1929, p. 949.

Schizonts produce twenty fusiform merozoites with a central nucleus. The microgametocytes produce a number of hairlike microgametes. The oöcysts have a characteristic mitrelike form, the surface of which, unlike those of other Coccidia,



Fig. 90.—*Eimeria mitraria* (Lav. & Mes.). A, oöcyst with four sporoblasts; B, oöcyst containing four sporocysts, each with two sporozoites and a residue. (After Laveran and Mesnil.)

presents four or five ornamental projections. One pole of the oöcyst always bears a single projection, and the other pole, which is truncated at the base, bears three (or four) projections round it. The protoplasm retracts itself from the wall of the oöcyst, becomes spherical, and divides into four sporoblasts without leaving any cystic residue. Each sporoblast gives rise to an ovoid sporocyst containing two sporozoites and a sporal residue.

Dimensions.—Schizonts  $10-12\mu$ ; merozoites  $3-5\mu$  in length; microgametocytes  $10-15\mu$  in diameter; oöcysts  $10-15\mu$  in diameter.

*Remarks.*—All the stages of development are extracellular, but are more or less intimately attached to the epithelial cells of the host. Lavoran and Mesnil searched in vain for the intracellular stages, and remarked that if they exist they must be of a very short duration. They thought it probable that the organism nourishes itself at the cost of the epithelial cells by means of pseudopodia, and that the projections of the oöcyst are their chitinous representations.

Habitat.—Intestine of the tortoise, Chinemys reevesii (Gray) : CEYLON.

### 117. Eimeria najæ Ray & Das-Gupta. (Fig. 91.)

†Eimeria najæ, Ray & Das-Gupta, 1936 b, p. 345; 1937 d, pp. 275-7, pl. viii.

Schizogony results in the formation of eight merozoites, belonging to two types, either spherical or oval in form. One type shows vacuolar cytoplasm and a distinct karyosome



Fig. 91.—*Eimeria najw* Ray & Das-Gupta. A, intestinal epithelial cells showing (a) young male gametocyte, (b) microgamete formation, (c) growing macrogamete; B, macrogamete with micropyle and reserve granules; C, oöcyst showing a buttonlike plug at the micropyle; D, mature oöcyst. (After Ray and Das-Gupta.)

in the nucleus and later develops into female gametes. The other also shows hyalin cytoplasm, but the nucleus contains few chromatin granules besides the karysome, and later gives rise to male gametes. Fully formed female gametocyte shows a large number of reserve granules in the alveoli of the vacuolar cytoplasm, and a micropyle is also visible, but disappears when the oöcyst is formed, a button-like plug being seen instead. Fully formed male gametocyte contains a large number of biflagellate male gametes. Oöcysts oval, thin-

walled. Oöcystie residuum absent. Sporocysts spindleshaped, with a small residue. Segmented and unsegmented oöcysts discharged from the host. Sporulation takes place in 4 to 5 days.

Dimensions.—Male gametocyte  $20-24\,\mu$  in diameter, female gametocyte 23–27  $\mu$  by 16–18 $\mu$ ; occysts 23–27 $\mu$  by 16–18 $\mu$ ; sporocysts,  $12.14 \mu$  by  $6.8 \mu$ .

Remarks.—Some of the occusts showed sporozoites lying free in the occyst, formed without the formation of sporoblasts. This abnormality has been previously described by Wasielewski for E. steidæ and Paracoccidium prevoti.

Habitat.-Epithelial cells of the small intestine of the cobra, Naja naja Linn. : BENGAL, Sunderbans.

### 118. Eimeria piscatori Ray & Das-Gupta.

†Eimeria piscatori, Ray & Das-Gupta, 1936 a, p. 345.

Oöcysts oval; oöcystic residuum present. Sporocysts spindle-shaped.

Dimensions.—Occysts 29-31  $\mu$  in length by 22.5-24.5  $\mu$ in breadth ; sporocysts 14  $\mu$  by 4–6  $\mu$ .

Habitat.--Rectum of the snake, Natrix piscator (Schneid.): BENGAL, Calcutta.

## 119. Eimeria sardinæ (Thélohan). (Fig. 92.)

Coccidium sardinæ, Thélohan, 1890, p. 1216; Labbé, 1899, p. 69; Minchin, 1903, p. 340.

†Coccidium oxyspora, Dobell, 1919, p. 188, pl. viii, fig. 3.

Eimeria oxyspora, Castellani & Chalmers, 1919, p. 476.

Eimeria sp., Snijders, 1920, pp. 427-32.

Etimeria sp., isinjaers, 1820, pp. 427-92.
 Eimeria snijdersi, Dobell, 1920, pp. 433-6.
 Esmeria oxyspora, Dobell, 1922, p. 1499, fig. 533 D; Hogner & Taliaferro, 1924, pp. 289, 290, 291; Craig. 1926, pp. 355-7, fig. 64; Wenyon, 1926, pp. 851, 852, 861, figs. 350, 368; Knowles, 1928, p. 361, fig. 83, *I*.
 Eimeria snijdersi, Wenyon, 1926, p. 855; Knowles, 1928, p. 361, fig. 92

fig. 83, 3.

Eimeria sardinæ, Wenyon, 1926, pp. 851, 852, 861, figs. 350, 368; Thomson & Robertson, 1926 a, pp. 282-3, 2 figs.; 1926 b, pp. 420-1; Knowles, 1928, p. 362, fig. 83, 3; Reichenow, 1929, pp. 951-2, fig. 918 A; Kudo, 1931, p. 274, fig. 113 m.

Oöcyst spherical, having a faintly yellow transparent wall, composed of at least two distinct layers, and containing four dizoic spores and a small oöcystal residue. Sporocysts long, sharply pointed at both ends, and possessing a tough endospore and deciduous epispore, the remains of which give the spore a frilled appearance. In each spore there are two sporozoites, with pointed anterior and rounded posterior ends, the latter containing the nucleus.

Dimensions.—Oöcyst  $36-52 \mu$  in diameter; sporocysts  $30-32 \mu$  by  $7.5 \mu$ .

**Remarks.**—Dobell (1919) found a Coccidium in a young man who had been in South Africa. India, and Ceylon, and described it as a new species under the name *Coccidium oxyspora*. He could not determine whether the organism was pathogenic, as the patient was also infected with *Entamæba histolytica* and *Ancylostoma*. He further analysed records of over seventy cases of coccidiosis in man. These cases all belong to *Eimeria clupearum* (Thélohan) (=*E. wenyoni* Dobell, 1918) or to *Isospora belli* Wenyon (=*I. hominis* Railliet & Lucet).

Snijder (1920) described another case of human coccidiosis and, as the oöcysts were definitely larger, Dobell (1920) named



Fig. 92.-Eimeria sardinæ (Thélohan). (After Dobell.)

this parasite E. snijdersi. Broughton-Alcock and Thomson (1922), however, in another case of E. oxyspora infection found oöcysts quite as large as those of E. snijdersi, and considered the two species to be identical. Brug (1922 a) suggested that E. snijdersi was of animal origin and ingested with food. Thomson and Robertson (1926 a) made a careful study of the Coccidia of fish and came to the conclusion that the oöcysts of E. sardinæ (Thélohan, 1890), parasitic in the "soft roe" (testis) of herrings, sprats and mackerel, are identical with the oöcysts of E. oxyspora and E. snijdersi. They further (1926 b) proved their contention by giving a man a strong saline aperient and afterwards making him eat a large quantity of soft roe of herring. Next morning he passed a large number of oöcysts of E. sardinæ which were

identical with those found in cases of human coccidiosis. Thus E. oxyspora and E. snijdersi are synonymous with E. sardinæ.

In India Setna and Bana (1935) have studied the coccidial infection in a number of fish, and described the oöcysts and sporocysts from ten different species, but the oöcysts in no case correspond with those of E. clupearum or E. sardinæ.

Habitat.—Alimentary canal of man who had visited South Africa, India, and Ceylon.

## 120. Eimeria smithi Yakimoff & Galonzo. (Fig. 93.)

Coccidium oviforme (part), Guillebeau, 1893, p. 81.

Eimeria zurni (part), Smith & Graybill, 1918, p. 89; Wenyon, 1926, pp. 842-3, fig. 362.

†Eimeria zurni (part), Cooper, 1926 a, p. 290 ; 1926 b, p. 291.

Bovine Coccidia (part), Cooper, 1927, pp. 92-7, pl. x.

Eimeria smithi, Yakimoff & Galouzo, 1927, pp. 185-200, figs. 1-7; Reichenow, 1929, p. 945; Rastegaïeff, 1930, pp. 390-1, fig. 2. †Bovine Coccidia, Sen, 1932, p. 34.

+Eimeria smithi, Ware, 1936, p. 35.

Oöcysts are ovoid, with one pole pointed, have a thin wall, and are provided with a micropyle. They are brownish in colour, and are distinctly larger in size than those of E. zürni. Mature cysts do not show any oöcystic residue. Sporocysts pear-shaped, with a sporocystic residue.



Fig. 93.—Eimeria smithi Yakimoff & Galouzo. (After Yakimoff and Galouzo.)

Fig. 94.—Eimeria wassilewskyi Rastegaïeff. (After Rastegaïeff.) Fig. 95.—Eimeria zürni (Rivolta). (After Yakimoff and Galouzo.)

Dimensions.—Oöcysts  $25-35 \mu$  in length; sporocysts  $10.8-14.4 \mu$  by  $7-9 \mu$ ; sprozoites  $3.6-5.8 \mu$  by  $3-5 \mu$ .

*Remarks.*—Zschokke (1892), Hess (1892), and Guillebeau (1893) were the first to describe coccidiosis of cattle as a distinct disease. Guillebeau noted that in certain years the disease became epidemic and caused considerable mortality. Multiplication occurred in the epithelial cells of both the small

and large intestine. The oöcysts described by him possessed a distinct micropyle. Zublin (1908) studied the development of the oöcyst outside the body and also noted that although the majority of oöcysts were  $12-15\mu$  in diameter, larger forms, which measured  $30-35\mu$  in length by  $20\mu$  in breadth, also occurred. Theobald Smith and Graybill (1918), who investigated coceidial dysentry of calves in America, also encountered oöcysts of two types. The second type, which evidently corresponds with *E. smithi*, were said to be brownish in colour, possessed a thick wall, and measured  $25\cdot8-41\cdot8\mu$  in length by  $16\cdot4-24\cdot6\mu$  in breadth. They did not contain any oöcystic residue, but sporocystic residue was present.

Wenyon (1926) considered it possible that there were two species of *Eimeria* found in cattle, but was led to identify and figure the larger, oval type as  $E. z \ddot{u} rni$ .

Cooper (1924) apparently was the first to record bovine coccidiosis in India. He (1926 a) noted the extreme variation in shape and size of the occysts, but considered these variations to be connected with the rate of multiplication. According to him, in normal "carrier" infection all occysts are well formed, with a strong, thick capsule and with a relatively large amount of dense and granular protoplasm, but very large and extremely small forms are encountered, and display considerable range of variation in shape. At the height of multiplication occysts are small, and they show an almost uniformly ovoid to a nearly round shape, with distinctly thinner capsules. When clinical symptoms occur still smaller oöcysts appear, usually in small numbers; these have an extremely thin capsule and much reduced amount of almost transparent protoplasm. In discussing the pathogenicity of bovine Coccidia, Cooper (1926 b) maintained that a latent or " carrier " type of infection is of almost universal occurrence in cattle in India, and that, although usually innocuous, the parasites are capable, under certain conditions, of overcoming the animal's natural defences and then set up a clinical disease identical with what is described in other countries as "red dysentery." He also observed that clinical coccidiosis also occurs as a sequel to rinderpest, even a mild attack of which may result in an appreciable resuscitation of Coccidia. Later (1927) he again reviewed bovine coccidiosis, but the possibility of the existence of two species did not occur to him.

Yakimoff and Galouzo (1927) surveyed the previous literature on bovine Coccidia and gave a table noting the measurements of oöcysts and other character as given by previous workers, and came to the conclusion that the name *E. zürni* should be restricted to the forms with smaller and spherical oöcysts, while larger and oval oöcysts should be referred to a new species, *E. smithi*.

Sen (1932) recorded a case of coccidiosis in a buffalo calf in the Coorg District, and stated that the parasites comprised two species, one a larger form, of yellow colour, provided with a micropyle and the other a small colourless form without a micropyle. I think the former can be identified as E. smithi and the latter as E. zürni. Ware (1936) has for the first time noted the occurrence in India of E. smithi as such.

Habitat.—Alimentary canal of Bos indicus Linn. (?): UNITED PROVINCES, Muktesar; alimentary canal of Bos bubalus Linn. : SOUTH INDIA, Coorg.

### 121. Eimeria southwelli Halawani. (Fig. 96.)

†Eimeria southwelli, Halawani, 1930 a, pp. 1-3. fig. 1; 1930 b, p. 326.

Oöcysts polymorphic. Immature oöcysts generally peashaped. Mature oöcysts cylindrical or sausage-shaped;



Fig. 96.—*Eimeria southwelli* Halawani. A, an immature pear-shaped oöcyst containing a large spherical zygote; B, an immature oöcyst rotaining its hind-bulb to a slight degree; C, oöcyst containing four sporoblasts; D, fully mature oöcyst containing oval sporocysts. (After Halawani.)

cyst-wall colourless and transparent. A residual mass is usually present in the immature cyst, but often disappears as the cyst matures. Sporoblasts four, arranged lengthwise, end to end, sometimes in pairs or in a chain. Sporocysts oval. *Dimensions.*—Oöcysts 25–50 $\mu$  in length, average 38 $\mu$ ,

Dimensions.—Oöcysts  $25-50 \mu$  in length, average  $38 \mu$ , average width  $12 \mu$ ; sporocysts  $10-12 \mu$  in length.

*Remarks.*—It is presumed by Halawani that the mature oöcysts from the parent pass, via the cloaca, up the oviduct to the uterus, and by liberation of their sporozoites infect the intra-uterine embryos.

Habitat.—Spiral valve of the intestine of the embryo of a shark, Ætobatis narinari Agassiz, from the Indian Ocean: CEYLON, Colombo.

SPOR.

### SPOROZOA.

## 122. Eimeria wassilewskyi Rastegaïeff. (Fig. 94.)

†Eimeria wassilewskyi, Rastegaïeff, 1930, pp. 391-2, fig. 3.

Occysts egg-shaped, flattened at one end, with a distinct micropyle.

Dimensions.—Oöcysts  $18 \mu$  by  $14.4 \mu$ ; micropyle  $4.5 \mu$ .

Habitat.—Alimentary canal of Axis axis Erxl. (=Cervus axis), from INDIA, in the Zoological Gardens at Leningrad.

## 123. Eimeria yakimovi Rastegaïeff. (Fig. 97.)

*†Eimeria yakimovi*, Rastegaieff, 1930, p. 389, fig. 1.

Occysts oval, with distinct micropyle and containing four sporocysts. Sporocysts oval, each containing two sporo-



Fig. 97.-Eimeria yakimovi Rastegaïeff. (After Rastegaïeff.)

zoites, which are pyriform, and lie in a tête-bêche manner. No cystal or sporal residue present.

Dimensions. Occysts  $32\cdot4-41\cdot4\mu$  by  $21\cdot6-28\cdot8\mu$ ; sporocysts 13-19  $\mu$  by 3.5-4  $\mu$ ; sporozoites 11.7-13.6  $\mu$  by 4.5  $\mu$ .

Habitat .- Alimentary canal of the nilgai, Boselaphus tragocamelus (Pall.), from India, in the Zoological Gardens at Leningrad.

## 124. Eimeria zürni (Rivolta). (Fig. 95.)

Coccidium perforans (part), Zürn & Proger, 1877, p. 113.

Cytospermum zürni, Rivolta, 1878.

Coccidium zürni, Railliet & Lucet, 1891, p. 247.

Coccidium perforans var., Labbé, 1899, p. 67.

Eimeria zürni, Jowett, 1911, p. 207; Smith & Graybill, 1918, p. 89; Hegner & Taliaferro, 1924, p. 287; Wenyon, 1926, pp. 842-3.

+Bovine Coccidia, Cooper, 1924, p. 48.
+Eimeria zürnii, Cooper, 1926 a, p. 290.
Eimeria zürni, Yakimoff & Galouzo, 1927, pp. 185-200, figs. 8-15; Reichenow, 1929, p. 945; Rastegaïeff, 1930, p. 399.

†Smaller form, Sen, 1932, p. 34.

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Oöcysts colourless or faintly greenish, almost or quite spherical, without a micropyle, and smaller than those of E. smithi. Mature cysts contain no oöcystal residue, and the sporoblasts, when first formed, are spherical. They soon become ovoid and secrete a sporocyst which has a thick cap at one pole. The sporocysts are completely filled by sporozoites and there is no sporocystal residue.

Dimensions.—Oöcysts  $12-25 \mu$  in diameter ; sporocysts  $9.9-11 \mu$  by  $5.3-5.7 \mu$ .

Remarks.—Zublin (1908) was the first to differentiate the larger oöcysts found more rarely (now identified as *E. smithi*) from the smaller ones which are more commonly found, and which measure 12-25 in diameter. Jowett (1911) gave the measurements as  $14\cdot4-27\cdot2\mu$  by  $12\cdot8-20\cdot8\mu$ . Theobald Smith and Graybill (1918) also described two types of oöcysts, and gave the measurements of the smaller ones as  $13\cdot1-28\cdot7\mu$  by  $12\cdot3-20\cdot5\mu$ .

Habitat.—Alimentary canal of Bos indicus Linn. (?): UNITED PROVINCES, Muktesar; alimentary canal of Bos bubalus Linn.: SOUTHI INDIA, Coorg.

## 125. Eimeria sp.

†Eimeria sp., Cooper & Gulati, 1926, pp. 191-2.

Occysts of an unidentified species of *Eimeria* were found in the fæces of a cow at Tocklai and a heifer at Shillong, in ASSAM.

### 126. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Oöcysts spherical; residual body absent. Sporocysts ovoid, with a small rounded knob-like thickening at one end; sporocystal residue absent.

Dimensions.—Oöcysts  $10.6 \mu$  in diameter ; sporocysts  $5.3 \mu$  by  $3 \mu$ .

Habitat.—Intestine of the fish Trichiurus savala Cuv. & Val. : BOMBAY, Bombay.

### 127. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Oöcysts spherical; residual body occasionally present. Sporocysts small ovoid, with one pole slightly narrower than the other; sporocystal residue absent.

Dimensions.—Oocysts  $7.6\,\mu$  in diameter; sporocysts  $3.4\,\mu$  by  $2.1\,\mu$ .

Habitat.—Intestine of the fish Batrachus grunniens (Bl. & Schn.): BOMBAY, Bombay.
#### 128. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935a, p. 167.

Oöcysts spherical; residual body absent. Sporocysts ovoid; the sporocystal residue in the form of two darkly staining globules.

Dimensions.—Oöcysts  $10 \mu$  in diameter: sporocysts  $5.5 \mu$  by  $3.1 \mu$ .

Habitat.—Intestine of the fish Epinephelus tauvina (Forskal): BOMBAY, Bombay.

#### 129. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Oöcysts spherical; residual body absent. Sporocysts broadly ovoid; sporocystal residue absent.

Dimensions.—Oöcysts  $12 \mu$  in diameter; sporocysts  $7.6 \mu$  by  $6.1 \mu$ .

Habitat.—Intestine of the fish Engraulis mystax (Bl. & Schn.): BOMBAY, Bombay.

#### 130. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Oöcysts spherical, residual body absent.

Dimensions.—Oöcysts  $10.3 \mu$  in diameter.

Habitat.—Intestine of the fish Otolithus ruber (Bl. & Schn.): BOMBAY, Bombay.

#### 131. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Oöcysts almost spherical; residual body absent. Sporocysts ovoid, with one end more pointed than the other; sporocystal residue absent.

Dimensions.—Oöcysts  $8.4 \mu$  in diameter; sporocysts  $4 \mu$  by  $2.5 \mu$ .

Habitat.—Intestine of the fish Sillago sihama (Forskal): BOMBAY, Bombay.

#### 132. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Occysts spherical; residual body absent. Sporocysts ovoid, with frequently a knob at one end; sporocystal residue absent.

Dimensions.—Oöcyst 15–18 $\mu$  in diameter ; sporocysts 8.7 $\mu$  by 5.3 $\mu$ .

Habitat.—Intestine of the fish Coilia dussumieri (Cuv. & Val.): BOMBAY, Bombay.

# 133. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Habitat.—Intestine of the fish Plotossus canius (Ham.-Buch.): BOMBAY, Bombay.

#### 134. Eimeria sp.

†Eimeria sp., Setna & Bana, 1935 a, p. 167.

Oöcysts spherical; residual body absent. Sporocysts ovoid; sporocystal residue absent.

Dimensions.—Oöcysts 11  $\mu$  in diameter ; sporocysts  $4 \cdot 2 \mu$  by  $3\mu$ .

Habitat.—Intestine of the fish Epinephelus diacanthus (Cuv. & Val.): BOMBAY, Bombay.

#### 135. Eimeria (?) sp.

Certain Coccids, Ross, 1898, p. 173; 1906, pp. 102, 104.

Well-defined oval organisms,  $8\mu$  by  $4\mu$  in size, were found lying by the side of the nucleus within the stomach-cells of a few individuals of *Culex* sp. Each possessed a vacuole (?) surrounded by a faint granulation. According to Ross they probably belonged to the Coccidiidæ, but were never seen again by him.

Habitat.—Stomach-cells of Culex sp.: MADRAS, Ootacamund.

# Genus WENYONELLA Hoare, 1933.

Wenyonella, Hoare, 1933, pp. 369-74; Ray, 1935, pp. 112-13; 1937, pp. 117-20.

Oöcysts develop four sporocysts, each containing four sporozoites.

*Remarks.*—Hoare (1933) founded this genus for a species from the small intestine of a snake in Uganda. Ray (1935, 1937) has described a new species from a squirrel.

# 136. Wenyonella hoarel Ray. (Fig. 98.)

†Wenyonella hoarei, Ray, 1935, pp. 112–13, 6 figs.; 1937, pp. 117– 20, pl. vii.

Schizonts more irregular in shape than the sexual forms, and lacking a clear outline. Merozoites in groups of six to eight, escaping from a host-cell into the lumen of the intestine. Two types of merozoites : one with hyaline cytoplasm and a dark-staining granule in the neighbourhood of the nucleus; the other slightly longer, more opaque when stained, and showing numerous dark granules scattered irregularly. On entering new epithelial cells the merozoites assume a round form and become gametocytes, which are differentiated into microgametocytes and macrogametocytes respectively; the former give rise to a large number of biffagellate microgametes round a small cytoplasmic residuum; they then break off from the residuum and swim actively, congregating at one end of the female gamete. The latter increase in size and become macrogametes.

Oöcysts perfectly spherical; tetrasporocystid. Sporocysts show a characteristic lenticular knob at one pole; tetrazoic. Sporozoites more or less regularly arranged, with broad ends of two sporozoites at each pole.

Dimensions.—Merozoites  $6\mu$  by  $2\mu$  or  $8\mu$  by  $2\mu$ ; young gametocytes  $6-8\mu$ ; oöcyst  $14-18\cdot5\mu$  in diameter; sporocysts  $10\mu$  by  $8\mu$ .

Remarks.—In a fresh preparation merozoites were seen actively swimming about with the pointed end directed



Fig. 98.—Wenyonella hoarei Ray. A, oöcyst with developing sporoblasts; B. oöcyst with distinct sporoblasts; C, mature oöcyst showing sporocysts each containing four sporozoites. (After Ray.)

forward. Female gametocyte showed a micropyle which did not persist in the oöcyst. Development of the male gametes was also observed. Advanced male gametes were seen adhering round a central mass of cytoplasm with their actively moving tail-ends directed away from it. Within an hour and a half these gametes broke off and congregated round the micropylar end of the female gamete.

On keeping the occysts in 1 per cent. chromic acid these showed the first sign of development after forty-eight hours, when the sporoblasts were seen to be budding out. They separated from each other on the fourth day, were invested with sporocysts on the fifth day, and the sporozoites were well differentiated on the seventh.

Habitat.—Gut-contents of a squirrel, Sciurus sp.: BENGAL, Calcutta.

# Subfamily BARROUSSIINÆ (Wenyon, 1926).

Oöcysts contain many sporocysts, each containing one or four sporozoites.

# Genus PYTHONELLA Ray & Das-Gupta, 1937.

Pythonella, Ray & Das-Gupta, 1937, p. 2.

Oöcyst develops sixteen sporocysts, each containing four sporozoites.

# 137. Pythonella bengalensis Ray & Das-Gupta.

†Pythonella bengalensis, Ray & Das-Gupta, 1937, p. 2.

Oöcysts spherical. Segmenting oöcysts found in the epithelial cells of the intestine and in the fæces. Oöcyst develops eight primary sporoblasts, and after some time these divide to form sixteen, and are converted into as many sporocysts. Each sporocyst contains four sporozoites and a central residuum. Male gametes are formed as in the family Eimeriidæ. Female gametes often seen lying in the submucosa.

Dimensions.—Oöcysts 25–30  $\mu$  in diameter ; sporocysts 8–10  $\mu$  by 6–7  $\mu$ .

*Remarks.*—Full description of this species has not yet been published. Heccaidecasporocystid and tetrazoic condition marks it out from all known CocciDIA. Segmented or unsegmented oöcysts were seen in the fæcal matter and matured within seven to ten days when kept in 1 per cent. chromic acid.

Habitat.-Intestine of Python sp. : BENGAL, Calcutta.

# Subfamily AGGREGATINÆ Reichenow, 1929 (emend. Hoare, 1933).

Schizogony in one type of host and sporogony in another. Oöcysts contain many sporocysts, each sporocyst giving rise to from two to many sporozoites.

*Remarks.*—Labbé (1899) founded the family Aggregatidæ to include the genus Aggregata. As originally defined by Frenzel, Aggregata was a genus of Gregarines, characterized by sporozoites being formed directly in the cyst round a number of residual masses. The researches of Dobell (1914, 1925) and Pixell-Goodrich (1924) showed that the Aggregatidæ may be safely regarded as COCCIDIA, which have their schizogony in one host (usually a crab) and the sporogony in another (usually a Cephalopod). It is now known that the bodies produced in the cyst are merozoites, and not sporozoites as was formerly supposed.

Reichenow (1929) amended the family and included in it a number of other genera, such as Pseudoklossia Léger & Duboscq, Merocystis Dakin, Myriospora Lermantoff, Caryotropha Siedlecki, and Angeiocystis Brasil, all parasites of marine worms, molluscs and crustaceans, in which the occyst develops many sporocysts (except in Angeiocystis Brasil, in which only four are developed) containing two or more (up to thirty) sporozoites. In some of these genera schizogony is not known, and therefore is presumed to take place in some other host. Hoare (1933), in accordance with his scheme of classification, has transferred Angeiocystis to the subfamily Eimeriinæ, and has retained the others in his subfamily Aggregatinæ. Mackinnon and Ray (1937) have shown that the sporozoan parasite hitherto known as Monocystis thalassemæ Lankester is a coccidian, and have placed it in a new genus, Ovivora, belonging to this subfamily.

#### Genus AGGREGATA Frenzel, 1885.

"Sac à Psorospermies," Van Beneden, 1854, p. 9.

Monocystis sp., Lieberkühn, 1855, p. 9, pl. viii, figs. 9, 12. Benedinia, Aimé Schneider, 1875 a, pp. xl-xlv. Klossia, Aimé Schneider, 1883, pp. 78-104, pls. viii, ix.

Aggregata, Frenzel, 1885, p. 560.

Gregarina, Frenzel, 1885, pp. 572, 576, 578.

Benedinia, Labbé, 1895, p. 381; 1899, pp. 54-5.

Klossia, Labbé, 1896, p. 535, pl. xii, fig. 20; pls. xv, xvi, xviii, figs. 1-12; 1899, p. 54.

Aggregata, Labbé, 1899, p. 6.

Legeria, Blanchard, 1900, p. 159.

Eucoccidium, Lühe, 1902, pp. 771-3.

Legerina, Jacquemet, 1903, pp. 193-4.

Aggregata, Léger & Duboscq, 1906, pp. 1001-3; 1908, pp. 44-108; Moroff, 1906 a, pp. 652-4; 1908, pp. 1-224; Pixell-Goodrich, 1914, pp. 159-74; Dobell, 1914, pp. 1-7; 1925, pp. 1-136; Wenyon, 1926, pp. 870-5, figs. 376-8; Knowles, 1928, pp. 366-8; Reichenow, 1929, pp. 928-31, figs. 894-7; Kudo, 1931, p. 270, fig. 111; Calkins, 1933, p. 566; Reichenow, 1935, p. 373.

Oöcyst contains many sporocysts, each developing three sporozoites. Schizogony takes place in a crab, and sporogony in a cephalopod host.

Remarks.-The life-cycle of A. eberthi Labbé has been studied by a number of workers. The nucleus of the zygote divides repeatedly, and numerous sporoblasts, and finally sporocysts, are developed in the occyst in the body of a cuttlefish. Each sporocyst contains three sporozoites and a residual mass. A crab is infected by eating the infected material passed in the dejecta of a cuttlefish. The sporozoites are liberated in the intestine of the crab and, passing through the lining cells, grow into schizonts, which form cysts that bulge into the body-cavity, and by schizogony produce innumerable



Fig. 99.—Life-cycle of Aggregata eberthi Labbé. The stages above the dotted line occur in the cuttlefish, those below in the crab. R, merozoite swallowed by the cuttlefish; A, undifferentiated parasite in submucous tissue; B, C, D, growth into microgametocyte and production of microgametes; E, F, G, growth into macrogamecyte and fertilization; H, zygote; I, first nuclear division in zygote; J, K, nuclear multiplication in zygote and production of sporoblasts; L, sporocyst containing three sporozoites and small residual body; M, escape of sporozoites in intestine of crab; N-Q, growth of schizont and production of merozoites in subepithelial connective tissue. (From Wenyon, after Dobell.)

merozoites. When the crab is eaten by a cuttlefish, the merozoites penetrate the gut-wall and develop into microand macrogametocytes, and further into gametes. Anisogamy results in zygote formation, and the occysts are passed out and ingested by another crab.

# 138. Aggregata sp.

†Aggregata sp., Setna & Bhatia, 1934, pp. 42-3, fig. 23.

Schizonts spherical or oval, and are enclosed in a cyst. Each cyst has six or seven pear-shaped apertures through which merozoites escape. The merozoites are curved spindleshaped bodies, with a broad central portion and pointed ends; the nucleus consists of deeply staining granules and occupies one end. The merozoites move with sudden jerky or springy movements, and the nuclear end is usually anterior in these movements.

Dimensions.—Schizonts  $193-246 \mu$  in diameter; merozoites  $11-16\,\mu$  in length.

Habitat.-Intestine of the prawn, Parapeneopsis sculptilis (Heller): BOMBAY, Bombay.

# INCERTÆ SEDIS.

# Genus TOXOPLASMA Nicolle & Manceaux, 1909.

Toxoplasma, Nicolle & Manceaux, 1909 a, pp. 369-72; 1909 b, pp. 97-103; Splendore, 1909, pp. 462-5; Carini, 1909, pp. 465-9; 1911 a, p. 518; Ugo Mello, 1910, pp. 359-63; Yakimoff & Kohl-Yakimoff, 1911 a, pp. 617-18; 1912, pp. 1-14; Bonrret, 1911, p. 373; Sangiorgi, 1913, p. 323; 1914, p. 83; Marullaz, 1913, pp. 323-6; Laveran & Marullaz, 1914, pp. 21-5; Castellani, 1914, p. 113; Castellani & Chalmers, 1919, pp. 488-90; Nöller, 1920, pp. 907-18; Hegner & Taliaferro, 1924, pp. 293, 300-2; Brug,

b) 107-18, Heger & Haga, 1925, pp. 232-8.
 Babesia (Toxoplasma), Calkins, 1926, p. 445.
 Toxoplasma, Wenyon, 1926, pp. 1041-50; Knowles, 1928, pp. 463-5; Reichenow, 1929, p. 965; Nöller, 1931, pp. 804-7.

Organism small, elongated, slightly curved, with a central nucleus. Found in the host-cells either singly or in groups resulting from repeated binary fission. When occurring singly they often lie against the nucleus of the host-cell and indent it, thus bearing some resemblance to the leucocytic Hæmogregarines. Parasites of body fluids, leucocytes, and cells of the spleen, liver, kidneys, lungs, etc., of various Vertebrates.

Species have been described from man, monkeys, dogs, gondis, rabbits, rats, guinea-pigs, moles, birds, and snakes.

Remarks.-It is not possible to determine the correct position of Toxoplasma in the scheme of classification till more

#### TOXOPLASMA.

is known about the life-history. Calkins (1926) provisionally regarded it as a subgenus of *Babesia*, while Wenyon (1926) placed it among parasites of doubtful nature. Reichenow (1929) placed the genus as an addendum to the family Eimeridæ, as, according to Nöller, certain stages found in the wall of the alimentary canal of birds may be interpreted as schizonts, microgametocytes, and macrogametes of the type found in that family. Nöller (1931) thinks that they should most probably be placed in the neighbourhood of the Coccidia occurring in the blood. Kudo (1931) and Calkins (1933) have altogether excluded them from consideration.

#### Key to Indian Species.

1 (8).	Schizogony known to occur	2.
2 (7).	Schizonts not differentiated.	3.
3.	Form circular or pointed, $3-7\mu$ ; schizo-	
	peripheral blood or bone-marrow	
	sometimes binary fission. In leuco-	(Kohl-Yak., p. 204,
	cytes of pigeon	T. columbæ Yak. &
4.	Smaller forms ring-like; schizonts	
	showing signs of binary fission. In	[p. 203.
r.	Bulastur	T. butasturis de Mello,
υ.	reproduction by binary fission or	
	schizogony. In smears from internal	[p. 205,
	organs of rabbit	T. cuniculi Splendore,
6.	Form semilunar or oval. In cells of	[p. 204.
	internal organs of dog	T. canis Ugo Mello,
T(2).	form ovoid fusiform or falsiform	
	In leucocytes or eudothelial cells of	In. 206.
	lung of coot	T. fulicæ de Mello,
8 (1).	Schizogony not known, even animal	•
	nature doubtful. Form round, ovoid	
	or pyriform. In blood and spleen of	[lani, p. 207.
	man	1. (1) pyrogenes Castel-

#### 139. Toxoplasma butasturis de Mello.

†Toxoplasma butasturis, de Mello, 1937 a, p. 111.

Small parasites resemble the ring-forms of Plasmodids, larger schizonts with the nucleus better formed and showing evidence of binary fission. Often more than one parasite in the same host-cell.

Remarks.—The organism differs from T. fulicæ in that the cytoplasm is stained uniformly pale rose and does not show two types of coloration suggestive of sexual differentiation. It resembles T. columbæ Yakimoff & Kohl-Yakimoff, of the pigeon.

Habitat.—Leucocytes of Butastur teesa (Franklin): PORTU-GUESE INDIA.

#### 140. Toxoplasma canis Ugo Mello.

Toxoplasma canis. Ugo Mello, 1910, pp. 359-63 : Minchin, 1912, p. 389; Carini & Maciel, 1913, pp. 681-3; Castellani & Chalmers, 1919, p. 489; Boëz, 1921, pp. 479-82; Wenyon, 1926, p. 1047.
 †Toxoplasma canis, Donovan (first recorded in Wenyon, 1926, p. 1357).

In fresh preparations the organisms are transparent, colourless, immobile, semilunar or oval in form, rarely rounded or pyriform. Stained with Giemsa each organism is a clear blue homogeneous mass, surrounded by a transparent and thin envelope. It contains one or (rarely) two chromatic masses representing the nucleus, which are stained reddish-violet; generally rounded, but sometimes irregular, oval or linear in form. The organism occurs singly or in groups of any number up to sixty in endothelial cells, leucocytes, or cells of the spleen or kidney.

Habitat.—In the endothelial cells or cells of the spleen or kidney of the dog : INDIA (locality not cited).

# 141. Toxoplasma columbæ Yakimoff & Kohl-Yakimoff. (Fig. 100.)

Toxoplasma columbæ, Yakimoff & Kohl-Yakimoff, 1912, p. 198. †Hæmogregarina francæ, de Mello, 1915, pp. 93-4, pl. ii, figs. 1-10. †Leucocytogregarina francæ, de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, p. 13. Toxoplasma francæ, Nöller, 1920, p. 914.

Toxoplasma columbæ, Nöller, 1920, p. 914.

Toxoplasma francæ, Wenyon, 1926, pp. 1043, 1047.

Toxoplasma columbæ, do Mello, 1935 a, p. 706 ; 1937 a, p. 111.

Parasite has a circular form when at rest, but slight movements in the interior of the white cell change the form to a



Fig. 100.—Toxoplasma columbæ Yak. & Kohl-Yak. A, young form; B, C, schizogony. (After de Mello.)

pointed one, with the nucleus in the rounded portion. Nucleus in the fresh state strongly refringent, surrounded by such clear protoplasm that it has the appearance of a vacuole. TOXOPLASMA.

Stained by Giemsa the nucleus is coloured a brilliant red; the cytoplasm clear and faintly blue. Small or large forms,  $3-7\mu$  in size. Schizogony in the hepatic cells, more rarely in the peripheral blood and bone-marrow. Sometimes binary division takes place, when rudimentary karyokinetic figures can be demonstrated. Sporogony not known.

Habitat.—Mononuclear leucocytes of the pigeon : PORTU-GUESE INDIA, Mapuça (Bardéz).

#### 142. Toxoplasma euniculi Splendore. (Fig. 101.)

Toxoplasma cuniculi, Splendore, 1909 a, p. 109; 1909 b, pp. 462-5; Carini, 1909, pp. 465-9, figs. 1-14; 1910, p. 167; 1911 a, pp. 518-19; Bourret, 1911, p. 373; Castellani & Chalmers, 1919, pp. 489, 490; Brug, den Heyer, & Haga, 1925, pp. 232-8, pls. i-vi; Wenyon, 1926, p. 1047; Reichenow, 1929 p. 965; Nöller, 1931, p. 805.

†Toxoplasma cuniculi, Krishnan & Lal, 1933, pp. 1049-50, pl. xlv.

Intracellular in the endothelial cells, or leucocytes, or in the body fluids. Crescentic in shape, pointed at both ends; but one end is frequently more pointed than the other. Nucleus central and has a definite karyosome. Occurs singly or in groups. Reproduction by schizogony has also been described.

Remarks.—Krishnan and Chiranji Lal (1933) found this infection in two out of twelve rabbits that had received a course



Fig. 101.—*Toxoplasma cuniculi* Splendore. A, a large cluster of parasites. B, a large monenuclear cell from spleen showing a dividing form and a group of parasites. (After Krishnan and Lal.)

of fifteen injections of Indian ink and colloidal iron, followed by three intravenous injections of a culture of *Leishmania donovani*. Eight weeks after the last injection the two rabbits began to look sickly and lose weight. They suffered from diarrhee and died. Smears from spleen, liver, bone-marrow, and heart-blood showed large numbers of intracellular and extracellular forms of this parasite. The parasite is morphologically indistinguishable from T. gondi. Splendore (1909) was able to infect rats, guinea-pigs, rabbits, and frogs, while Carini (1909) infected pigeons with this parasite.

Habitat.—Smears from liver, spleen, bone-marrow, and heartblood of rabbit, Lepus sp.: BENGAL, Calcutta.

#### 143. Toxoplasma fulicæ de Mello. (Fig. 102.)

†Toxoplasma fulicæ, de Mello, 1935 a, pp. 708-9, pl. ii; 1937 a, p. 111.

Free, or included in endothelial cells or mononuclear leucocytes. Form ovoid, fusiform or falciform in young intracellular stages; large ovoid, occupying most of the host-cell when full grown, but not invading its nucleus. Schizonts show sexual differentiation. In one type the cytoplasm is



Fig. 102.—Tozoplasma fulicæ de Mello. A, B, female schizonts; C, D, division stages; E, F, male schizonts; G-I, division stages. (After de Mello.)

#### TOXOPLASMA.

stained dark blue and the nucleus is a compact chromatic dot, sometimes vacuolated, and surrounded by a white halo: in the other type the cytoplasm is stained light violet, rather rosy, and the nucleus is compact or vacuolated, sometimes constituted by an irregular spirematic thread, never showing the vacuolar space round it. The former are interpreted as female and the latter as male schizonts. Multiplication takes place by repeated binary fission. Merozoites produced also show the same cytoplasmic differentiation as the parent schizonts.

*Remarks.*—The parasite was found only in smears from the lung, and was not found in the peripheral blood or in smears from other organs.

Habitat.—Lung-smears of Fulica atra Linn. : PORTUGUESE INDIA, lakes of Carambolin and Taleigão.

#### 144. Toxoplasma sp.

†A parasite in the sparrow, J. R. Adie, 1908, p. 176.

Toxoplasma avium, Castellani & Chalmers, 1918, p. 489.

Toxoplasma sp., Wenyon, 1926, pp. 1042, 1048; Knowles, 1928, p. 465.

*Remarks.*—Wenyon (1926) regards the form as undoubtedly a species of *Toxoplasma*.

Habitat.—Blood of the sparrow, Passer sp. : PUNJAB.

#### 145. Toxoplasma sp.

†Toxoplasma sp., Plimmer, 1916 a, p. 295.

*Toxoplasma* sp., Wenyon, 1926, pp. 1043, 1048; de Mello, 1937 *a*, p. 111.

Habitat.—Blood and exudation from the lungs of the chat, Saxicola caprata Linn., from India, in the Zoological Gardens, London.

# DOUBTFUL SPECIES.

#### 146. Toxoplasma (?) pyrogenes Castellani, 1914. (Fig. 103.)

†Toxoplasma pyrogenes, Castellani, 1914 b, pp. 113-14, 2 pls.

Some unknown protozoon, Plate, 1914.

Contaminating organisms of a vegetable nature, Wenyon, 1923, pp. 527-50, 855, 1 pl.; 1926, p. 1048, fig. 433, A-C.

Toxoplasma pyrogenes, Knowles, 1928, p. 465; Reichenow, 1929, p. 965

Intracellular, round, ovoid or pyriform in shape, and about  $6\mu$  in diameter. A second form,  $12\mu$  in diameter, and containing several chromatin granules, has been described as a schizont.

Remarks.—Wenyon (1923) discussed the probable nature of this form and came to the conclusion that what had been described as T. pyrogenes was only a contaminating organism of a vegetable nature. Knowles (1928) and Reichenow (1929) support this view. Knowles has also been struck



Fig. 103.—Toxoplasma (?) pyrogenes Castollani. A-D, small forms; E, large form described as schizont. (From Wenyon, after Castollani.)

with the resemblance which some of the forms depicted by Castellani bear to the breaking-down malarial parasites encountered in spleen-puncture films from cases of chronic and relapsing malaria.

Habitat.-Blood and spleen of a man : CEYLON.

# III. Order HÆMOSPORIDIA Danilewsky.

The HÆMOSPORIDIA are Coccidia-like forms specially modified for parasitic life in the blood. There is alternation of hosts, asexual reproduction or schizogony taking place in the blood of Vertebrates and sexual reproduction or sporogony in the alimentary canal of some blood-sucking Invertebrates. They are minute, usually intracellular parasites of red bloodcorpuscles, showing motile amœboid forms in their schizogonous cycle in the Vertebrate host. Gametocytes and dimorphic gametes are formed, as in the Coccidia; but the microgametes have no flagella as a rule and move like spirochotes, fertilizing a spherical macrogamete in the body of the Invertebrate host. The zygote is motile, and is known as an oökinete; after becoming encysted it gives rise to a large number of naked sporozoites, which are introduced into the blood of a Vertebrate host. As they do not pass any stage of their life-history outside the body of a host, the sporozoites are not enclosed within a resistant membrane.

Vertebrates of all classes—mammals, birds, reptiles, amphibians, and fish—are parasitized by different species. A number of them are known to occur in man and cause malaria, which works such havoc in India and other tropical countries. The Arthropods were their primary hosts, and they became secondarily introduced into the Vertebrates and adapted to a parasitic mode of life in the blood of the latter.

The HEMOSPORIDIA are divided into four families.

# Identification Table of Families.

1 (4).	Parasites form hæmazoin pigment (with	
	the exception of Leucocytozoon)	2.
2 (3).	Schizogony in the endothelial cells of the	
• •	blood-vessels of internal organs of	
	Vortebrates. Gametocytes in the peri-	
	pheral blood-corpuscles	Hæ
3 (2).	Schizogony in the peripheral blood of	
• • •	Vertebrates	Pla
4(1).	Parasites do not form hæmazoin pigment.	5.
5 (6).	Schizogony in the endothelial cells of the	
• /	blood-vessels of Vertebrates; finally the	
	parasites invade the red corpuscles,	
	within which they occur as round, ovoid,	
	rod-like or irregular forms. Show no	
	tendency towards a naired arrangement	The
	pointer of the of point of the	~ ~~ ~

SPOR.

{Doflein, p. 210. Hæmoproteidæ [p. 244. Plasmodlidæ Mesnil, 5.

{p. 294. Theileriidæ Du Toit, P 6 (5). Schizogony in the red blood-corpuscles of Vertebrates, with division into two or four; of varying size and shape, and have a tendency to arrangement in couples of pear-shaped individuals .....

[p. 301. Babesiidæ Poche,

# 1. Family HÆMOPROTEIDÆ Doflein, 1916.

Schizogony takes place in the endothelial cells of the bloodvessels of Vertebrates. Certain merozoites penetrate into the circulating red blood-cells, in which they develop into gametocytes. If the blood is taken up by a specific blood-sucking Invertebrate host, the gametocytes develop into gametes, which unite to form the zygotes, and the latter undergo sporogony, as in the family Plasmodiidæ.

Remarks.—It is important to remember that the schizogony cycle can only be observed in sections or smears from the internal organs of the host. In the absence of definite know-ledge regarding the occurrence of schizogony in the internal organs, the generic position can only be inferred from the morphology of the parasites themselves and their occurrence in relation to the nucleus of the infected corpuscle. In Hæmoproteus the organism is halter-shaped and grows round the nucleus without displacing it, whereas in Proteosoma (family Plasmodiidæ) the nucleus of the red blood-corpuscle is ordinarily pushed to one side by the invading organism.

Stiles (1925) gives "oökinete not known to encyst" as the character which serves to distinguish the family Hæmoproteidæ from the family Plasmodiidæ. This statement appears to be based on the earlier work of Aragão. Helen Adie (1915) followed the complete development of the oökinete of *Hæmoproteus columbæ* in the fly *Lynchia maura*. She confirmed that work in 1924, and Aragão (1927) also confirmed it. Numerous oöcysts were found in the wall of the stomach of the fly, and the various stages of development were found to resemble closely those of malarial parasites in mosquitoes.

# Key to India Genera.

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#### Genus HÆMOPROTEUS Kruse, 1890.

(Syn. HALTERIDIUM Labbé, 1894.)

Hæmoproteus, Kruse, 1890, p. 371.

Laverania (part), Grassi & Feletti, 1890, p. 463.

Hæmamæba (part), Grassi & Feletti, 1891, p. 463.

Halleridium, Labbé, 1894, pp. 129, 151; 1899, pp. 78-9; Minchin, 1903, pp. 257-9, 267-9; Stephens & Christophers, 1904, pp. 319-20.

Laverania (part), Laveran, 1899, pp. 603-6.

Hæmocystidium, Castellani & Willey, 1905, pp. 84-5.

 Haemogratum, Gathin, 1912, pp. 365-9; Prowazek, 1912, pp. 566-88, ; Castellani & Chalmers, 1919, pp. 518-26; Wenyon, 1926, pp. 885-902; Knowles, 1928, pp. 372-8; Reichenow, 1929, pp. 972-9; Kndo, 1931, p. 288; Calkins, 1933, p. 566; Reichenow, 1935, p. 375.

The parasites grow in the endothelial cells of the bloodvessels of various organs into fairly large multi-nucleated schizonts, which then break up into very numerous merozoites. It is possible that some of them enter other endothelial cells and again become schizonts, or, entering the red bloodcorpuscles, grow into gametocytes. They produce hæmozoin pigment granules at the expense of the hæmoglobin of the host-cells. The fully-formed gametocyte encircles the nucleus of the red blood-corpuscle like a halter (hence the name *Halteridium* Labbé) and usually does not force it out of place. Parasitic in birds and reptiles. Sporogony takes place in the body of an Arthropod.

Remarks.---Danilewsky (1889) was the first to record the parasites referable to this genus. Labbé (1899) considered that the halteridia of different birds belong to one species, to which he restricted the name H. danilewskyi (Grassi & Feletti). They are now known from hundreds of different species of birds, and many species are recognized. They have often been confused in the past with another pigmented parasite of the blood of birds which belongs to the same family as the human malarial parasite. Researches of Danilewsky, Laveran. Kruse. Grassi, Feletti, and others have shown the difference between Hæmoproteus (Halteridium) and Proteosoma, viz., that schizogony takes place in the endothelial cells in the former and only gametocytes are found in the red blood-corpuscles. while in the latter schizogony takes place in the red bloodcorpuscles and thus both merozoites and gametocytes can be found in them.

It was in the *Hæmoproteus* in the blood of the crow that MacCullum (1897) demonstrated for the first time that the so-called "flagellating body" was the male gametocyte, and the process of ex-flagellation produced the male gametes, which eventually fertilized female gametes and led to the formation of motile zygotes or oökinetes. Coatney (1936) has catalogued 45 species of *Hæmoproteus* and has given a complete list of 650 avian and 22 reptilian hosts from which they had been recorded up to the time of his compilation.

# 147. Hæmoproteus antigonis de Mello.

†Hæmoproteus antigonis, de Mello, 1935 b, p. 471; 1937 a, p. 100.

Female gametocyte with alveolar cytoplasm, stained blue with Lieshman's stain; nucleus compact, oval, slightly subcentral, stained rose, lodged in a vacuole. Male gametocyte pale violet; nucleus granular, central. Pigment coffee-brown, scattered over the body; in the female gametocyte often in clusters or in two large masses united by a batonette.

*Remarks.*—The parasite may possibly be the same as that recorded by Scott (1926) from *Antigone antigone* (Linn.).

Habitat.—Blood of Anthropoides virgo (Linn.): PORTUGUESE INDIA, Junagad.

#### 148. Hæmoproteus asturis de Mello.

*†Hæmoproteus asturis dussumieri*, de Mello, 1935 b, p. 469; 1937 a, p. 100.

Female gametocytes with alveolar cytoplasm, stained light blue with Leishman's stain, nucleus pale rose. Male gametocyte colourless, nucleus more conspicuous. General form oval, especially in male, rarely halter-like in female. Nucleus sometimes subcentral. Pigment granules dark brown, of different sizes, irregularly scattered in both kinds of gametocytes.

*Remarks.*—De Mello has given to this form a trinomial designation to avoid confusion with the unnamed *Hæmoproteus* of *Astur plumbarius* recorded by Wasielewsky in Germany in 1908.

Habitat.—Blood of Astur badius dussumieri (Temm. & Lang.): PORTUGUESE INDIA, Corlim (Ilhas).

# 149. Hæmoproteus cerchneisi, sp. nov.

†Hæmoproteus sp., de Mello, 1935 b, p. 472 ; 1937 a, p. 100.

Female gametocyte halter-like, cytoplasm alveolar, stained greyish-blue with Leishman's stain; nucleus generally round and compact, but often quadrangular and consisting of a central mass with appendages all round, subcentral on the convex border of the parasite, stained light red and lodged in a vacuole : pigment coffee-brown with olive tone, granules or clusters irregularly distributed. Male gametocyte oval or round, and causing a sort of hernia on the periphery of the red cell; cytoplasm colourless or light violet; nucleus subcentral or central, small, elliptic or irregular, containing one chromatic dot fairly visible, surrounded by the rest of the chromatic substance which is hardly visible, stained pale rose; pigment coffee-brown with olive tone, situated at the poles.

Remarks.—Unnamed species of Hæmoproteus have previously been recorded from several species of Cerchneis in different parts of the world. According to de Mello, Wasielewski and Wülker (1918) recorded the parasite of this bird in Europe as H. danilewskyi var. tinnunculus, and not having access to that paper he has refrained from giving his form a specific name. On comparing his description with Wasielewski and Wülker's figure, as reproduced by Reichenow, the nucleus of the microgametocyte in var. tinnunculus is large and does not correspond with the description as given by de Mello. I have, therefore, named the parasite found by de Mello as H. cerchneisi.

Habitat.-Blood of Cerchneis tinnunculosus objurgatus Stuart Baker : PORTUGUESE INDIA, Praganã.

#### 150. Hæmoproteus columbæ Celli & San Felice. (Figs. 104, 105.)

Hæmoproteus danilewskii (part), Kruse, 1890, p. 359.

Hæmoproteus columbæ, Celli & San Felice, 1891, pp. 517-18, 541-8, pl. vi; Ed. & Et. Sergent, 1907, pp. 264-74, pls. vi, vii.

Halteridium columbæ, Aragão, 1908, pp. 154-67, pls. xi-xiii.

Hæmcproteus columbæ, Minchin, 1912, pp. 365-6, fig. 157.

Hæmoproteus sp., Sergent & Beguet, 1914, pp. 21-3.

*Hæmoproteus* sp., Acton & Knowles, 1914, pp. 663-90, pls. xlvii-li ;

Alcock, 1914, p. 584; Helen Adie, 1915, pp. 671-80. Hæmoproteus columbæ, Castellani & Chalmers, 1919, pp. 525-6, figs. 185, 186; Helen Adie, 1924, pp. 605–13, 2 pls.; 1925, pp. 9–15, 5 figs.; Wenyon, 1926, pp. 886, 888–96, 1314, figs. 383–6; Knowles, 1928, pp. 372–7, figs. 87, 88; Reichenow, 1929, pp. 973–5, 978, figs. 938–40, 942; Kudo, 1931, p. 288, fig. 122 e, f; Ruiz Martinez, 1934, pp. 96–8; Coatney, 1936, p. 88; de Mello, 1937 a, p. 99.

Schizogony takes place in the endothelial cells of the bloodvessels of various organs of the pigeon, particularly the lungs. The youngest schizonts are minute cytoplasmic bodies with a single nucleus within the cytoplasm of an endothelial cell (fig. 104, P). Growth, nuclear multiplication, and segmentation into fifteen or more small uninucleated unpigmented masses take place (fig. 104, Q). Each of these cytomeres grows; its nucleus undergoes repeated division till the host-cell, now considerably hypertrophied, is filled by a number of multinucleate bodies, each of which is surrounded by a fine membrane. Within this membrane the multinucleate cytomere divides into an enormous number of merozoites (fig. 104, R-W). The endothelial cell finally breaks down and the merozoites escape into the blood-stream. It is possible that some of them



Fig. 104.—Life-cycle of  $Hxmoproteus\ columbx$  Celli & San Felice. The stages above the dotted line occur in the pigeon, those below in the fly.  $A_1-C_1$ , growth of female gametocyte in red blood-corpuscle;  $A_2-C_2$ , growth of male gametocyte and escape from cell;  $D_2$ ,  $E_2$ , rounding off of male gametocyte and escape from cell;  $D_2$ ,  $E_2$ , rounding off of male gametocyte and order to do obtain the stomach-wall of Lynchia maura; M, young occyst on stomach-wall of Lynchia maura; N, mature occyst filled with sporozoites, which eventually enter the salivary glands of the fly and are thence injected into the pigeon; O, eporozoite entering an endothelial cell of a blood-vessel of the pigeon; P, growth of the eporozoite in a mononuclear cell; Q, primary schizogony into a number of uninucleate bodies; R-V, each uninucleate body increases in size and becomes multinucleate; W, segmentation into numerous minute young gametocytes which enter red blood-corpuscles. (From Wenyon, after Aragão and Adie.)

enter other endothelial cells and again become schizonts. Others enter the red blood-corpuscles and are seen as minute cytoplasmic bodies with a single nucleus and often a vacuole (fig. 104,  $A_1$ ,  $A_2$ ). Sometimes as many as a dozen young forms may be present in a single corpuscle, and thus in dry films, by an approximation and overlapping of the parasites, falsely suggest a schizogony. The corpuscles with such multiple infections generally die, so that it is rare to find even two fully developed gametocytes in a single corpuscle. The young



Fig. 105.—Development of Hæmoproteus columbæ Celli & San Felice on stomach-wall of Lynchia maura. A, flattened stomach showing numerous oöcysts of various sizes (× c. 72); B, edge of stomach more highly magnified, showing mature and immature oöcysts with pigment granules (× c. 450); C, intact oöcyst and ruptured oöcyst, with discharged sporozoites and residual cytoplasm (× c. 600); D, free sporozoites. (From Wenyon, after Adie.)

gametocyte becomes elongate, and granules of brown or black pigment appear in the cytoplasm (fig. 104, B, C): it grows round one side of the nucleus, pushing it to one side of the cell, but the latter retains its original shape and size. The fully developed gametocyte is of an elongate sausage-shape, and almost completely encircles the nucleus : it shows a central nucleus and a number of pigment granules distributed through its cytoplasm. Gametocytes are distinguishable as male and female. Male gametocyte possesses a hyaline cytoplasm, staining a pale blue or pinkish colour with Romanowsky stains, and a rather larger nucleus consisting of a membrane enclosing a number of fine chromatin granules (fig. 104,  $B_2, C_2$ ). Female gametocyte has a denser cytoplasm, stains more deeply blue, and possesses a more compact nucleus (fig. 104,  $B_1, C_1$ ).

Further development of the gametocytes takes place in the fly, Lynchia maura. The male gametocyte produces male gametes by exflagellation, the female gametocyte gives rise to a single female gamete, and fertilization produces a zygote, which is a motile vermicule or oökinete. The oökinetes penetrate the hinder portion of the mid-gut of the fly and produce pigmented oöcysts on the outer surface of the wall. The mature oöcyst (fig. 105, B) measures about  $36 \mu$  in diameter, and gives rise to a very large number of sporozoites (fig. 105, C, D), measuring up to  $10 \mu$  in length. These are set free by the rupture of the cysts, invade the salivary glands of the fly, and are reintroduced into the body of the pigeon.

*Remarks.*—Acton and Knowles (1914) studied the development in the pigeon and concluded that schizogony takes place only in the lung, but other observers have shown that it takes place in the endothelial cells of the blood-vessels of other organs as well. Clean pigeons exposed to infection by infected flies show young gametocytes in the blood in about four weeks, during which schizogony takes place a number of times.

Ed. and Et. Sergent were the first (1906) to transmit the infection experimentally to pigeons in Paris by means of infected flies (Lynchia maura), received from Algiers. The asexual cycle as occurring in the endothelial cells was first described by Aragão (1908). Helen Adie (1915), working in the Punjab, was the first to follow the sporogony of the oökinetes in L. maura, and later, working at Algiers (1924, 1925), confirmed her previous work in India. According to her researches, flies which have lived on infected pigeons for ten to twelve days, during which they fed daily, showed all stages of development of the parasite from the oökinete to the sporozoites in the salivary glands.

De Mello and de Sá (1916) have described a process of schizogony in *Hæmoproteus columbæ* taking place in schizonts which were originally in the red blood-corpuscles and then became free in the plasma. If their observations were correct, they must have been dealing with a species of *Proteosoma*. These observations are recorded under *Proteosoma columbæ* (de Mello & de Sá).

Habitat.—Blood of the common pigeon, Columba livia Gmelin, and body of Lynchia maura Bigot : PUNJAB, Amballa ; BENGAL, Calcutta.

#### 151. Hæmoproteus coraciæ de Mello & Afonso. (Fig. 106.)

*Hæmoproteus coraciæ benghalensis*, de Mello & Afonso, 1935, pp. 67-8, pl. i ; de Mello, 1937 a, p. 100.

Sexual dimorphism of gametocytes shown by tinctorial reactions of cytoplasm, which stains blue in femile gametocytes and is colourless or slightly yellowish-blue in male gametocytes. Young female gametocytes small, more or less ovoid; when full grown are typical halteridia, embracing the



Fig. 106.—Hæmoproteus coraciæ de Mello & Afonso. A-C, female gametocytes; D, free female gametocyte; E-G, male gametocytes; H, free male gametocyte. (After de Mello and Afonso.)

nucelus of the red cell and displacing it to the periphery. These halteridia may be pointed, with a tail-like appendage, but when full grown they are broad, regular, lodging the nucleus of the host-cell in their concavity; cytoplasm alveolar, not staining uniformly, the blue colour more pronounced at the poles and lighter in the centre, and showing violet rings in some specimens; nucleus compact or more or less granular; pigment yellow-brown, in minute granules or big dots, showing a tendency to collect at the poles. When free the female gametocytes are roundish or oval, with blue cytoplasm, nucleus compact or irregular, pigment irregularly scattered over the body in clusters or isolated granules. Male gametocytes show the same form as the female, a little more irregular in young stages : nucleus in the form of an irregular spireme. Pigment has the same appearance, but is more definitely collected at the poles. When free the male gametocytes are round, the cytoplasm showing a slight violet-bluish tone. Infected cells are slightly hypertrophied.

Remarks.—Hæmoproteus has been previously recorded from Coracia indica Linn. by Plimmer (1912, 1914) and from other species of Coracia from other parts of the world, but none of these appears to have been given a specific name. De Mello rejects the idea that all Hæmoproteids from birds in general are identical with H. danilewskyi.

Habitat.—Blood of Coracia benghalensis benghalensis Linn. : PORTUGUESE INDIA, Corlim (Ilhas).

#### 152. Hæmoproteus corvi, nom. nov. (Fig. 107.)

- *†Hxmoproteus* sp., Donovan, 1904; Castellani & Willey, 1905, p. 385, pl. xxiv, fig. 5; p. 400; Dobell, 1910, p. 71.
- †Hæmoproteus du Corvus macrorhynchus, de Mello, de Sá. de Sousa, Dias, & Noronha, 1917, pp. 10-11, pl. i, figs. 45-71; de Mello, 1937 a, p. 100.



Fig. 107.—Hæmoproteus corvi, nom. nov. A, female gametocyte; B, male gametocyte; C, so-called "Acton body," showing schizogony. (After de Mello, de Sá, de Sousa, Dias, and Noronha.)

Young parasites small, lanceolate. Female gametocyte halter-shaped, stained blue by Leishman's stain. Nucleus a large chromatic mass, stained deep red, situated usually in the concavity of the halteridium, nearer one pole than the other, and quite close to the nucleus of the infected corpuscle; pigment collected at one pole, but in the fully developed gametocyte may occur at both poles. Male gametocyte ovoid or halter-shaped, stained but little, almost yellowish; nucleus contains two or three chromatin granules, and is situated near one pole, while the pigment is situated near the other.

When fully developed the nucleus may be vesicular or compact and linear; sometimes the pigment may occur near both the poles also. Schizogony takes place in the lung and not in the peripheral blood. It is very similar to schizogony as described in H. columbæ. In the intracorpuscular phase the schizonts are small, medium-sized or large, and in the extradorpuscular phase the "Acton body " shows a single nucleus or, as the result of repeated division, forms a number of nuclei leading to the formation of as many merozoites.

Remarks.—De Mello and his colleagues referred the parasite to the genus Hæmoproteus, but described schizogony as taking place in "Acton bodies" that have come out of the bloodcorpuscle. As, however, they state that schizogony takes place in the lung (presumably in the blood-vessels), it is conceivable that the so-called "Acton bodies" may have been endothelial cells of the blood-vessels that are being carried along in the plasma. The form is consequently retained in the genus Hæmoproteus pending further investigation.

Habitat.-Blood and smears from the lung of the erow, Corvus levaillanti macrorhynchus Blanf. & Oates: CEYLON; PORTUGUESE INDIA, Nova Goa; and the Indian house-crow, Corvus splendens Vieill. : INDIA (locality not cited by Donovan); CEYLON ; PORTUGUESE INDIA, NOVA GOA.

# 153. Hæmoproteus danllewskyl (Grassi & Feletti). (Fig. 108.)

Hæmoproteus (part), Kruse, 1890, p. 359.

Laverania danilewskyi, Grassi & Feletti, 1890, p. 463.

Halteridium danilewskyi, Labbé, 1899, p. 79, fig. 145; Minchin, 1903, pp. 266, 268, 269, 347, 348, 349.

*Halteridium danilewskyi*, Stephons & Christophers, 1904, pp. 319-21, fig. 67; Castellani & Willey, 1904, pp. 83-4, figs. 7-9. Hæmoproteus danilewskyi, Minchin, 1912, p. 365. †Hæmoproteus danilewskyi, Plimmer, 1912, pp. 416, 417; 1913,

p. 148; 1914, p. 189; 1915, p. 130; 1916, p. 85; 1917, p. 32. Hæmoproteus danilewskyi var. tinnunculus, Wasielewski & Wülker, 1918, p. 115.

Hæmoproteus sp., Wenyon, 1926, p. 1383.

Hæmoproteus danilewskyi, Reichenow, 1929, pp. 975-7, figs. 943-6; Coatney, 1936, p. 88.

*†Hæmoproteus* sp., de Mello, 1937 a, p. 99.

Young oval stages of the trophozoite present, though not common. Two kinds of fully developed gametocytes (referred to as trophozoites by the discoverers) present in approximately equal numbers. The female gametocytes are stained distinctly blue with Leishman's stain, leaving a clear tract in the centre, and with pigment granules scattered more or less throughout the cytoplasm. The male gametocytes are shorter and stouter, appearing nearly white or very faintly bluishwhite, owing to greater density of cytoplasm, and the pigment

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granules are aggregated at the two ends. The male becomes shorter, thicker, and finally nearly round. In one instance a double infection of a blood-corpuscle by the two kinds of gametocytes was seen.

Remarks.—Wasielewski and Wülker (1918) described from the kestrel a parasite which they named *H. danilewskyi* var. tinnunculus, accepting the view that many species of birds can harbour the same species of parasite. They point out that in kestrels the infection is either acute or chronic. Acute infection occurs in young birds, which are heavily infected and show schizonts in the various organs. When this subsides chronic infection supervenes and is characterized by relapses



Fig. 108.—Hæmoproteus danilewskyi var. tinnunculus Was. & Wülk. A, female gametocyte; B, male gametocyte; C, double infection with both types of gametocytes. (After Wasielewski and Wülker.)

of a milder type extending over several years. Different stages in the life-cycle are fully described by them.

Habitat.—Blood of the scops owl, Otus bakkamæna bakkamæna Pennant: CEYLON, Colombo. Also in the blood of the following birds from India in the Zoological Gardens, London :—

Anas (Fuligula) baeri Radde (Bayer's pochard).

Kittacincla macroura Blanf. & Oates (shama).

Copsychus saularis (Linn.) (Indian dial-bird).

Coracia benghalensis indica Linn. (Indian roller).

Garrulax albigularis Gould (white-throated jay-thrush).

Glareola pratincola Linn. (pratincole).

Garrulus lanceolatus Vigors (jay).

Melophus melanicterus (Gmelin) (crested black bunting).

Mesia argentauris Hodgs. (silver-eared babbler).

Nettapus coromandelianus Gmelin (cotton-teal).

Propasser rhodochrous (Vigors) (finch).

Prunella strophiata jerdoni Brooks (=Tharrhaleus jerdoni Blanf. & Oates) (Jerdon's hedge-sparrow).

# 154. Hæmproteus glaun cidii de Mello.

111 Hamproteus glaucid & ii, de Mello, 1935 b, p. 473 ; 1937 a, p. 100.

acuse gametocyte<sup>17</sup>, vacualated, stained deep blue with "Yman's stain", "ucleus spherical and central or elongate "structured son the convex border, stained rose; pigment scaere" u. "Male gametocyte almost unstained; nucleus large, without definite borders, containing irregular chromatic masses, subcentral in halter-like forms, central in the rounded ones. Infected red cell hypertrophied, with its nucleus retaining its position when the corpuscle contains a female gametocyte, and displaced when there is a male gametocyte.

Remarks.--De Mello thinks that perhaps the parasite is the same as the unnamed Hæmoproteus of Glaucidium perlatum recorded by A. & M. Léger (1914) in Niger, or perhaps it is only a new variety of H. noctuæ Celli & San Felice (1891).

Habitat.—Blood of Glaucidium radiatum Tickell : PORTU-GUESE INDIA, Canacona.

#### 155. Hæmoproteus gymnorhidis de Mello.

+Hæmoproteus sp., Plimmer, 1913, p. 148.

†Hæmoproteus gymnorhidis, de Mello, 1935 b, p. 474; 1937 a, p. 100.

Female gametocytes halter-shaped, very slender, larger at the poles, cytoplasm alveolar, stained deep blue with Leishman's stain, more deeply at the poles; nucleus central or subcentral, rod-like, oval or bilobed; pigment in clusters of large granules at the centre and at the poles, in the latter situation sometimes as a fine dust, rendering more intense the dark staining of the cytoplasm in this part. The pigment spreads a little round the granules, diffusing into the cytoplasm. Male gametocyte halter-shaped or oval, not stained or slightly blue; nucleus as in the female gametocyte. Pigment granules very minute, mostly at the poles. Infected red cell hypertrophied.

Remarks.—The parasite is probably the same as the unnamed Hæmoproteus from the Indian Gymnoris flavicollis recorded by Plimmer (1913), as Gymnoris flavicollis is a synonym of G. xanthocollis.

Habitat.—Blood of the yellow-throated fineh, Gymnoris xanthocollis Burton : PORTUGUESE INDIA, Praganã ; also from a specimen of the same species in the Zoological Gardens, London.

#### 156. Hæmoproteus herodiadis de Mello. (Fig. 109.)

*Hæmoproteus herodiadis*, de Mello, 1935 a, pp. 351-2, pl. xlii, fig. 2; 1937 a, p. 101, pl. 1, fig. 1.

Young stages roundish oval, fully developed halteridia never embracing the nucleus of the red cell. Fusiform or irregularly bent forms also occur. Sexual dimorph only visible in the tinctorial reaction's. The nucle metocyces Romanowsky stains, it being light blue ind. In one iplastace gametocytes and colourless, almost white, by the two kinds of ?



Fig. 109.—Hæmoproteus herodiadis de Mello. A, fomale gametocyte; B, male gametocyte. (After de Mello.)

Brownish-black pigment irregularly distributed in both. Infection heavy.

*Remarks.*—De Mello thinks that the form is perhaps similar to that found in *Herodias alba* in the Belgian Congo by Rodhain, Pons, Vandenbranden, and Bequaert (1913).

Habitat.—Blood of the heron, Egretta intermedia intermedia (Wagler): PORTUGUESE INDIA, lake of Carambolim.

# 157. Hæmoproteus kopki (de Mello). (Fig. 110.)

*Hæmocystidium kopki*, de Mello, 1916, pp. 8-10, pl. i, figs. 1-17; de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, p. 13.

Hæmoproteus simondi, Wenyon, 1926, p. 902. †Hæmoproteus kopki, de Mello, 1934 a, p. 378; 1934 b, pp. 1785-

99, pl. i.

Hæmoproteus kopki, Coatney, 1936, p. 88.

Young stages round and non-pigmented. Female gametocyte variable in form, usually halter-shaped or oval; cytoplasm finely granular, staining deep blue with Leishman's



Fig. 110.—Hæmoproteus kopki (de Mello). A, B, female gametocytes; C, D, male gametocytes. (After de Mello.)

or **Oproteus glau**emsa stain, with small irregular alveoli unit *poproteus glauenid* all over the body, sometimes with larger vacuate gamet-cyte arger clear zones; nucleus a compact chy man's stath; tained faintly rose; pigment granules very fir situated s'on the d irregularly all over the body. Male ga ere we make ga<sup>35</sup> ovoid, not halter-shaped; cytoplasm not staining finite *i* staining light straw-yellow or light grey, with very dis ntravacuoles, rarely without them; nucleus granular, compact, sausage-shaped or oval, only faintly staining, and always larger than in the female gametocyte; and situated in one, two or three well circumscribed vacuoles. Infected red cell hypertrophied, dehæmoglobinized, with its nucleus displaced. Schizogony in the monocytes or endothelial cells of the lung.

Remarks.—De Mello (1934 b) has described the schizogony occurring in the pulmonary epithelium of the infected lizard as very similar to that of *Hæmoproteus* in birds. This, along with similarity in other characters, definitely establishes the identity of *Hæmocystidium* with *Hæmoproteus*. He has further shown that *H. kopki* is specifically distinct from *H. simondi*, and not identical as was believed by Wenyon.

Habitat.—Blood and lungs of Hemidactylus brooki Gray: PORTUGUESE INDIA, Nova Goa.

# 158. Hæmoproteus machlolophi de Mello. (Fig. 111.)

†Hæmoproteus danilewskyi, Plimmer, 1912, p. 417.

†Hæmoproteus machlolophi, de Mello, 1935a, pp. 353-4, pl. xliii, fig. 3; 1937 a, p. 100.

Halteridia forms not very conspicuous; nucleus subcentral, stained pale rose with Leishman's stain. Female



Fig. 111.—Hæmoproteus machlolophi (de Mello). A, female gametocyte ; B, male gametocyte. (After de Mello.)

gametocytes very irregular, denticulated in outline, stained greyish-blue; pigment dark-brown, granules irregularly scattered in the cytoplasm. Male gametocytes of a regular outline, stained clear, almost white; pigment  $dr_k$ - $br_0\psi$ , granules, showing a tendency to collect at the poles.

Habitat.—Blood of the yellow-cheeked tit, Machlolaphus xanthogenys (Vigors): from INDIA, in the Zoological Garden<sup>18</sup>, London; also from PORTUGUESE INDIA, Siroda (Pondá).

#### 159. Hæmoproteus metchnikovi (Simond). (Fig. 112.)

- *†Hæmamæba metchnikovi*, Simond, 1901 a, pp. 150-2; 1901 b, pp. 338-43, pl. viii, figs. 1-20.
- Hæmocystidium metchnikovi, Castellani & Willey, 1905, pp. 84-5;
  Castellani & Chalmers, 1919, p. 516; Wenyon, 1926, pp. 899, 901, 1396; Knowles, 1928, p. 378; Reichenow, 1929, p. 978;
  Coatney, 1936, p. 89.

Gametocytes rarely exceeding half the blood-corpuscle in size, with a small number of pigment granules, and not causing



Fig. 112.—Hæmoproteus metchnikovi (Simond). A, tailed non-pigmented form; B, large non-pigmented vermicule form; C, young pigmented form; D, female gametocyte; E, female gametocyte and male gametocyte in the same corpuscle; F, fully developed female gametocyte showing nucleus; G, leucocyte from the spleen showing two female and one male gametocytes; H, leucocyte from the lung showing male gametocyte. (After Simond.)

a displacement of the nucleus or distortion of the corpuscle. In some cases a blunt prolongation of the parasite extends up the side of the nucleus, but typical halteridia forms are not seen. Male gametocytes faintly staining, containing larger nucleus and coarse irregularly distributed pigment; female gametocytes deeply staining, containing smaller nucleus and finer pigment. Sometimes two gametocytes are found in the same corpuscle.

Dimensions.—Gametocytes  $6-10\mu$  in diameter.

Remarks.—Simond (1901 a, b, e) described two distinct species from the same host, one non-pigmented and hæmogregarine-like, the other pigmented hæmamæba-like. Hæmogregarina hankini is very rare, but shows the tailed and vermieule forms similar to the Hæmogregarines of other tortoises. Hæmoproteus metchinikovi shows sexual differentiation, the faintly staining forms with coarse grains of pigment being regarded as male and the deeply staining with finer grains of pigment as female gametocytes.

Habitat.—Blood of the tortoise, Chitra indica (Gray) (=Trionyx indicus) of the Ganges and the Jumna: UNITED PROVINCES, Agra.

#### 160. Hæmoproteus orioli de Mello.

†Hæmoproteus orioli, de Mello, 1935 b, p. 469; 1937 a, p. 100.

Female gametocyte stained light blue with Leishman's stain; nucleus eentral or subcentral, constituting a more or less compact chromatic dot. Male gametocyte whitish; nucleus central or subcentral, composed of an irregular spireme. Pigment light brown, granules of different sizes irregularly distributed, sometimes situated near the poles in both sexes.

*Remarks.*—De Mello thinks that possibly the parasite may be identical with the unnamed *Hæmoproteus* of *Oriolus* sagittarius recorded by Cleland and Johnston in Australia in 1912, and of *O. galbula* recorded by Cardamatis in Greece in 1919.

Habitat.—Blood of Oriolus oriolus kundoo Sykes: PORTU-GUESE INDIA, Nova Goa.

161. Hæmoproteus raymundi de Mello & Raimundo. (Fig. 113.)

*†Hæmoproteus raymundi*, de Mello & Raimundo, 1934 a, pp. 97-9, pls. xii, xiii, text-fig. 1; 1934 b, pp. 1437-40, pl. i; 1937 a, pp. 98, 100.

Free trophozoites, uni- or multinucleated, are found in the lung, giving rise to schizonts, rosettes, and merozoites. The



Fig. 113.—Hæmoproteus raymundi de Mello & Raimundo. A, female gametocyte; B, male gametocyte; C, schizogony in the lung. (After de Mello and Raimundo.)

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schizogony cycle is particularly simple. The merozoites remain free among the blood-corpuscles or the cells of the host-tissue. Sections of lungs and spleen also show schizonts within the red corpuscles and free merozoites among them. In the blood the red corpuscles show halteridia or reniform gametocytes. Female gametocyte stained deep blue with Leishman's stain; cytoplasm finely vacuolated; nucleus elliptical; pigment granules deep brown and slightly larger than in the male. Male gametocyte stained light blue; nucleus granular; pigment granules finer. Gametocytes may become extracellular and free in the plasma, and assume an oval or rounded form.

Dimensions.—Gametocytes  $6-8\mu$  by  $2\cdot 5-3\mu$ ; infected corpuscles  $8-9\mu$  by  $4-4\cdot 5\mu$ .

*Remarks.*—When the gametocytes are full grown the parasitized eorpuscles become hypertrophied, decolorized, and sometimes reduced to a thin border round the parasite. The nucleus of the corpuscle is dislocated to the periphery, and finally disappears.

Habitat.—Peripheral blood and sections of the lung and the spleen of Leptocoma zeylonica (Linn.): PORTUGUESE INDIA, Nova Goa.

162. Hæmoproteus rlleyi Malkani. (Fig. 114).

†Hæmoproteus rileyi, Malkani, 1936, pp. 155-7, pl. iv.

Gametocytes show a variety of form, such as rings, pearshaped or elongate forms, spindles, boomerangs, and typical



Fig. 114.-Hæmoproteus rileyi Malakani. (After Malakani.)

halteridia encircling the nucleus of the infected red bloodcorpuscles. Male gametocytes stained a pale blue with Leishman's stain; nucleus central, large, consisting of a number of chromatin granules stained intensely red enclosed by a membrane; brownish-black pigment granules collected together. Female gametocytes stained more deeply and the nucleus not granular but compact. The number of gametocytes in each corpuscle varied from one to four. In some there was a mature gametocyte on each side of the nucleus, both of the same sex or opposite sexes. No schizonts could be seen in the blood. *Remarks.*—The parasite is believed to be responsible for a serious illness which ended fatally.

Habitat.—Red blood-eorpuscles of the peacoek, Pavo cristatus Linn. : BIHAR, Patna.

# 163. Hæmoproteus simondi (Castellani & Willey). (Fig. 115).

- *†Hæmocystidium simondi*, Castellani & Willey, 1904, pp. 84–5, figs. 10–16; Robertson, 1908, pp. 181–2; Dobell, 1910 a, pp. 68, 69; 1910 b, pp. 123–32, pl. vii; Castellani & Chalmers, 1919, p. 516, fig. 173.
  - Hæmoproteus simondi, Wenyon, 1926, pp. 899, 902, 1388; Knowles, 1928, p. 378; Reichenow, 1929, pp. 978-9, fig. 949.

+Hæmoproteus simondi, de Mello, 1934 a, pp. 6-10, 14-15, pl. ii. Hæmoproteus simondi, Coatney, 1936, p. 89.

Trophozoite small, rather irregular or amœboid, with a zone of pigment granules across the centre, at first only slightly displacing the nucleus of the corpuscle; by its further growth the nucleus of the corpuscle is more displaced. Sometimes



Fig. 115.—Hæmotropeus simondi (Castellani & Willey). A. female gametocyte; B. male gametocyte: C. double infection with a male and a female gametocyte. (After Dobell & Müblens.)

the parasite is round, lenticular or oval, the oval form nearly filling the corpuscle and moulding itself on the nucleus of the blood-cell. Spherical or discoidal forms are gametocytes. In the male type, body faintly granular; stains a delicate pale blue with Leishman's stain, and possesses numerous small pigment granules seattered round the periphery. In the female type, body stains dark blue; pigment granules, though numerous, are slightly larger, and a varying number of vacuoles always present.

Dimensions.—Gametoeytes about  $18\,\mu$  in length by  $9\,\mu$  in breadth.

*Remarks.*—Dobell (1910) describes schizonts in the red blood-corpuscles. These are round bodies about  $8\mu$  in diameter, and are supposed to divide into two or four merozoites : but probably this apparent division was an artefact due to fusion of adjacent parasites during the drying of the

blood-films, as shown by Wasielewski and Wülker in the case of the Hæmoproteus of the kestrel. De Mello (1934) has carefully compared his observations on H. kopki with the original descriptions of H. simondi, and has come to the conclusion that the two are distinct, and not identical as was supposed by Wenyon. The chief characters in which the two differ are as follows :- In H. simondi the male gametocyte is without vacuoles and with pigment granules finer than in the female and scattered over the body, whilst in H. kopki it is without distinct vacuoles; the pigment granules are generally larger than in the female and are collected in vacuoles. As regards the female gametocyte, this has numerous vacuoles and the pigment granules are larger than in the male in the former species; but is without definite vacuoles and the pigment granules are finer than in the male in the latter species.

Habitat.—Blood of Hemidactylus leschenaulti Dum. & Bibr.: CEYLON, Trincomalee ; PORTUGUESE INDIA, Mamadu, near Vavuniya.

#### 164. Hæmoproteus sturni de Mello.

†Hæmoproteus sturni, de Mello, 1935 b, pp. 473-4; 1937 a, p. 100.

Female gametocyte vacuolated, stained light blue with Leishman's stain; nucleus ovoid, central or subcentral; pigment absent or irregularly scattered. Male gametocyte halter-like or oval, almost unstained; nucleus eonspicuous, eompact, central or subcentral; pigment at the poles.

*Remarks.*—De Mello thinks that the parasite may perhaps be the same as the unnamed *Hæmoproteus* of *Sturnus vulgaris* Linn. recorded by Celli and San Felice (1891) in Italy, by Labbé (1894) in France, by Wasielewski (1896) in Germany, and by Coles (1914) in England.

Habitat.—Blood of Sturnia malabarica (Gmelin): PORTU-GUESE INDIA, Pondá.

#### 165. Hæmoproteus upupæ de Mello.

†Hæmoproteus upupæ, de Mello, 1935 b, p. 472 ; 1937 a, p. 100.

Female gametocyte stained bluish, but not uniformly, with Leishman's stain; nucleus compact, spherical, ovoid or ribbon-like, subcentral, stained deep rose; pigment in granules or clusters, irregularly distributed. Male gametocyte colourless, with the borders having a very slight bluish tinge; nucleus large, granular, pseudospirematic, the threads being irregularly interwoven or disposed in parallel lines obliquely to the long axis of the parasite, stained pale rose or violet-rose; pigment granules small, often dust-like, and never fused together, scattered over the body or at the poles, fewer than in the female gametocyte. Remarks.—The parasite may possibly be the same as that recorded by Danilewsky (1889) in Southern Russia from Upupa epops.

Habitat.—Blood of Upupa epops orientalis Stuart Baker: PORTUGUESE INDIA, Daman.

# 166. Hæmoproteus wenyoni de Mello, de Sá, de Sousa, Dias, & Noronha. (Fig. 116.)

*†Hæmoproteus wenyoni*, de Mello, de Sá, Sousa, Dias, & Noronha, 1917, pp. 11-12, pl. i, figs. 18-44; 1937 a, p. 100.

Female gametocyte ovoid or halter-shaped, not displacing the nucleus of the infected red blood-corpusele, stained deep blue with Leishman's stain; nucleus small, spherical, compact, and almost always situated near one pole of the gametocyte at its convex border; pigment granules brown, scattered all over the body, rarely confined to one pole. Male gametocyte stained blue; nucleus usually central, polymorphic,



Fig. 116.- Hæmoproteus wenyoni de Mello, de Sá, de Sousa, Dias, & Noronha. A, female gametocyte : B, male gametocyte ; C, extra-corpuscular schizont. (After de Mello and others.)

being a compact chromatic granule or comma-shaped, linear or doubled up, or pyriform, etc.; pigment yellowish-brown, darker than in the female gametocyte, and nearly always situated at both the poles.

Schizogony observed in smears from the liver and the lung, but never in the peripheral blood. Schizonts small or of medium size. "Acton body" rounded, showing nuclear multiplication. Merozoites not seen.

*Remarks.*—The form of the gametocytes, and the occurrence of schizogony in smears from the liver and the lung, make it reasonably certain that the parasite is a *Hæmoproteus*. The so-called "Acton body" is probably a detached endothelial cell of the blood-vessel.

Habitat.—Blood and smears from the liver and the lung of the tailor-bird, Orthotomus sutorius (Pennant): PORTUGUESE INDIA, Nova Goa.

#### 167. Hæmoproteus sp.

*†Hæmoproteus* (?) sp., Scott, 1926, p. 237, Wenyon, 1926, p. 1365. *Hæmoproteus* (?) sp., Coatney, 1936, p. 90; de Mello, 1937 a, p. 99.

Habitat.—Blood of the Sarus crane, Antigone antigone Linn., from India, in the Zoological Gardens, London.

# 168. Hæmoproteus sp.

*Hæmoproteus* sp., Scott, 1926, p. 237; Wenyon, 1926, p. 1368. *Hæmoproteus* sp., Coatney, 1936, p. 91; de Mello, 1937 a, p. 99.

Habitat.—Blood of the Nicobar pigeon, Calænas nicobarica Linn., from INDIA, in the Zoological Gardens, London.

#### 169. Hæmoproteus sp.

*Hæmoproteus* sp., Donovan (first recorded in Wenyon, 1926, p. 1369).

Hæmoproteus sp., Coatney, 1936, p. 91; de Mollo, 1937 a, p. 99.

Habitat.—Blood of the cuckoo, Centropus sinensis (Stephen): INDIA (locality not cited).

# 170. Hæmoproteus sp.

*†Hamoproteus* sp., Scott, 1926, p. 237; Wenyon, 1926, p. 1369. *Hamoproteus* sp., Coatney, 1936, p. 91; de Mello, 1937 a, p. 99.

Habitat.—Blood of the golden-headed chloropsis, Chloropsis aurifrons (Temm.), from INDIA, in the Zoological Gardens, London.

#### 171. Hæmoproteus sp.

†Hæmoproteus sp., Castellani & Willey, 1905, p. 385, pl. xxiv, fig. 4. Hæmoproteus sp., Wenyon, 1926, p. 1371.

Rare parasites of an elongate shape, with two or three vacuoles, free in the plasma. Sometimes the free parasite appears to be surrounded by a clear halo. Nucleus oval, central.

*Remarks.*—The parasite appeared to be non-motile, but showed a slight change of shape.

Habitat.—Blood of the common babbler, Turdoides striatus striatus (Swainson): CEYLON.

#### 172. Hæmoproteus sp.

*Hæmoproteus* sp., Plimmer, 1916, p. 85. *Hæmoproteus* sp., Wenyon, 1926, p. 1371; Coatney, 1936, p. 93.

Habitat.—Blood of yellow-fronted barbet, Cyanops flavifrons (Cuvier), from CEYLON, in the Zoological Gardens, London.

#### 173. Hæmoproteus sp.

*Hæmoproteus* sp., Plimmer, 1913, p. 148. *Hæmoproteus* sp., Wenyon, 1926, p. 1371; Coatney, 1936, p. 93.

Habitat.—Blood of the wandering tree-pie, Dendrocitta rufa vagabunda Latham, from INDIA, in the Zoological Gardens, London.

#### 174. Hæmoproteus sp.

*Hæmoproteus* sp., Scott, 1926, p. 237; Wenyon, 1926, p. 1375. *Hæmoproteus* sp., Coatney, 1936, p. 95; de Mello, 1937 a, p. 99.

Habitat.—Blood of Pallas's fishing eagle, Haliaëtus leucoryphus Pallas, from INDIA, in the Zoological Gardens, London.

#### 175. Hæmoproteus sp.

*Hæmoproteus* sp., Donovan (first recorded in Wenyon, 1926, p. 1388).

Habitat.—Blood of Hemidactylus sp.: INDIA (locality not cited).

#### 176. Hæmoproteus (?) sp.

†Hæmoproteus (?) sp., Scott, 1926, p. 237; Wenyon, 1926, p. 1377.

Habitat.—Blood of the Ceylon loriquet, Coryllis beryllinus (Forster), from CEYLON, in the Zoological Gardens, London.

#### 177. Hæmoproteus sp.

†Hæmoproteus sp., Plimmer, 1913, p. 148; Knowles, 1928, p. 372.

Habitat.—Blood of the red-breasted parrakeet, Psittacula alexandri fasciatus (Müller), from INDIA, in the Zoological Gardens, London; also from parrots in the Alipore Zoological Gardens: BENGAL, Calcutta.

#### 178. Hæmoproteus sp.

†Hæmoproteus sp., de Mello, 1935, p. 470; 1937 a, p. 100.

Female gametocyte stained light prussian-blue with Leishman's stain; nucleus pale rose. Male gametocyte stained light bluish-grey or colourless; nucleus a little larger than in the female; pigment coffee-coloured, in granules, seldom in rods, scattered singly or in clusters. Shows the peculiarity of almost completely hæmolysing the infected cell.

*Remarks.*—Hæmoproteids have been previously recorded from several species of *Strix* from different parts of the world, and in view of the possibility that it might be specifically
identical with one or the other of those parasites, de Mello has not given a name to the form observed by him.

Habitat — Blood of Strix ocellata (Lesson): PORTUGUESE INDIA, Praganã.

## 179. Hæmoproteus sp.

†Hæmoproteus sp., Knowles, 1928, p. 372.

Habitat.—Blood of water-fowls, buzzards, Java sparrows, munias, hawks, parrakeets, and canaries in the Alipore Zoological Gardens: BENGAL, Calcutta.

#### 180. Hæmoproteus (?) ægithinæ de Mello.

†Hæmoproteus æyithinæ, de Mello, 1935 b, p. 471; 1937 a, p. 100.

Female gametocyte stained light blue with Leishman's stain; nucleus ovoid, central or subcentral; pigment in minute granules, scattered over the body. Male gametocyte colourless; nucleus oval, subcentral or central; pigment in fine granules, dust-like, collected mostly at the poles. Red cell hypertrophied, with its nucleus displaced.

Habitat.—Blood of Ægithina tiphia (Linn.): PORTUGUESE INDIA, Nagoa (Salcete).

#### 181. Hæmoproteus (?) anthi de Mello.

†Hæmoproteus anthi, de Mello, 1935 b, p. 474 ; 1937 a, p. 100.

Female gametocyte stained light blue, but not uniformly, with Leishman's stain; nucleus compact, central or subcentral, stained deep rose. Male gametocyte not stained; nucleus oval or triangular, central. Pigment in both kinds of gametocytes brown, coffee-coloured, irregularly scattered, seldom located at the poles. Infected red cell hypertrophied, with its nucleus displaced.

Remarks.—De Mello thinks that the parasite is perhaps similar to the unnamed Hæmoproteus recorded from Anthus trivialis by Galli Valerio (1902) in Europe, from A. japonicus by Ogawa (1911) in Japan, and from A. pratensis by Niesehulz (1921, 1922) in Heligoland.

Habitat.—Blood of Anthus richardi rufulus Vieillot : PORTU-GUESE INDIA, Nagoa (Salcete).

#### 182. Hæmoproteus (?) bramæ de Mello.

*Hæmoproteus* sp., Donovan (first recorded in Wenyon, 1926, p. 1366).

†Hæmoproteus bramæ, de Mello, 1935 b, pp. 474-5; 1937 a, p. 100.

Female gametocyte halter-shaped, slender, more or less irregular, somewhat constricted in the middle, rarely oval, cystoplasm homogeneous, stained blue with Leishman's stain, more deeply at the poles; nucleus oval, central or subcentral, stained pale rose; pigment granules isolated or in clusters, irregularly scattered. Male gametocyte halter-shaped, commashaped, or oval; nucleus without definite outline, often scarcely visible; pigment granules fewer than in the female gametocyte, generally at the poles, but sometimes over the whole body. Infected red cell unaltered or slightly hypertrophied, with its nucleus displaced.

Habitat.—Blood of Athene brama Temm. : INDIA (locality not cited); PORTUGUESE INDIA, Nagoa (Salcete).

#### 183. Hæmoproteus (?) centropi de Mello.

†Hæmoproteus centropi, de Mello, 1935 b, p. 471 ; 1937 a, p. 100.

Female gametocyte with alveolar cytoplasm, stained light blue with Leishman's stain; nucleus central, pale rose; pigment yellow-brown, in large or minute dust-like granules, situated either at the poles or irregularly scattered over the body. Male gametocyte eolourless, or with a slightly pale blue tinge in the periphery or at the poles; nucleus central, much larger than in the female gametocyte, and containing minute distinct chromatic dots or an irregular spirematic thread; pigment very irregularly distributed, but showing a tendency for a polar location. Infected red cell hypertrophied, with its nucleus displaced.

*Remarks.*—Donovan recorded an unnamed Hæmoproteid from *Centropus sinensis* from India, and unnamed Hæmoproteids have also been recorded from two other species of the same genus from other parts of the world.

Habitat.—Blood of Centropus sinensis parroti Stresemann : PORTUGUESE INDIA, Diu.

#### 184. Hæmoproteus (?) dicruri de Mello.

†Hæmoproteus dicruri, de Mello, 1935 b, p. 473 ; 1937 a, p. 100.

Female gametocyte stained bluish with Leishman's stain, but not uniformly, the poles and the convex border remaining unstained; nucleus small, subcentral, stained pale rose; pigment yellow-brown, in granules or rods, collected at the poles. Male gametocyte ovoid, not stained; nucleus rod-like, subcentral, sometimes indistinct, stained pale rose; pigment in large granules, scattered or collected in clusters at the poles. Infected red cell hypertrophied, with its nucleus displaced.

*Habitat.*—Blood of *Dicrurus macrocercus macrocercus* Vieillot : PORTUGUESE INDIA, Praganã.

#### 185. Hæmoproteus (?) elani de Mello.

†Hæmoproteus elani, de Mello, 1935 b, p. 472 ; 1937 a, p. 100.

Female gametocyte stained uniformly blue with Leishman's stain; nucleus compact, small, ovoid, rarely triangular, subcentral, stained reddish. Male gametocyte not stained; nucleus large, granular, without a definite outline. Pigment of sepia colour, disposed in both male and female gametocytes in a characteristic manner, being in the form of large mulberrylike masses at the poles. Infected red cell hypertrophied, with its nucleus displaced.

Habitat.—Blood of Elanus cæruleus vociferus Latham : Portuguese India, Daman.

#### 186. Hæmoproteus (?) gallinulæ de Mello.

*Hæmoproteus gallinulæ*, de Mello, 1935 b, p. 469 ; 1937 a, p. 100.

Female gametocyte with vacuolated cytoplasm, stained deep blue with Leishman's stain; nucleus stained pale rose; pigment brownish-black, in granules of different sizes scattered all over the body, rarely collected at the poles, sometimes grouped in clusters. Male gametocyte stained light blue, rather colourless, roundish or oval, almost never halter-like; pigment in irregular clusters collected at the poles. Infected red cells hypertrophied, with the nucleus displaced. No schizogonic forms observed in lung-smears.

Habitat.—Blood of Gallinula chloropus (Linn.): PORTU-GUESE INDIA, lake of Carombolim.

#### 187. Hæmoproteus (?) halcyonis de Mello.

*Hæmoproteus halcyonis*, do Mello, 1935 b, p. 474 ; 1937 a, p. 100.

Female gametocyte halter-like, oval, or cordiform, vacuolated or not, stained deep blue with Leishman's stain; nucleus oval, central or subcentral; pigment at the poles, sometimes in the centre or near the periphery. Male gametocyte not stained; nucleus indistinct, oval or comma-shaped, central or subcentral; pigment scattered over the body, often denser at the periphery. Infected red cell slightly hypertrophied, sometimes not at all, with its nucleus displaced.

Habitat.—Blood of Halcyon smyrnensis (Linn.): PORTU-GUESE INDIA, Canacona.

# 188. Hæmoproteus (?) halcyonis var. fuscæ de Mello & da Fonseca. (Fig. 117.)

†Hæmoproteus halcyonis fuscæ, de Mello & da Fonseca, 1937, pp. 215-16, text-figs.

Female gametocyte oval, fusiform or like a slender halter when young, definitely halter-like when full grown; often

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the halteridia are somewhat irregular, and when fully grown both poles of the gametocyte fuse together, surrounding the nucleus of the red cell; stained definitely blue with Romanowsky's stain; nucleus oval, generally central; pigment granules blackish-brown or coffee-brown, with irregular distribution. Male gametocyte oval when young, halter-like when fullgrown, sometimes completely surrounding the nucleus of the red cell, not stained or slightly blue with Romanowsky's



Fig. 117.—Hæmoproteus (?) halcyonis var. fuscæ de Mello & da Fonseca. A, B, female gametocytes; C, D, male gametocytes. (After de Mello & da Fonseca.)

stain; nucleus large, central or subcentral, stained rose by Leishman's stain, reddish by May-Grunwald Giemsa stain; pigment granules variable in size and location, showing a tendency to be callected at the poles. Infected red cell not altered when the parasite is young; when the latter is full grown the nucleus of the red cell is displaced to the periphery.

Habitat.—Blood of Halycyon smyrnensis fusca (Bodd.): PORTUGUESE INDIA, Santo Estevam.

#### 189. Hæmoproteus (?) lanli de Mello. (Fig. 118.)

Hæmoproteus lanii, de Mello, 1937 a, pp. 102-3, pl. i, fig. 2.

Female gametocyte halter-shaped, staining blue with



Fig. 118.—*Hæmoproteus* (?) *lanii* de Mello. *A*, female gametocyte; *B*, male gametocyte. (After de Mello.)

Leishman's stain ; nucleus small, oval or triangular, generally subcentral, not staining or very lightly with Leishman's stain, staining well with May-Grunwald Giemsa stain ; pigment granules large, scattered irregularly over the body. Male gametocyte irregularly halter-shaped, almost quadrangular, not stained or very light blue ; nucleus large, central, without definite outline, with chromatinic masses irregularly scattered or disposed in a reticulum ; pigment granules very minute, located at the poles. Infected red cell hypertrophied, with its nucleus displaced.

*Remarks.*—Unnamed species of *Hæmoproteus* have been previously recorded from many different species of the genus *Lanius* in different parts of the world.

Habitat.—Blood of Lanius schach erythronotus Vigors: POR-TUQUESE INDIA, Pondá.

# 190. Hæmoproteus (?) otocompsæ de Mello.

*Hæmoproteus otocompsæ*, de Mello, 1935 b, p. 473 ; 1937 a, p. 100.

Female gametocyte with cytoplasm stained pale blue with Leishman's stain and rose with May-Grunwald Giemsa stain, seldom vacuolated; nucleus spherical, subcentral, seldom elongated, situated on the convex border; pigment scattered over the body. Male gametocyte not stained with Leishman's stain, pale rose with May-Grunwald Giemsa stain; nucleus very large, granular, without definite outline, subcentral; pigment at the poles. Infected red cell hypertrophied, with its nucleus displaced.

*Remarks.*—According to de Mello the parasite is perhaps identical with the unnamed *Hæmoproteus* of *Picnonotus barbatus* recorded by A. and M. Léger in Senegal.

Habitat.—Blood of Elathea jocosa (Linn.) [=Otocompsa emeria (Linn.)]: PORTUGUESE INDIA, Malim (Bardéz).

#### 191. Hæmoproteus (?) pastoris de Mello.

*†Hæmoproteus pastoris*, de Mello, 1935 b, p. 470. *Hæmoproteus* sp., Coatney, 1936, p. 98. *Hæmoproteus pastoris*, de Mello, 1937 a, p. 100.

Female gametocyte stained blue with Leishman's stain, with a small ovoid nucleus, central or subcentral, stained pale rose; halter-shaped or ovoid, and in the latter ease nucleus of the host-cell completely displaced; pigment seattered all over the body. Male gametocyte colourless or very slightly blue, halter-shaped, ovoid or irregularly deformed; nucleus indistinct, composed of chromatic masses disposed like the spokes of a wheel, or in irregularly weaved threads; pigment polar in halter-shaped forms, seattered over the body in ovoid or deformed gametocytes. Infected red cell hypertophied, with its nucleus displaced.

Habitat.—Blood of Pastor roseus (Linn.): PORTUGUESE INDIA, Praganã.

# 192. Hæmoproteus (?) plataleæ de Mello.

*†Hæmoproteus platalex*, de Mello, 1935 b, p. 471. *Hæmoproteus* sp., Coatney, 1936, p. 99. *Hæmoproteus platalex*, de Mello, 1937 a, p. 100.

Female gametocyte with alveolar cytoplasm, stained deep blue with a greenish tone with Leishman's stain; nucleus central; pigment coffee-olive tone, scattered over the body, showing a tendency to undergo solution and to diffuse through the cytoplasm. Male gametocyte not stained, or very slightly greyish-blue, the margins at the poles being often the only stained part; pigment in minute granules, often in clusters, generally at the poles. Infected red cell hypertrophied, with its nucleus displaced.

Habitat.—Blood of Platalea leucorodia major Temm. & Schleg. : PORTUGUESE INDIA, Diu.

# 193. Hæmoproteus (?) tephrodornis de Mello.

†Hæmoproteus tephrodornis, de Mello, 1935 b, p. 473; 1937 a, p. 100.

Female gametocyte halter-shaped or ovoid, stained blue with Leishman's stain, but not uniformly; nucleus oval, central or subcentral. Male gametocyte oval, stained light greyish; nucleus small, indistinet. Pigment black or sepia eoloured, in granules or rods disposed in lines or clusters along the eoncave border. Infected red cell hypertrophied, with its nucleus displaced.

Habitat.—Blood of Tephrodornis pondicerianus pondicerianus (Gmelin) : PORTUGUESE INDIA, Praganã.

# 194. Hæmoproteus (?) thereicerycis de Mello.

†Hæmoproteus thereicerycis, de Mello, 1935 b, p. 470; 1937 a, p. 100.

Female gametocyte halter-shaped, ovoid or spherical, cytoplasm finely vacuolated, stained blue with Leishman's stain; nucleus hardly distinguishable with Leishman, well stained with May-Grunwald Giemsa stain, spherical, central or subcentral, lodged in a definite vacuole; pigment granules of different sizes scattered over the body. Male gametocyte always spherical, colourless with Leishman's stain, rose with May-Grunwald Giemsa stain; nucleus very small, commashaped; pigment granules at the poles a little larger than those scattered over the body. Infected red cell hypertrophied, with its nucleus displaced. Shows a marked tendency to hæmolyse the infected cell and become free, taking in this frec condition aberrant forms like deformed halteridia or spheres.

Habitat.—Blood of Thereiceryx zeylanicus inornata Walden ; PORTUGUESE INDIA, Corlim (Ilhas).

# 195. Hæmoproteus (?) thereicerycis var. zeylonica de Mello.

*†Hæmoproteus thereicerycis var. zeylonica*, de Mello, 1935, p. 470 ; 1937 a, pp. 100, 101-2.

Female gametocyte with vacuolated cytoplasm, stained dark blue with Leishman's stain; nucleus round, seldom elongated, central or subcentral; pigment granules scattered, minute, often in clusters. Male gametocyte colourless; nucleus large and without definite outline, always central, and containing chromatic masses with irregular disposition; pigment granules very minute, situated at the poles. Infected red eell hypertrophied, with its nucleus displaced.

*Remarks.*—This variety differs from the preceding species as regards the structure of the male gametocyte, and does not show the same tendency to hæmolyse the host-cell.

Habitat.—Blood of Thereiceryx zeylanicus zeylanicus (Gmelin): PORTUGUESE INDIA, Malim (Bardéz); blood of Thereiceryx viridis Bodd.: PORTUGUESE INDIA, Pondá.

# Genus LEUCOCYTOZOON Danilewsky, 1889.

Leucocytozoon, Danilewsky, 1889, p. 23; Berestneff, 1903, pp. 376– 86; Wenyon, 1910, pp. 63–72; Franca, 1912, pp. 173–6; Mathis & Lóger, 1912, pp. 77–82; Minchin, 1912, pp. 357, 369–71, 390, 392; Moklovan, 1914, pp. 249–62; Wenyon, 1926, pp. 903–8; Kudo, 1931, p. 288; Calkins, 1933, p. 566; de Mello, 1935 a, pp. 355–7; 1935 b, pp. 68–73; Reichenow, 1935, p. 375.

Schizogony takes place in the internal organs of the host, probably in the endothelial cells of the blood-vessels (as in *Hæmoproteus*). Certain cells in the peripheral blood, which were originally thought to be leucoeytes (hence the name of the genus) but are now believed to be young erythrocytes, in which the colouring matter has not yet developed, contain the gametocytes in various stages of growth. No pigment is produced. Micro- and macrogametocytes are differentiated, the nucleus of the microgametocyte is large and contains numerous diffuse chromatin granules, while that of the macrogametocyte is compact and contains a definite karyosome. The host-cell is profoundly modified into an elongate fusiform body, much larger than the normal red blood-corpuscle, and contains the elongate and hypertrophied nucleus of the cell and the elongate gametocyte. The gametocytes leave the host-cell, the microgametocyte producing the microgametes by a process of flagellation and the macrogametocyte rounding off to form a macrogamete. Finally, a large unpigmented oökinete is formed. Blood parasites of various birds.

**Remarks.**—Wenyon (1926) gives a useful summary of the previous literature on this genus and places it in the same family with *Hæmoproteus*, from which it differs chiefly in the absence of pigment in the gametocytes and the peculiar modification of the host-cell. Reichenow (1931), stressing the affinities with the COCCIDIA, places it in a family by itself under EIMERIDEA. I have followed Wenyon, Kudo, and Calkins in placing it here. De Mello (1937 a) has surveyed the previous literature dealing with the genus, and come to the conclusion that species with a rounded form should be placed in an independent genus from *Leucocytozoon*, which should be restricted to the fusiform species.

# 196. Leucocytozoon ardeolæ de Mello.

†Leucocytozoon ardeolæ, de Melle, 1937 a, p. 106.

Form round. No sexual differentiation made out. Cytoplasm with very minute alveoli, staining deep blue with Leishman's stain; nucleus circular, staining pale rose.

Habitat.-Blood of Ardeola grayi Sykes: PORTUGUESE INDIA.

# 197. Leucocytozoon chloropsidis de Mello. (Fig. 119.)

†Leucocytozoon chloropsidis, de Mello, 1935 a, pp. 356-7, pl. xliii, fig. 2; 1937 a, p. 105.

Form round or oval. Cytoplasm alveolar. Sexual dimorphism of the gametocytes not shown by the cytoplasm, which stains deep blue in both the male and female, but by the



Fig. 119.—Leucocytozoon chloropsidis de Mello. A, female gametocyte; B, male gametocyte. (After de Mello.)

appearance of the nucleus. In the male gametocytes the nucleus is long, thread-like, irregular, and stains violet by Romanowsky stains. In the female gametocytes the nucleus is roundish or oval, weakly stained pale rose or not stained at all. Infection very scanty.

Habitat.—Blood of the golden-headed chloropsis, Chloropsis aurifrons davidsoni Stuart Baker : PORTUGUESE INDIA, Mardol.

# 198. Leucocytozoon coraciæ de Mello & Afonso. (Fig. 120.)

†Leucocytozoon coraciæ benghalensis, de Mollo & Afonso, 1935 b, pp. 71-2, pl. ii; de Mello, 1937 a, p. 105.

Form of the adult gametocyte ovoid, included in a fusiform cell. Cytoplasm finely alveolar stains blue with Romanowsky stains, but lighter than in L. melloi. Ratio of length to breadth is 5 to 2 in intracellular forms and 7 to 4 in free forms. Nucleus



Fig. 120.—Leucocytozoon coraciæ de Mello & Afonso.

A, female gametocyte; B, male gametocyte. (After de Mello and Afonso.)

ovoid or bean-shaped, central or subcentral. Sexual dimorphism is recognizable in the structure of the nucleus as stained by iron hæmatoxylin. The nucleus of the female gametocyte is small, with a karyosome and a centriole or only with centriole. The nucleus of the male gametocyte is much larger, often oval, elongated, or even bean-shaped, filled with granules, but without a centriole.

Remarks.—No young forms or schizogony stages were recognized. Specimens of this species were far more abundant (in the ratio of 7 to 1) than those of L. melloi described from the same host.

The poles of the fusiform host-cell are stained greyish-blue by Romanowsky stains, and stained lightly by Heidenhain's iron hæmatoxylin. The nucleus of the host-cell is attached to one side of the parasite, giving it a bean-shaped form. The parasite is really ovoid, as can be seen in free forms or where the nucelus of the host-cell is still central. In many cases one or both poles of the host-cell are lost, and it appears to be represented only by its nucleus.

Habitat.—Blood or smears from the lungs of Coracias benghalensis benghalensis (Linn.): PORTUGUESE INDIA, Corlim.

# 199. Leucocytozoon enriquesi de Mello.

†Leucocytozoon enriquesi, de Mello, 1937 a, p. 106.

Form round. Differs from the preceding species in the nucleus being roundish, ovoid or sausage-shaped and always well stained in the female gametocyte; and in the nucleus being very large, circular, and occupying the greater part of the body, and stained pale rose, dust-like and with a more deeply staining body, in the male gametocyte.

Habitat.—Blood of Chloropsis jerdoni Blyth: PORTUGUESE INDIA.

## 200. Leucocytozoon (?) melloi, sp. nov. (Fig. 121.)

†Leucocytozoon (?) sp., "type B," do Mello & Afonso, 1935 b, pp. 72-3, pl. ii ; de Mello, 1937 a, p. 105.

Form of the adult gametocyte spherical, attached to the nuclear substance of the host-cell, the rest of the cell not to be seen. Cytoplasm compact, stained deep blue by Romanowsky stains, and nuch darker than *L. coraciæ* by iron hæmatoxylin;



Fig. 121.—Leucocytozoon (?) melloi, sp. nov.

A, B, forms with nucleus of the host-cell attached ; C, free form. (After de Mello and Afonso.)

containing small or large vacuoles, which do not take the stain. Only one type of gametocyte recognizable, with a nucleus containing a karyosome.

*Remarks.*—Forms are of different sizes, the free ones being absolutely spherical, and not ovoid as in *L. coraciæ* from the same host. The relationship of the nucleus of the host-cell SPOR. R to the parasite is entirely similar to that of the host-cell and the previous species.

Marcel Léger (1913) and Laverallan and Marullaz (1914) have previously described Leucocytozoons of a rounded form, and interpreted them as different stages of the typical form. De Mello and Afonso (1935 b) are, however, of the opinion that they are definitely dealing with two independent species, of which the first one may be classified as a *Leucocytozoon*, while the second has enough characters to constitute an independent genus, to which the name of Marcel Léger should be attached. As they have not given a definite name to the species, but have simply described it as "type B," I have for the present kept it in the genus *Leucocytozoon* and have given the name *melloi* to the species.

Habitat.—Blood and smears from the lungs of Coracias benghalensis benghalensis (Linn.): PORTUGUESE INDIA, Corlim.

#### 201. Leucocytozoon molpastis de Mello.

+Leucocytozoon molpastis, de Mello, 1937 a, pp. 106-7, pl., fig. 1.

Form round. Cytoplasm with very few minute vacuoles, not showing sexual differentiation. Nucleus circular, with a well-marked membrane and a conspicuous central granule in the female gametocyte; oval, with chromatin disposed in variously arranged threads in the male gametocyte.

Habitat.--Blood of Molpastes cafer cafer (Linn.): PORTU-GUESE INDIA, Pondá.

#### 202. Leucocytozoon sp.

*Leucocytozoon* sp., Donovan (first recorded in Wenyon, 1926, p. 1366).

Leucocytozoon sp., de Mello, 1937 a, p. 105.

Habitat.—Blood of the Indian little owl, Athene brama (Temm.): INDIA (locality not eited).

#### 203. Leucocytozoon sp.

†Leucocytozoon sp., Donovan (first recorded in Wenyon, 1926, p. 1373).

Leucocytozoon sp., de Mello, 1937 a, p. 105.

Habitat.—Blood of the falcon, Falco sp.: INDIA (locality not cited).

#### 204. Leucocytozoon sp.

†Leucocytozoon sp., Scott, 1926, p. 237; Wenyon, 1926, p. 1375. Leucocytozoon sp., de Mello, 1937 a, p. 105.

Habitat.—Blood of the rufous-chinned laughing-thrush, Ianthocincla rufogularis Gould, from INDIA, in the Zoological Gardens, London.

#### 205. Leucocytozoon sp.

†Leucocytozoon sp., Knowles, 1925.

Leucocytozoon sp., Wenyon, 1926, p. 1377; de Mello, 1937 a, p. 105.

Habitat.—Blood of the babbler, Leiothrix lutea (Scop.): BENGAL, Calcutta.

#### 206. Leucocytozoon sp.

Leucocytozoon sp., Plinmer, 1914, p. 190. Leucocytozoon sp., Wenyon, 1926, p. 1379; de Mello, 1937 α, p. 105.

Habitat.—Blood of the chat, Oreicola ferrea (Gray), from INDIA, in the Zoological Gardens, London.

#### 207. Leucocytozoon sp.

†Leucocytozoon sp., Plimmer, 1917, p. 32. Leucocytozoon sp., Wenyon, 1926, p. 1382; de Mello, 1937 a, p. 105.

Habitat.—Blood of the finch, Propasser rhodochrous (Vigors), from INDIA, in the Zoological Gardens, London.

#### 208. Leucocytozoon sp.

†Leucocytozoon sp., Knowles, 1928, pp. 379, 381.

*Remarks.*—The host-cells, although very much enlarged and showing the characterististic squeezing-out of the nucleus by the parasite against the cell-membrane, did not show the drawn-out tapering ends. Knowles thinks this may have been due to post-mortem ehanges, as the material was not fresh.

Habitat.—Blood from the heart and other viscera of Peking robins in the Alipore Zoological Gardens : BENGAL, Calcutta.

#### 209. Leucocytozoon (?) sp.

†Leucocytozoon (?) sp., de Mello, 1937 a, p. 105.

Form round. Female gametocyte with very small vacuoles, and staining deep blue with Leishman's stain; nucleus ovoid, granular, staining rose colour. Male gametocyte with large vacuoles, and staining light blue; nucleus in the form of an irregular thread.

*Remarks.*—According to de Mello, the form is perhaps similar to *L. annelloix* Cleland, 1912, described from *Oriolus* sagittarius in Australia.

Habitat.-Blood of Oriolus oriolus kundoo Sykes : PORTU-GUESE INDIA.

# 210. Leucocytozoon (?) sp.

†Leucocytozoon (?) sp., de Mello, 1937 a, pp. 105-6.

Form round. No sexual differentiation. Great tendency to aberrant forms in addition to round ones of regular type. Nucleus crescentic or irregular and very large in some specimens; oval, with a more deeply staining line or point in others.

Habitat.—Blood of Oriolus xanthornus xanthornus (Linn.): PORTUGUESE INDIA.

# 2. Family PLASMODIIDÆ Mesnil, 1903.

Schizogony takes place within the red blood-corpuscles of a Vertebrate host. During the growth of the trophozoite, pigment, known as hæmozoin, is formed from the hæmoglobin of the corpuscle. Gametocytes also occur in the red bloodcurpuscles and contain pigment. Further development takes place in the body of a mosquito. Microgametes are produced by exflagellation. Zygote becomes a motile oökinete, and later encysts as an oöcyst. The oöcyst grows enormously in size, and iunumerable sporozoites are produced, without the formation of sporoblasts or sporocysts.

The family, according to many authorities, contains a single genus, Plasmodium, as they consider Plasmodium Marchiafava & Celli, Laverania Grassi & Feletti, and Proteosoma Labbé as congeneric. Doflein (1916) and Reichenow (1929) consider these as distinct, and Reichenow also includes the genus Ductulosoma in the family. According to Reichenow it would be desirable to retain the generic name Proteosoma for the malarial parasites of birds, and to refer the malarial parasites of the reptiles also to it. He also considers it justifiable to place the human tropical parasite of malignant tertian malaria in a separate genus, Laverania. Both the genera Proteosoma and Laverania show some resemblance to COCCIDIA in that the gametocytes are spherical, oval, vermiform or sausage-shaped, whilst in *Plusmodium* the gametocytes are circular and disc-The genus Laverania, which includes parasites of man like. and anthropoid apes, is not easily marked off from the Proteosoma of birds. Slender gametocytes, like those characteristic of Laverania, occur also in some species of Proteosoma, and the characteristic that schizogony takes place in the internal organs in the case of Laverania has been found to occur, according to the recent researches of Hartmann, in Proteosoma also.

Of the English authors, Thomson and Woodcock (1922) justified the retention of *Laverania* as a separate genus on the ground that the gametocyte or crescent is enclosed by a capsule,

the male crescent possessing a more delicate capsule than the female crescent. Wenyon (1926) is not convinced that any such capsule exists, and is of the opinion that the lifecycles of the three species of malarial parasites are so similar in every respect that, even if a capsule did exist, it would not justify generic distinction. In my opinion the life-cycle is bound to be similar within the same family, and the difference in form of the gametocyte is a sufficiently important feature to serve as a basis for generic distinction. Stiles (1928) has announced that the International Commission of Zoological Nomenclature has placed both *Laverania* and *Plasmodium* in the official list of generic names.

History of the Discovery of Malarial Parasites and the part played by the Mosquitoes.-It will not be out of place to briefly review here the history of the discovery of malarial parasites and of the part played by the mosquitoes in their transmission. The study of the HÆMOSPORIDIA (or Hæmocytozoa or Hæmatozoa as they were then generally called) began with the discovery by Ray Lankester in 1871 of Drepanidium ranarum in the blood of the frog. Human malarial parasites are said to have been seen, but their significance was not comprehended until Laveran published his investigations (1880). Golgi (1885) demonstrated the relationship existing between the life-eycle of the parasites within the human body and the occurrence of the febrile attack. In India Vandyke Carter (1888), Evans (1888), Hehir (1893), Crombie (1894), and Ronald Ross (1895) were the earliest to study the malarial parasites in man. Ross (1895) observed the process of "flagellation" of crescentic parasites in the stomach of mosquitoes fed on the blood of a malarial patient. MacCullum (1897) found that the "flagella" of Hæmoproteus (Halteridium) and of æstivo-autumnal parasites constitute the male element, and serve to impregnate the "pigmented spheres" or female element, and further observed in the former that the impregnated spheres become converted into motile "vermicules." Ross (1897), working at Secunderabad, fed mosquitoes upon human blood containing "crescents." After examining hundreds of mosquitoes fed on malarial blood, with negative results, he obtained a few mosquitoes with spotted wings, in which he discovered peculiar pigmented cells lying within the walls of their stomachs. The pigment was similar to that within the malarial parasites in the blood upon which the mosquitoes had been fed. Ross concluded that he had found the mosquito which served as a host for the parasite. In February 1898 he again referred to his experiments with crescentic parasites and dapple-winged mosquitoes. After this, working at Calcutta, Ross observed the development of the malarial parasite of birds. Proteosoma, in a species of

Culex (subsequently determined as C. fatigans Wied.), the insects being fed on the blood of infected crows, larks, and sparrows, and found similar pigmented cells. In July of the same year Manson reported to the British Medical Association at Edinburgh further observations on behalf of Ross, which showed that the encapsulated parasites, on reaching a certain size, ruptured and emptied their contents into the cœlome, and these minute spindle-shaped bodies subsequently accumulated in the salivary gland of the insect, thus making it capable of transmitting the infection to healthy birds.

In October 1898 Grassi suspected three species of Culicidæ as being carriers of malarial infection, as they were confined in their geographical distribution to those regions where malaria was prevalent in Italy. It has since been proved that only one of these three, viz., Anopheles claviger, can serve as a host for human malarial parasites. A month later Grassi reported that Bignami had made an infection experiment with positive results. Bignami, Bastianelli, and Grassi (1898) observed the development of croscentic parasites in Anopheles claviger, and reported that the appearances correspond to those described by Ross for Proteosoma on the fourth day in Culex, and further that they had successfully infected a person with tertian fever by means of infected A. claviger. A few weeks later they followed the development of crescentic parasites in A. claviger to the formation of sporozoites, their escape into the cœlome, and their accumulation in the salivary glands of the insect. Koch (1899) reported the results of the German Malaria Commission and confirmed the development of Proteosoma in Culex as previously described by Ross. Daniels (1899) also reported to the Royal Society that he had been able to confirm Ross's observations with Proteosoma.

Grassi, Bignami, and Bastianelli (1899) observed the development of quartan parasites in A. claviger, and Bastianelli and Bignami (1899) reported further studies upon the development of tertian parasites in A. claviger, and later extended their studies of development in certain other species of Anopheles. In September 1899 Bastianelli and Bignami gave a detailed description of benign and malignant tertian parasites, and their papers were illustrated by the best coloured plates published till then, illustrating the development. Ziemann (1900) observed the development of the parasites of tropical malaria in two species of Anopheles and of tertian parasites in one species of Anopheles, and followed the development up to the appearance of sporozoites in the salivary glands of the insects. In September 1900 Manson reported a positive infection experiment with tertian-fed Anopheles imported from Rome, the insects being permitted to bite his son. Luhe (1900) showed the correspondence of the life-cycle of the

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malarial parasites with that of COCCIDIA, and introduced a terminology which is in use up till now. Nuttall (1901) gave a useful summary of the researches on malaria, recording the various works in a chronological order, and the interested reader may be referred to it for fuller details. Laveran (1907) also reviewed the published literature.

## Key to Indian Genera.

corpuscles in t	he internal organs;	
gametocytes spł	nerical, ovoid, vermi-	
form or sausage-s	shaped	2.
2 (3). Parasites of birds	or reptiles; gameto-	[p. 247.
cytes spherical, o	void, or vermiform	PROTEOSOMA Labbé,
3 (2). Parasites of man	and anthropoid apes;	
gametocytes cre	escentic or sausage-	[Feletti, p. 259.
shaped		LAVERANIA Grassi &
4 (1). Schizogony takes p corpuscles in t	place in the red blood- he peripheral blood;	
outline. Parasit- mammals	es of man and other	[fava & Celli, p. 270. Plasmodium Marchia-

#### Genus PROTEOSOMA Labbé, 1894.

Pseudovacuole + Polymitus + Pseudospirilles, Danilewsky, 1889. Hæmamæba (part), Grassi & Feletti, 1890, p. 463; 1891 a, p. 465;

1892, p. 10.

Hæmoproteus (part), Kruse, 1890, p. 359.

Hæmoproteus vars. b & c, Celli & San Felice, 1891, pp. 541-8, pls. vii, viii.

Corps sphérique + Corps à flagelles + Corps en rosette (part), Laveran, 1891.

Cytosporon+Polymitus malariæ avium, Danilewsky, 1891, p. 758.

Proteosoma, Labbé, 1894, pp. 142, 157.

Hæmoproteus, Labbé, 1899, p. 79.

Hæmoproteus (Proteosoma), Minchin, 1903, pp. 250, 257, 265, 267.

Plasmodium, Minchin, 1912, p. 358.

Proteosoma, Doflein, 1916, pp. 896-901; Franca, 1917, pp. 221-7. Plasmodium, Castellani & Chalmers, 1919, p. 513; Taliaferro, 1925, pp. 742-89; Wenyon, 1926, pp. 977-81.

Proteosoma, Calkins, 1926, p. 444.

Plasmodium, Hartmann, 1927, pp. 1-7; Knowles. 1928, pp. 412-13, p. 442; Hegner, 1929, pp. 59-82; Manwell, 1929, pp. 308-45. Proteosoma, Reichenow, 1929, pp. 983-7.

Plasmodium, Manwell, 1930, pp. 381-3; Kudo, 1931, p. 287; Ed. & Et. Sergent & Catanei, 1931, pp. 399-437; Calkins, 1933, pp. 407, 566; Giovannola, 1934, pp. 1-10.

Proteosoma, Reichenow, 1935, pp. 376-7.

Schizogony takes place in the red blood-corpuscles, usually in the internal organs, but rarely in the peripheral blood. The fully developed schizont may more or less displace the nucleus of the host-cell or not affect it at all. Gametocytes spherical, ovoid or vermiform, in some species closely resembling those of Laverania. Sporogony as in the human malarial parasities of the genus Laverania and Plasmodium. Blood-parasities of birds and reptiles.

Remarks.-Celli and San Felice (1891) reported three kinds of malarial parasites in birds, but these were not described sufficiently adequately to be recognizable. Till 1927 the organisms continued to be considered as belonging to a single species, usually called *Plasmodium* pracox. Hartmann (1927 b), however, believes that at least three species exist. For one of these, isolated from "Whitmore" strain, he retains the name P. præcox; the second, known as "Hartmann" strain, he named P. cathemerium; and the third strain, obtained by Huff in Virginia, was named P. inconstans. Evidence has been secured by Manwell (1929) that the "Whitmore" strain may consist of two distinct varieties or species. He seems to have established that the "Whitmore "strain of malaria, originally isolated by Whitmore from a New York sparrow in 1913, is a variety of P. inconstans, and is distinct from P. præcox, which was isolated by Huff in 1926 from canaries originally infected with the "Whitmore" strain. Ed. and Et. Sergent and Catanei (1929) described another species from Algerian birds as P. rouxi, and Huff (1930) yet another as P. elongatum from canary and sparrow from the United States of America. Manwell (1930) regards P. cathemerium Hartmann and P. elongatum Huff as valid species, and P. inconstans Hartmann, the "Whitmore" strain, and the "German" strain as three strains or varieties of P. præcox. Thus according to him there are three species, namely, P. præcox, P. cathemerium, and P. elongatum. These three species differ from one another in morphology, length of incubation period, size of occvsts produced in the mosquito, length of asexual cycle, etc. The morphological differences chiefly concern the gametocytes. Those of P. præcox are more or less round, displace the nucleus of the host-cell, and contain fine dust-like particles of pigment. The gametocytes of P. cathemerium are similar in shape and size, but contain much coarser granules of pigment, which tend to have a rod-like shape. P. elongatum takes its name from the elongate character of the gametocytes, which markedly resemble those of *Hæmoproteus*. Russell (1932) described another species. Besides these species, which have been employed in experimental work, a large number of other species of Proteosoma have been described as distinct, because they are found in different species of birds. It is now no longer possible to regard the malarial parasites of birds as all belonging to the single species P. præcox; but the opposite tendency towards the multiplication of species should be avoided unless definite morphological distinctions are noted. In recent years Ed. and Et. Sergent and Catanei (1931) and Giovannola (1934) have attempted a systematic revision of

species occurring in birds. The latter recognizes nine species of avian Plasmodia, and divides them into three groups according to their morphology, as follows :—

(1) Spheroidal gametocytes; nucleus of the parasitized cell displaced by gametocytes and adult schizonts (P. præcox with pointed pigment and P. cathemerium with rod-shaped pigment in the gametocytes).

(2) Elongate gametocytes, spheroidal schizonts; nucleus of the parasitized cell displaced by schizont and slightly by gametocytes (*P. elongatum*).

(3) Elongate gametocytes; nucleus of the parasitized cell never displaced. This may be divided into two subgroups:

- (a) Adult schizonts small and quadrangular (P. rouxi with four merozoites; P. tenue with 4 to 8 merozoites).
- (b) Adult schizonts circumscribe the nucleus of the parasitized cell (P. circumflexum and P. fallax).

Coatney and Roudabush (1936) give a list of malarial parasites with their hosts, without expressing any opinion about the number of valid species. De Mello (1937 a) considers the scheme proposed by Giovannola as premature, and refers to his description of P. centropi, which combines certain characters of all the three groups.

Buxton (1935) has shown that if females of *Culex fatigans* are infected with *Proteosoma*, deaths occur earlier than in controls, and thinks it due to the invasion of the wall of the mid-gut by the oökinete.

Wolfson (1936) was able to transmit bird malaria by the intravenous injection of sporozoites. The salivary glands of *Culex pipiens* infected with *P. præcox* were dissected out and the contents placed in a solution of 1 per cent. sodium citrate in 0.7 per cent. sodium chloride to which one drop of sterile bovine serum per c.c. was added. Canaries were injected intravenously with 150 mgm. of the solution, and two out of the three birds acquired an infection.

# 211. Proteosoma centropi (de Mello).

†Plasmodium centropi, de Mello, 1937 a, p. 97.

Gametocytes spheroidal or crescent-shaped; nucleus of the parasitized cell always displaced; pigment granular or rod-shaped.

*Remarks.*—Full description of this species has not yet been published. According to de Mello (1937 a) the species combines the characters of all the three groups of Giovannola's classification as given above.

Habitat.—Blood of Centropus sinensis parroti Stresemann: PORTUGUESE INDIA.

# 212. Proteosoma chloropsidis (de Mello). (Fig. 128.)

*Plasmodium præcox*, Scott. 1926, p. 237; Wenyon, 1926, p. 1369.
 *Plasmodium chloropsidis*, de Mello, 1935 a, pp. 354-5, pl. xliii, fig. 1.
 *Plasmodium præcox*, Coatney & Roudabush, 1936, p. 342; de Mello, 1937 a, p. 98.

Young trophozoites ring-shaped, resembling those of *Laverania malarıæ*. Larger trophozoites in the form of big rings with brownish pigment granules. Rosettes with (or without) pigment, situated generally on the periphery of the red cell, with varying number (6 to 11) of merozoites. Male gametocytes oval, with large, irregular nucleus. Female gametocytes roundish, generally stained deeply blue, with round or oval nucleus. Infected red corpuscles not altered.

Remarks.—De Mello believes that the form is the same as that registered as P. præcox, from the same host, by Scott and Wenyon. He, however, regards it as a distinct species, differing from true P. præcox in the irregular number of merozoites formed during schizogony, and in the compact structure of



Fig. 128.—Proteosoma chloropsidis (do Mello). A, ring-form; B, fullgrown trophozoito; C, rosottes; D, male gametocyte; E, F, female gametocytes. (After de Mello.)

the nucleus of the merozoites, which contrasts with the ringshaped nucleus of the merozoites in *P. præcox*.

Habitat.—Blood of the golden-headed chloropsis, Chloropsis aurifrons Temm., from India, in the Zoological Gardens, London; also blood of Chloropsis aurifrons davidsoni Stuart Baker: PORTUGUESE INDIA, Mardol (Pondá).

# 213. Proteosoma columbæ (Carini).

Plasmodium columbæ, Carini, 1912 pp. 396-8.
Plasmodium sp., Ed. & Et. Sergent, 1904, pp. 132-3.
†Hæmoproteus columbæ, de Mello & de Sá, 1916, pp. 731-4; de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, pp. 8-9.
Plasmodium columbæ, Coatney & Roudabush, 1936, p. 339.

Schizogony said to take place in the plasma of the blood in the lung. Merozoites are seen in red blood-corpuscles as small irregular bodies with cytoplasm and nucleus. The

#### PROTEOSOMA.

large schizont reaches nearly a quarter the size of a red cell. Stained by Giemsa's stain the nucleus is bright red and the cytoplasm deep blue; some minute brownish-yellow pigment granules can be seen in the latter. It is then said to become free in the plasma, lose its pigment, assume a circular form, and stain a pale blue, with the chromatin central in position. This is called the "Acton body." The chromatin divides, forming two, and later more numerous, nuclear masses, some of which continue their further division, whilst others change into chromidial dust. The "Acton body "grows, and eventually all the nuclei disappear, and in its interior no nucleus, but only idio-chromidia, can be seen. This stage, called the meroblast, gives origin directly to the merozoites (corps en rosace of French authors, roseta of Portuguese authors). The roseta breaks up, and the merozoites infect fresh red corpusces.

Sexual forms are seen in the peripheral blood. Gamete formation, their structure and distinction into male and female forms, as described by Acton and Knowles for *Hæmoproteus* columbæ. Young gametes can be recognized as male or female by their staining reaction, form, situation and arrangement of chromatin, and the quantity and size of pigment granules. Gametes are halter-shaped, and embrace the nucleus of the infected blood-corpuscle.

Habitat.—Blood of Columba sp. : PORTUGUESE INDIA, NOVA Goa.

# 214. Proteosoma gallinulæ (de Mello). (Fig. 129.)

†Plasmodium gallinulæ, de Mello, 1935 a, pp. 352-3, pl. xlii, fig. 3; de Mello, 1937 a, p. 98.

Young ring-forms very regular. Full-grown trophozoites larger in size and oval in form. Large amœboid trophozoite provided with brownish-black pigment set free by the bursting of the corpuscle. Gametocytes show a clear sexual dimorphism. Male gametocytes are roundish or oval, alveolar; nucleus



Fig. 129.—Proteosoma gallinulæ (de Mello). A, ring-form; B, fullgrown trophozoite; C, free amœboid trophozoite; D, E, male gametocytes; F, G, female gametocytes. (After de Mello.)

consisting of many chromatinic rods irregularly connected together; pigment granules smaller, more or less scattered through the cytoplasm. Female gametocytes stain a compact and deep blue, have a large nucleus, and the pigment granules are larger. Infection very scanty. Infected red cells hypertrophied.

Habitat.—Blood of the moor-hen, Gallinula chloropus Linn.: PORTUGUESE INDIA, lake of Carambolim.

#### 215. Proteosoma herodiadis (de Mello). (Fig. 130).

*Plasmodium herodiadis*. de Melle, 1935 a, p. 351, pl. xlii, fig. 1 ; de Mello, 1931 a, p. 98.

Ring form with a granular or rod-like chromatinic dot. Older trophozoites pyriform or rounded, with irregularly scattered brownish-black pigment. Rosettes with five to ten chromatinic bodies. Male gametocytes somewhat oval, with a larger, irregular nucleus. Female gametocytes roundish, with a round nucleus. Infection not very heavy. Parasitized red cells not altered.



Fig. 130.—Proteosoma herodiadis (de Mello). A, ring-form; B, C, pyriform or rounded trophozoites; D, schizont; E, male gametocyte; F, fomale gametocyte. (After de Mello.)

*Remarks.*—This form is said to be distinguishable from *Hæmoproteus herodiadis*, found in the same host, by the nucleus being stained a vivid red colour in the former and a pale rose colour in the latter.

Habitat.—Blood of the heron, Egretta intermedia intermedia Wagler: PORTUGUESE INDIA, lake of Carambolim.

#### 216. Proteosoma heroni (Basu).

†Plasmodium heroni, Basu, 1938, p. 244.

*Remarks*—Morphological description of the species has not yet been published. Infection is easily transmitted from one paddy-bird to another by blood inoculation, and the disease set up is severe.

Habitat.—Blood of the paddy-bird commonly known as "pond heron": BENGAL, Calcutta.

#### 217. Proteosoma moruony (de Mello & de Sá). (Fig. 131.)

*Hæmoproteus moruony*, de Mello & de Sá, 1916, pp. 734-5, pl. lxiv, figs. A, 1-13, B, 1-18; de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, p. 9. *Hæmoproteus moruoni*, Wenyon, 1926, p. 1370.

Hæmoproteus moruony, de Mello, 1937 a, p. 99.

Sehizogony said to take place in the plasma. Merozoite enters a red blood-corpuscle and assumes a circular form, with a small chromatinic mass in the centre. Pigment granules are present. Large schizonts not seen in the body of erythrocytes, and the largest form in a red cell does not reach a quarter of the size of the infected cell. The schizont becomes free in the plasma, as the "Acton body." It has no visible pigment, and is surrounded by a very delicate hyaline capsule. The chromatin undergoes binary division, forming eight chromatin dots. Later, all the chromatin changes into idiochromidia and the capsule eventually disappears, the merozoites constituting the *roseta*.

Gamete formation, their structure, and differentiation into male and female similar to that in *Hæmoproteus columbæ*.



Fig. 131.—Proteosoma moruony (de Mollo & de Sá). A, young trophezoite; B, full-grown trophozoite; C, "Acton body," showing nuclear division; D, schizont: E, rosetta containing morozoites. (After de Mello and de Sá.)

The nucleus in an adult gamete is situated in the convexity of the halter and nearer to one pole than the other. The chromatin of the macrogamete is described as undergoing division for regressive schizogony.

Remarks.—De Mello and de Sá, though describing this parasite as a Hæmoproteus species, remark that all the stages of its development are seen in the peripheral blood, and no special forms are to be found in the internal organs.

Habitat.—Blood of the magpie robin, Copsychus saularis (Linn.): from INDIA, in the Zoological Gardens, London; also from the same host: PORTUGUESE INDIA, Calangate (Bardez).

218. Proteosoma præcox (Grassi & Feletti). (Fig. 132.)

Hæmamæba præcox, Grassi & Feletti, 1890, p. 463. Hæmamæba relicta, Grassi & Feletti, 1890, p. 465. Hæmoproteus danilewskii (part), Kruse, 1890, p. 371. Hæmoproteus, vars. b & c, Celli & San Felice, 1891, pp. 541-8, pls. vii, viii. Proteosoma grassii, Labbé, 1894, p. 157, pl. ix, figs. 1-31.

\*Proteosoma sp., Ross, 1898, pp. 401-8, 448-50; 1899 a, pp. 1-3; 1899 b, pp. 136-44; Daniels, 1899, pp. 443-54.

Hæmproteus danilewskyi, Labbé, 1899, p. 80, fig. 146.

Proteosoma sp., Schaudinn, 1900, pp. 159-81; Grassi, 1900. pp. 115-24; Ruge, 1901, pp. 187-91, 2 figs.; Hartmann, 1907, pp. 148, 149, 152.

Plasmodium priecox, Minchin, 1912, p. 358. Plasmodium relictum, Minchin, 1912, p. 358.

Proteosoma præcox, Doflein, 1916, pp. 897-901, figs. 896-900, 903.

- †Proteosoma præcox, de Mello, de Šá, de Sousa, Dias, & Noronha, 1917, p. 15.
- *Plasmodium danilewskyi*, Castellani & Chalmers, 1919, p. 513.
- Protcosoma præcox, Calkins, 1926, p. 444.
- †Plasmodium præcox, Scott, 1926, p. 237; Wenyon, 1926, pp. 1365, 1385.
  - Plasmodium præcox, Wenyon, 1926, pp. 977-8, pl. vi, figs. 1-10; Hegner, 1926, pp. 479-80; Hartmann, 1927, pp. 1-7, pls. i, ii; Knowles, 1928, pp. 442-3, fig. 102, 1-4; Hegner, 1929, pp. 59-82.

Proteosoma præcox, Reichenow, 1929, pp. 983-4, figs. 952-4, 1000. Plasmodium præcox, Manwell, 1930, pp. 382-3; Kudo, 1931, p. 287, fig. 122 a-d.

Plasmodium relictum, Ed. & Et. Sorgent & Catanoi, 1930, pp. 223-38; 1931 a, pp. 399-437; 1931 b, pp. 327-35.

Plasmodium præcox, Giovannola, 1934, pp. 372-8, pl. i, figs. 1-4; Coatney & Roudabush, 1936, p. 340; de Mello, 1937 a, p. 95.

Schizogony takes place in the red blood-corpuscles, generally in the viscera. Young trophozoites resemble the small rings



Fig. 132.—Proteosoma præcox (Grassi & Feletti). (×2000.) A, young ring-shaped trophozoite; B, C, larger trophozoites; D, schizont splitting into a number of merozoites; E, female gametocyte; F, male gametocyte. (After Wenvon.)

of Laverania malariæ. As growth proceeds, dark pigment granules appear, and the host-cell becomes distorted, its nucleus becoming displaced more or less to one side or to one end of the cell (unlike Hæmoproteus, which develops in the red blood-corpuscle without deforming it or displacing the nucleus to any extent). Schizont varies in size and period of growth. Small schizonts measure  $4-5\mu$ , and produce only about six merozoites. Largest schizonts occupy about half the corpuscle and produce 16 to 24 merozoites.

the average number of merozoites formed being 15; in such cases the infected corpuscles are altered in shape, pale in colour, and have the nucleus displaced to one side of the corpuscle. Sometimes two or more schizonts occur in a single cell, and a crop of as many as 36 merozoites may result. In the fully developed schizont the pigment is seen as a single dark mass. Gametocytes are slightly elongate ovoid bodies about the size of the largest schizonts, and closely resemble those of Laverania. The female gametocyte stains more deeply and has a more compact nucleus than the male. Infection is transmitted from bird to bird by *Culex* mosquitoes. Sporogony is similar to that of the human malarial parasites in Anopheles, the time occupied by the cycle varying with temperature. Number of occysts varies with the number of gametocytes in the blood, in some cases the whole wall of the stomach of the mosquito being studded with the occysts.

The parasite usually occurs in the blood of small birds like the sparrow and lark, but can also be found in the blood of larger birds such as the crow, owl, pigeon, partridge, duck, fowl, etc.

*Remarks.*—The species is of special interest, as it was while investigating it that Ross (May, 1898) first discovered the stages of fertilization and occyst formation in the stomach of Culex sp. (probably C. fatigans Wied.). In June and July of the same year Ross discovered the liberation of sporozoites into the coelomic cavity of the mosquito and the invasion of the salivary glands, and succeeded in infecting healthy birds from infected *Culex* mosquitoes. He thus discovered the whole of the transmission cycle in P. pracox. He had previously (1897) discovered oöcysts of human malarial parasites in Anopheles mosquitoes, and predicted that the transmission cycle of human malarial parasites would be found to occur in Anopheles mosquitoes, and that it would be identical with that of P. præcox in Culex, thus paving the way for the discovery of the complete life-cycle of the human malarial parasites by the Italian workers.

Daniels (1899), working at Calcutta, confirmed Ross's observations. Koch, working in Italy (1899 a), studied the development in *Culex nemorosus* (= $A\ddot{e}des$  nemorosus), and Grassi (1899) in *C. pipiens*. Lühe (1900) clearly brought out the relation of the life-cycle of malarial parasites to that of COCCIDIA, and introduced a terminology for various stages that is in vogue till to-day. Ed. and Et. Sergent (1907) and Neumann (1908) demonstrated that the complete cycle could take place in a small proportion of  $A\ddot{e}des$  ægypti (Linn.) (=Stegomyia fasciata) that were fed on infected blood. Ed. and Et. Sergent showed later (1918) that the development could take place in certain other species of  $A\ddot{e}des$  and *Culex* 

also, and they (1919) carried out experiments on the development of *P. præcox* under varying conditions of temperature.

Celli and San Felice (1891) were the first to show that the infection could be transmitted from lark to lark by inoculation of blood from an infected bird to a healthy one. Grassi and Feletti (1891) and Laveran (1891) repeated the experiment in larks, and Ziemann (1898) in another type of bird. Ross (1898 b) showed that mosquitoes fed on sparrows were able to infect crows and larks. Koch (1899 a, 1899 b) showed that parasites naturally occurring in one type of birds can be inoculated into other types of birds. Ruge (1901) and Wasielewski (1908) were similarly able to infect various types of birds, thus showing that the same species of Proteosoma is able to infect a number of different genera and species. According to Huff (1927) infections had been noted in at least seventy-nine species of birds belonging to at least fifteen families. Originally the malarial parasites of birds were all considered as belonging to a single species, but they have now been split up into a number of species by Hartmann (1927), Huff (1930), Manwell (1930) and others, and parasites from a large number of different birds have also been described as distinct species. Mayne (1928) has recorded both natural and experimental infections in Anopheles subpictus Grassi (=rossii). According to Giovannola (1934) no less than four species are known to occur in Passer domesticus (Linn.), viz., P. præcox Grassi & Feletti, P. cathemerium Hartmann, P. elongatum Huff, and P. rouxi Ed. & Et. Sergent & Catanei. According to him P. præcox is distinguishable by spheroidal gametocytes containing pointed pigment granules, and spheroidal schizonts producing 14 to 32 merozoites.

In recent years a good deal of experimental work has been carried out on infection and resistance in bird malaria (Boyd, 1925; Taliaferro, 1925; Hegner, 1926), on relapses in bird malaria (Manwell, 1929), and on quinine and plasmochin therapy (Boyd, 1926; Hegner, Shaw, and Manwell, 1928; Manwell, 1930), resulting in the discovery of facts which are of considerable practical importance in the warfare against human disease.

"Black spores" of Ross.—Certain bodies described as "black spores" were originally noted by Ross (1899, 1905, 1923) and Daniels (1900) within oöcysts in the stomach of infected mosquitoes, as well as free in the tissues, and even in uninfected mosquitoes. According to Stephens and Christophers (1908) they are also found in the thoracic muscles and salivary glands. Castellani and Chalmers (1919) regard these so-called "black spores" as protozoal parasites of the genus Nosema, which have invaded the oöcyst.

Bruce Mayne (1929), after reviewing the previous literature on the subject, and after recording his observations to the effect that these "black spores" have been found during experiments with avian malaria in uninfected mosquitoes, as well as in mosquitoes harbouring the parasites of malaria, and even in freshly emerged, unfed, laboratory-bred females and males of Anopheles, Culex, and Musca, comes to the conclusion that most, if not all, "black spores" appear to be merely chitinogenous thickenings of the tracheal tubes. Knowles and Basu (1933) conclude that no less than three, if not four, different structures have been described as "black spores" or "chitin corpuscles" by different workers. These are: (a) True malaria oöcysts which have undergone degeneration, and in which a very heavy deposition of pigment has occurred ; these alone they believe to be the true "black spores" of Ross; (b) a hyper-chitinization of segment-like portions of the finer ramifications of the tracheal system; (c) fungus infections of the tracheal system; (d) infections of the mosquito tissues concerned with miscrosporidian parasites. Their observations, based upon material studied in sections, led them to conclude that (b), (c), and (d) have nothing to do with true "black spores," which had better be described as "degenerated and hyper-pigmented occysts" or ruptured contents of the same.

Habitat.—Blood of sparrows, larks, pigeons, crows : BENGAL, Calcutta ; some birds : CEYLON ; oöcysts in mosquitoes, Culex fatigans Wied., fed on these birds ; also in Anopheles subpictus Grassi (=rossii) : also blood of white-throated munia, Uroloncha malabarica Linn., and common Indian starling, Sturnus vulgaris poltaratskyi Finsch, from INDIA, and Tragopan satyra (Linn.), from the HIMALAYAS, in the Zoological Gardens, London.

# 219. Proteosoma sp.

†Plasmodium (?) sp., Scott, 1926, p. 237; Wenyon, 1926, p. 1365. Plasmodium sp., Coatney & Roudabush, 1936, p. 341; de Mello, 1937 a, p. 95.

Habitat.—Blood of the sarus crane, Antigone antigone (Linn.), from INDIA, in the Zoological Gardens, London.

# 220. Proteosoma sp.

Plasmodium sp., Donovan (first recorded in Wenyon, 1926, p. 1371).

Plasmodium sp., de Mello, 1937 a, p. 95.

Habitat.—Blood of the Indian house crow, Corvus splendens Vieillot : INDIA (locality not cited).

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#### 221. Proteosoma sp.

†Plasmodium sp., Plimmer, 1914, p. 190.

Plasmodium sp., Wenyon, 1926, p. 1371; Coatney & Roudabush, 1936, p. 342.

Habitat.—Blood of the barbet, Cyanops flavifrons (Cuvier), from CEYLON, in the Zoological Gardens, London.

#### 222. Proteosoma sp.

†Plasmodium sp., Plimmer, 1913, p. 148.

Plasmodium sp., Wenyon, 1926, p. 1372; Coatney & Roudabush, 1936, p. 342; de Mello, 1937 a, p. 95.

Habitat.—Blood of the grey-headed bunting, Emberiza fucata Pallas, from INDIA, in the Zoological Gardens, London.

#### 223. Proteosoma sp.

†Plasmodium sp., Plimmer, 1912, p. 415; Scott, 1925, p. 237; Wenyon, 1926, p. 1372.

Plasmodium sp., Coatney & Roudabush, 1936, p. 342; de Mello, 1937 a, p. 95.

Habitat.—Blood of the wattled starling, Gracula indica (Cuvier) [=Eulabes religiosa Auct.], from SOUTH INDIA, in the Zoological Gardens, London.

#### 224. Proteosoma sp.

†Plasmodium sp., Plimmer, 1912, p. 415.

Plasmodium sp., Coatney & Roudabush, 1936, p. 342; de Mello, 1937 a, p. 95.

Habitat.—Blood of the jay-thrush or babbler, Garrulax leucolophus (Hardwicke), from NORTH INDIA, in the Zoological Gardens, London.

#### 225. Proteosoma sp.

*Plasmodium* sp., Scott, 1926, p. 237, Wenyon, 1926, p. 1377. *Plasmodium* sp., de Mello, 1937 a, p. 95.

Remarks.—Laveran and Marullaz (1914) have described Proteosoma (Hæmamæba) tenue and Proteosoma (Hæmamæba) liothiris from the same host from Japan.

Habitat.—Blood of the babbler, Leiothrix lutea (Scop.), from INDIA, in the Zoological Gardens, London.

# 226. **Proteosoma** (?) sp.

†Plasmodium (?) sp., Scott, 1926, p. 237; Wenyon, 1926, p. 1377. Plasmodium sp., Coatney & Roudabush, 1936, p. 343.

Habitat.—Blood of the Ceylon loriquet, Coryllis beryllinus (Forster) [=Loriculus indicus Gmel.], from CEYLON, in the Zoological Gardens, London.

#### 227. Proteosoma sp.

†Plasmodium sp., Plimmer, 1913, p. 148.

Plasmodium sp., Wenyon, 1926, p. 1377; Coatney & Roudabush, 1936, p. 343; de Mello, 1937 a, p. 95.

Habitat .- Blood of the bunting, Melophus melanicterus (Gmel.), from INDIA, in the Zoological Gardens, London.

# 228. Proteosoma sp.

+Plasmodium sp., Plimmer, 1915, p. 130.

Plasmodium sp., Wenyon, 1926, p. 1377; de Mello, 1937 a, p. 96. Habitat.-Blood of the thrush, Turdus boulboul (Latham), from INDIA, in the Zoological Gardens, London.

#### 229. Proteosoma sp.

†Plasmodium sp., Plimmer, 1912, p. 415. Plasmodium sp., Wenyon, 1926, p. 1383; de Mello, 1937 a, p. 96.

Habitat.-Blood of the bulbul, Elathea jocosa (Linn.) [=Otocompa eimeria], from INDIA, in the Zoological Gardens, London.

# 230. Proteosoma sp.

†Plasmodium sp., Plimmer, 1913, p. 148. Plasmodium sp., Wenyon, 1926, p. 1382.

Habitat.-Blood of the chat, Saxicola caprata (Linn.), from INDIA, in the Zoological Gardens, London.

#### Genus LAVERANIA Grassi & Feletti, 1890.

A third type of malarial parasite, Golgi, 1886 a, p. 109; 1886 b, p. 419; 1889, p. 173.

Malignant tertian parasite, Marchiafava & Celli, 1889. Laverania, Grassi & Feletti, 1890, p. 4; 1892, p. 10. Hæmatomonas, Hehir, 1893, pp. 207-12, 241-8, 273-80.

Hæmatozoon, Welch, 1897.

Plasmodium (part), Labbé, 1899, pp. 80-2; Lühe, 1900, p. 392; Schaudinn, 1902.

Laverania, Neveu-Lomaire, 1900, p. 9; Minchin, 1903, pp. 243, 267.

Hæmamæba (part), Laveran, 1907, pp. 110-60.

Plasmodium (part), Minchin, 1912, p. 358.

Laverania, Doflein, 1916, pp. 902-12; Castellani & Chalmers, 1919, pp. 517-18.

Plasmodium (part), Mühlens, 1921, pp. 1502–11. Laverania, Thomson & Woodcock, 1922, pp. 1530–4.

Plasmodium (part), Hegner & Taliaferro, 1924, pp. 327-9; Wenyon, 1926, pp. 934-41; Knowles, 1928, pp. 392-400.

Laverania, Stiles, 1928, p. 881; Reichenow, 1929, pp. 987-98.

Plasmodium (part), Kudo, 1931, p. 286; Calkins, 1933, pp. 406, 407, 566.

Laverania, Reichenow, 1935, p. 377.

Plasmodium (part), Brumpt, 1936, pp. 418-24, Coatney & Roudabush, 1936, pp. 338-53.

Trophozoites very small rings. Schizonts also remain small s 2

and do not completely fill the red-blood corpuscles. Hæmozoin pigment granular. Gametocytes elongated, wormlikeand somewhat sickle-shaped, so-called "crescents." Parasites of man and anthropoid apes.

Remarks .-- Laverania malariæ, the parasite of malignant tertian or tropical epidemic malaria in man, and Laverania reichenowi, the parasite of the gorilla and chimpanzee, are included in this genus. The species of this genus show considerable resemblance with *Proteosoma* of birds.

## 231. Laverania malariæ Grassi & Feletti\*. (Figs. 122 (Pl. I), 133, & 134).

Malarial parasites, Laveran, 1880 a, p. 158; 1881, pp. 627-30; 1882, p. 737.

Oscillaria malariæ (part), Laveran, 1883, p. 113.

A third type of malarial parasite, Golgi, 1886 a, p. 109; 1886 b, p. 419; 1889, p. 173.

Hæmamæba malariæ, Laveran, 1890, p. 374.

Hæmamæba malariæ præcox+H. malariæ immaculatum, Grassi & Feletti, 1890, p. 10.

Hæmamæba præcox, Grassi & Feletti, 1890, p. 6; 1892, p. 10.

Laverania malariæ, Grassi & Feletti, 1890, p. 4; 1892, p. 10.

Hæmamæba febris quotidinæ, Marchiafava & Bignami, 1891.

- Hæmamæba immaculata, Grassi, 1891, p. 14; Grassi & Feletti, 1892, p. 10.
- +Hæmatomonas malariæ, Hehir, 1893, pp. 207-12, 241-8, 273-80, pls. i–viii.

Hæmatozoon falciparum, Welch, 1897, pp. 36, 47.

Plasmodium malariæ præcox, Labbé, 1899, p. 82. Plasmodium malariæ immaculatum, Labbé, 1899, p. 82.

Hæmomenas præcoz, Ross, 1899, pp. 322-4, 439; 1900, pp. 522-7. Plasmodium præcoz, Lühe, 1900, p. 460. Laverania præcoz, Neveu-Lemaire, 1900, p. 9, pl. i, fig. 37.

Laverania malariæ, Minchin, 1903, pp. 243-54, 267, 332, 351, fig. 68.

Hæmamæba malariæ, Laveran, 1907, pp. 110-60; fig. ii, pl. i.

Plasmodium præcox, Doflein, 1909, pp. 662-70, figs. 596-609.

Laverania malariæ, Doflein, 1909, pp. 662-70, figs. 596-609.

Plasmodium falciparum, Bass & Johns, 1912, pp. 567-79; Minchin, 1912, pp. 358-60, fig. 156.

Malignant tertian malarial parasites, Thomson & Thomson, 1913, pp. 77-87, pl. x.

†Plasmodium falciparum, Row, 1917, p. 392, pl. xxiii.

†Laverania præcox, Row, 1917, p. 392, pl. xxiv.

Laverania malariæ, Castellani & Chalmers, 1919, pp. 517-18, figs. 174, 175, pl. i, figs. 1 c-4 c.

Plasmodium immaculatum, Mühlens, 1921, pp. 1502-11, pl. xxxi, figs. 20-50; pl. xxxii, figs. 26-40; pl. xxxiii, figs. 2, 5-21; text-figs. 4-7.

\* The retention of the genus Laverania for the parasite of malignant tertian Malaria is in accordance with Opinion 104 of the Intern. Comm. on Zool. Nomenclature. Many malariologists, however, consider that the separation of this parasite from those of benign tertian and quartan Malaria is not required on zoological grounds, and include all three species in the genus Plasmodium ; if this latter classification be followed, the name of the malignant tertian parasite will be Plasmodium falciparum (Welch).-EDITOR.

Laverania malariæ, Thomson & Woodcock, 1922, pp. 1530-6, pls. lxv, lxvi; text-figs. 545, 546.

Plasmodium falciparum, Hegner & Taliaferro, 1924, pp. 327-9, pl. ii, figs. 9-16.

Laverania falciparum, Stiles & Hassall, 1925, p. 42.

- Plasmodium falciparum, Craig, 1926, pp. 433-52, figs. 74-8; Wenyon, 1926, pp. 934-41, pls. vii-ix, pl. xiii, figs. 16-40; figs. 391, 401-3; Hehir, 1927, pp. 166-73, pl. xii, figs. 76-117; pl. xiii, figs. 118-29; pl. xiv, figs. 1-8; figs. 59-60.
- †Plasmodium falciparum, Knowles, 1927, pp. 14-20, pl. iii, figs. 1-24; 1928, pp. 392-400. pl. xiii.
- Laverania malaria, Reichenow, 1929, pp. 987-97, figs. 958-73, 975 a - e.
- †Plasmodium falciparum, Row, 1929, pp. 1120-5, pls. lxxxviii, lxxxix; 1930, pp. 221-6, pls. xxiii, xxiv.
- Plasmodium falciparum, Aragão, 1930, pp. 41-56, 1 pl.; Kudo, 1931, p. 286, fig. 121, *k-n*; Calkins, 1933, p. 407, pl. i, figs. 13-17; pl. ii, figs. A-L; Chaoulitch, 1936, pp. 716-22; Brumpt, 1936, pp. 418-24, figs. 185-7; Coatney & Roudabush, 1936, p. 339. Plasmodium immaculatum, Giovannola, 1935, pp. 90-1.

<sup>†</sup>Plasmodium tenue, Stephens, 1914, pp. 169-72.

- The so-called Plasmodium tenue, Balfour & Wenyon, 1914, p. 353.
- *Plasmodium tenue*, Sinton, 1922, pp. 215-35, pls. ii & iii ; Knowles, 1923 a, p. 276
- Plasmodium tenue, Wenyon, 1926, pp. 951-2, fig. 405; Craig, 1926, pp. 464-70, fig. 81.

†Plasmodium tenue, Knowles, 1928, p. 404, fig. 93.

Plasmodium tenue, Reichenow, 1929, p. 988, fig. 961.

The Cycle in Man.—Young trophozoites are very small, often seen as a tiny bead of chromatin, with a wisp of bluestaining cytoplasm adhering to the margin of the red corpuscle (the marginal or accolé form). Multiple infection of red blood-corpuscles is very common, many of them containing two, three, or sometimes a larger number. The smallest "rings" are very narrow, appearing as a thin blue line surrounding a vacuole, with a red-staining granule of chromatin or nucleus protruding at one side, occupying no more than one-sixth of the diameter of the red blood-corpuscle. Chromatin often rod-like, and divides very early into two granules (nuclei). Fully grown trophozoites may be as large as half the size of the red blood-corpuscle, and the cells containing these big rings show Maurer's dots. Occasionally more irregular or amœboid forms occur. Infection is usually heavy, as many as 25 per cent. of the corpuscles being found infected. Infected red corpuscles do not become enlarged (as they do in infections with *Plasmodium vivax*), but on their surface appear red-staining markings usually known 88 Maurer's dots (the markings were actually first described by Stephens and Christophers, who regarded them as clefts in the red corpuscle in which the red of the stain is deposited). Maurer's dots are larger and fewer in number than Schüffner's dots (which are found in P. vivax), and are best seen in deeply stained films : they are stellate or crack-like rather than dotlike, and stain brick-red rather than the bright pink of



Fig. 133.—Life-cycle of Laverania malariæ Grassi & Feletti in man and the mosquito. (× c. 1000.) A-G, schizogony in red blood-corpuscles; H, immature gametocyte;  $I_1$ ,  $I_2$ , male and female gametocytes in blood of man;  $J_1-L_1$ , development of male gametocyte and production of microgametes;  $J_2-L_2$ , development of female gametocyte and extrusion of polar body in the stomach of mosquito; M, fertilization; N, transformation of zygote into oökinete which penetrates through the intestinal epithelium; O, P, development of oöcyst between the epithelium and the elastic membrane; Q-S, growth of cytoplasm accompanied by multiplication of nuclei and vacuolation of cytoplasm; T, vacuoles run together to reduce cytoplasm Schüffner's dots. Sooner or later hæmozoin pigment appears in the growing trophozoite, the pigment being dark brown or black, and is collected into a distinct round mass, and this early collection of the pigment into a round compact mass in the preschizogonic stages is a diagnostic feature of the asexual forms of this species.

All the infected corpuscles soon disappear from the peripheral circulation, and schizogony takes place almost exclusively in the internal organs. Sometimes this takes place before pigment has formed, at others not till some growth has taken place and pigment granules have developed in the cytoplasm. It was probably this that led the earlier observers to describe two separate species of malignant tertian parasities (P. malariæ præcox and P. malariæ immaculatum). The pigment granules run together to form a dark granular mass at one side of the parasite. The fully developed schizont occupies one-half to two-thirds of the diameter of the red blood-corpuscle, and gives rise to 8 to 24, usually 10 to 20, very small merozoites. Sometimes a larger number, up to 32, are set free, but this may be due to two schizonts being contained in a single corpuscle. The merozoites are arranged in a grape-like cluster ("rosette") round a central residual mass, with a considerable amount of pigment.

The attack of malaria synchronizes with the setting free of the merozoites, but the three stages of the attack are not so sharply defined as in the case of P. vivax infections. The fever may occur every forty-eight hours (tertian) or every twenty-four hours (quotidian). In the latter case it is probably due to two infections having originally taken place, schizogony due to each taking place on alternate days, causing a daily attack of fever. In certain cases of heavy infections schizonts are seen in the peripheral blood along with the trophozoites; and this is usually an indication that the infection is a very serious one and may prove fatal. Schizogony may be seen in a certain number of ordinary infections in thick blood-films; but it can, however, be studied best in cultures or by carrying out spleen punctures during the acute phases of the attack. After schizogony, which, as remarked above, usually takes place in the internal organs, ring-forms appear again in the peripheral blood.

> to a coarse network, with sporozoites commencing to form as finger-like buds; U, sporozoites fully formed and attached to several masses of cytoplasm into which the network has broken, two residual masses containing nuclear matter also present; V, detached sporozoites irregularly distributed in the oöcyst, which ruptures, liberating them into the colonic cavity; W, sporozoites entering the salivary glands; X, sporozoites injected into man by the mosquito: they invade the red bloodoorpuscles and start the schizogony cycle. (After Wenyon.)

Merozoites grow into gametocytes almost exclusively in the blood-stream in the internal organs. Only mature gametocytes are generally seen in the peripheral blood, though occasionally young forms may also be found. Young gametocytes are slightly elongate, with one side concave and the other convex. Fully developed gametocytes are crescentic or sausage-shaped, and are generally referred to as " crescents." They are about one and a half times the length and about half the breadth of a red corpuscle. They usually have rounded ends, but sometimes the ends are pointed, and the "crescents" are then sickle-shaped. In deeply stained films the margin of the blood-corpuscle is seen to be closely applied to the convex side of the crescent and stretched across the concave side to show a bulge. Sometimes the crescent appears to be completely surrounded by a somewhat irregular red-staining border, which J. D. Thomson (1917) interpreted as a definite capsule. Two types of crescents are found. The male, or microgametocyte, is shorter and broader than the female, and has more rounded ends; it has hyaline cytoplasm, which stains a faint blue or a faint dirty pink colour with Romanowsky stains; the nucleus is larger and paler when stained, and the pigment is irregularly distributed throughout the middle two-thirds of the crescent. The female gametocyte is thinner and longer and has more pointed ends; its cytoplasm is denser and stains more deeply blue; the nucleus is compact and stains more intensely, and the pigment is aggregated in the centre of the crescent round the nucleus. In well-stained, wetfixed films the nucleus of the female gametocyte is seen to have a large karyosome, while that of the male has fine chromatin granules distributed through it. The number of crescents varies considerably ; sometimes a single crescent will be found after a prolonged search, at other times they are so numerous that one or more will be found in nearly every field. Relapses are less common than in the benign tertian fever, and the infection as a rule dies out more quickly.

The Cycle in the Mosquito.—As in the other species of human malarial parasites, the gametocytes undergo their further development in the stomach of Anopheline mosquitoes. There the crescents retract to a spherical form, thus causing the rupture of the remains of the red blood-corpuscles, and escaping out of them. Subsequent development and fertilization takes place as in the other species. The male gametocyte gives rise to a number of microgametes by ex-flagellation, and the female gametocyte forms a single macrogamete. Macroand microgametes unite in pairs, and zygotes, called oökinetes, are formed, which, after penetrating the stomach-wall, become lodged between the epithelium and the elastic layer of the wall of the stomach. There they grow; the nuclei undergo rapid and repeated divisions, and finally produce an enormous number of minute sporozoites. These sporozoites are set free through the rupture of the cyst-wall into the body-cavity, find their way into the salivary glands, and are inoculated into a new victim when the mosquito bites one. The oökinetes and the developing oöcysts of this species are distinguished



Fig. 134.—Oöcysts of Laverania malariæ Grassi & Feletti projecting from the wall of the stomach of an infected Anopheles; c, crop; m.t., Malpighian tubules; o, oöcysts of Laverania; r, ventral reservoir; s, stomach, the enlarged portion of the mid-gut; s.gl., salivary glands. (From Reichenow, after Ross and Grassi.)

from those of P. vivax by the dark colour of the pigment in the former and the light brown or yellow pigment in the latter. Also a much larger number of occysts occurs, and they

may be so numerous as to be actually in contact with one another on the stomach-wall.

Remarks.—There is a vast literature on this, as well as the other species causing malaria in man, and it is difficult to refer to even the more important works here. Sinton (1929) has published a very valuable and complete bibliography on malaria in India, indexing about 2200 papers and reports dealing with the causation, prevention, and treatment of the disease. Covell (1927, 1931) has similarly collected all the available data relating to the transmission of malaria by different species of Anopheline mosquitoes. For information on the subjects of pathology of malaria infections, the occurrence and mechanism of relapses, the factors which influence the development and survival of malarial parasites in mosquitoes, the possibility of animal reservoirs and the susceptibility of man and animals to inoculation, reference may be made to text-books and other special works.

L. malariæ is usually present in tropical countries, and is responsible for severe outbreaks of epidemic malaria. The fever is referred to as malignant, as it does not yield readily to treatment. Two types are known, a quotidian, which recurs every day, and a tertian, which recurs every other day. Whether or not these are due to distinct species or the quotidian is due to double infection is still uncertain.

Knowles (1919) noticed crescents, without asexual forms, in blood-films taken from a convalescent malaria patient. It is quite common to come across crescents and other forms in blood films from persons living in an endemic area who are quite free from all symptoms of malaria at the time. Knowles, Chopra, Gupta, and Das-Gupta (1923) record the examination of twelve villagers from the Kuki Hills in Assam admitted to hospital in Calcutta suffering from frambœsia. All except one were afebrile, and yet blood examination showed that eleven out of twelve patients harboured malarial parasites. all three species being found among different patients, and both trophozoites and gametocytes were encountered. Sinton (1926) has made a thorough study of the problems relating to game to cyte formation. He finds that (a) there is a marked seasonal variation in crescent production in patients; (b) different localities show a different proportion of crescent-carriers; (c) there is a distinct correlation between the numerical prevalence of asexual forms in the peripheral blood and the number of crescents which appear about ten days later; (d) the development of crescents appears to be associated with a lowered immunity, possibly due to a change in the reaction of the pulp in the bone-marrow and spleen; (e) no marked correlation can be traced between the degree of splenic enlargement and the occurrence of crescents in the peripheral blood; (f) duration of life of a crescent may be as long as forty to **h**fty days in the peripheral blood, but the great majority of them disappear within three weeks after the asexual cycle has been destroyed; and (g) the reduction in the number of crescent carriers by efficient treatment is an important factor in all anti-malarial campaigns.

Row (1929), discussing the subject of evolution of the crescents of malarial parasites grown anærobically in simple cultures, recorded definite variations during different attacks of the same infection. Thus, when a man fresh from Europe, and otherwise healthy, is infected, one may find in his first paroxysm a very severe clinical reaction, and yet the number of parasites in his peripheral blood is very small. These, however, yield a larger number of merozoites in culture, whereas during the second and third paroxysms, though the number of parasites in his peripheral blood may be very large, the number of merozoites yielded in culture is much smaller, and this diminution progressively continues for each paroxysm until a time is reached when one merozoite entering a red blood-corpuscle produces no more than one individual. This is the point when the crescent and gamete production is initiated, and, once started, it continues, so as ultimately to flood the blood with these resistant forms of the parasite, which persist for several weeks during the apparent well-being of the patient. The factors contributing to this change in the direction of parasitic development have been observed to depend (a) on the vigour of the phagocytes, and (b) on the biochemical action of the plasma or serum in which the free merozoites find themselves either after the rupture of the fully developed schizont or after their escape from the phagocytes or on (a) and (b) combined. Row described the phagocytosis in leucocytes and plasmolysis by blood serum of such of the merozoites as are liberated by degenerating leucocytes. In vivo these liberated merozoites probably initiate a fresh paroxysm. The merozoites during each succeeding paroxysm become more resistant owing to their passage through the leucocytes, but lose their capacity to multiply, until the point is reached when one merozoite yields but one individual for each corpuscle attacked, and the crescent formation is inaugurated and continued. This is an ideal state of equilibrium between the host and the parasite. Row also described the actual development of crescents in cultures, and advanced evidence for the belief that antibodies are formed in a malarial infection. Row (1930) further showed that an accelerated effort toward crescent formation was brought about by using for culture a mixture of Laverania malariæ and Plasmodium vivax, and put forward the suggestion that in nature also such mixed infections may be one of the factors responsible for the presence of the crescents in carriers in endemic areas.
Aragão (1930) has studied the development of the gametocytes of *Laverania malariæ*, and finds that there are two types of merozoites, corresponding to the male and the female gametocytes. Merozoites destined to form male gametocytes after entering a corpuscle are spherical with a distinct nucleus and without the vacuole typical of ring forms, while those destined to give rise to female gametocytes stretch across the corpuscle in the form of an elongate bar.

Knowles and Das-Gupta (1931) observed scanty phagocytosis in a case of infection with L. malariæ, and Chopra, Das-Gupta, and Sen (1932) observed heavy phagocytosis of parasites by the polymorphonuclear and large hyaline leucocytes in another case. Stott (1933) has reviewed the previous literature on the phagocytosis of malarial parasites and recorded his own observations in two cases. In severe malignant tertian infection approximately 50 per cent. of red blood-cells may be infected, and in such cases merozoite formation may be seen in the peripheral circulation. Large mononuclear cells may show their phagocytic power, even in the peripheral circulation of severe cases, by the presence of (1) pigment, (2) rings, (3) red cells, normal or dehæmoglobinized, and non-infected or infected with sporulating or, less commonly, with other forms of malarial parasites.

Missiroli (1933), working with Culex fatigans infected with bird malaria, and Knowles and Basu (1935) with Anopheles stephensi infected with Laverania malariæ and Plasmodium vivax, have observed division of the nucleus of the sporozoites into two, three or more parts. The sporozoite is then said to divide into a corresponding number of small rounded bodies consisting almost entirely of chromatin with scarcely any visible cytoplasm. These apply themselves to the surface of the red corpuscles and presumably grow into the usual ringtrophozoite forms. If these observations are confirmed it would mean that the sporozoite is not the end of the sporogony cycle, but that occasionally, at any rate, it breaks up into smaller bodies which, after entering the corpuscles, grow into trophozoites.

Chaoulitch (1936) has recently observed that asexual multiplication can also take place by binary fission of the schizont.

Alessandrini (1933) reported a new method of reproduction for L. malariæ, i. e., simple amitotic division, occurring both in man and mosquito, but only when the resistance of the host is distinctly lowered by infection, adverse environmental influences, etc. This amitotic reproduction results in a more rapid and virulent multiplication of the parasite, accounting for the occasional pernicious character of the disease. Hingst (1934) has also adduced evidence to show that in some indi-

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viduals of L. malariæ repeated direct division and formation of two daughter organisms of equal size occurs, and according to him this explains the multiple infection of the red bloodcorpuscles so characteristic of the species. He believes that schizogony may follow direct division.

Boyd (1935) has discussed the comparative morphology of the sporozoites of the human malarial parasites. The form with dissimilar extremities appears most frequently in P. vivax, while in L. malariæ both ends tend to be pointed. The sporozoites of L. malariæ are the finest, those of P. malariæ the coarsest.

Cultivation.-Bass and Johns (1912) were the first to successfully cultivate this species outside the body, and to observe three generations of schizogony in these cultures. J. G. Thomson and D. Thomson (1913) introduced a reliable modification of Bass and Johns's technique. Row (1917) successfully cultivated the malarial parasites in blood drawn from the finger, and published his observation on the forms observed in culture of Plasmodium falciparum and Laverania præcox (synonyms of Grassi and Feletti's two subspecies of Laverania (Hæmamæba) malariæ). In the former he noted the small size of the infected corpuscle and the formation of six or less merozoites: in the latter the mature schizont formed four or less merozoites and occupied the whole of the infected corpuscle, which was much larger than the normal one. The schizogony cycle completed itself in twenty-four hours, and there was a marked tendency on the part of the developing parasite to agglutinate in larger or smaller masses. Later workers do not regard the two subspecies as distinct. Sinton (1922) described the culture of malarial parasites from the finger-blood in specially constructed glass tubes. The details of some of these methods will be found in the part dealing with practical methods.

"Plasmodium tenue."-Stephens (1914), under the name of Plasmodium tenue, described a malarial parasite seen by him in a single blood-film sent to him from the Central Provinces. Only young forms were present, and were distinguished by their marked amœboid form, large size of the nuclear chromatin, and markedly irregular shape. Balfour, Andrew, and Wenyon (1914) pointed out that such forms were not uncommon in Laverania malariæ, and concluded that P. tenue was not a distinct species. Knowles (1923 and 1926) expressed a similar Sinton (1922 b) found these forms first in five opinion. cases of malaria in the Central Provinces, and later in other cases at Lahore, and made out a strong case for the validity of P. tenue as a distinct species. Christophers (1925), in his study of malaria in Singhbhum, notes that the parasites encountered in that district resembled P. tenue rather than

Laverania malariæ. Callanan (1926) found "tenue" forms in blood-films containing Laverania malariæ, and noted that the "tenue" forms tended to preponderate in certain portions of the film, whereas ring forms showed a predilection for others. He concluded that near the edge of the blood-film, where the red corpuscles are evenly distributed and lie singly, pressure in spreading the film causes the delicate hair-like rings of Laverania malariæ to become distorted into band-like "tenue" forms. So the case for P, tenue as a distinct species is by no means proved.

Habitat.-Blood of man, in almost all parts of INDIA, and the body of the following species of mosquitoes :- Anopheles annularis Van der Wulp, A. culicifacies Giles (=A. fuliginosus), A. fluviatilis James (= A. listoni Liston), A. maculatus Theo., A. minimus Theo., A. philippinensis Ludl., A. stephensi Liston, A. sundaicus Rodenwaldt, and A. varuna Iyengar. For the localities in INDIA, BURMA, and CEYLON where Laverania malariæ infection is known to occur, the map reproduced from Knowles, Senior White, and Das-Gupta (1930) at the end of the description of malarial parasites of man (p. 279) may be consulted, and for the relative importance of various species of Anopheles in the transmission of malaria in different parts, the useful summaries taken from Covell (1931) and Christophers (1933) may be referred to.

## Genus PLASMODIUM Marchiafava & Celli, 1885.

Malarial parasites, Laveran, 1880 a, p. 158; 1881, pp. 627-30; 1882, p. 737.

Oscillaria (part), Laveran, 1883, p. 113.

Plasmodium, Marchiafava & Celli, 1885, p. 791; Golgi, 1889. p. 173.

Laverania+Hæmamæba, Laveran, 1890, p. 374; Grassi & Feletti, 1892, p. 10.

Hæmamæba, Labbé, 1894, p. 170.

Plasmodium, Labbé, 1899, pp. 80-2.

Hæmomenas, Ross, 1899, pp. 322-4. Hæmamæba, Ross, 1899, pp. 322-4. 439; 1900, pp. 522-7.

 Plasmodium, Lübs, 1853, pp. 322-4, 435, 1860, pp. 322-7.
 Plasmodium, Lühe, 1900, pp. 367-84, pp. 436-60; Neveu-Lemaire, 1900, p. 9; Minchin, 1903, pp. 239, 265, 267.
 Hæmamæba (part), Laveran, 1907, pp. 110-60.
 Plasmodium, Minchin, 1912, p. 357; Row, 1917, p. 392; Castellani & Chalmers, 1919, pp. 504-16; Muhlens, 1921, pp. 1495-502; Thomson & Woodcock, 1922, pp. 1517-30; Hegner & Taliaferro, 1924, pp. 317-40. 1924. pp. 317-40; Craig, 1926, pp. 362-470; Wenyon, 1926, pp. 908-85; Hehir, 1927, pp. 164-6; Knowles, 1928, pp. 382-410; Row, 1929, pp. 1120-5; Reichenow, 1929, pp. 998-1009; Kudo, 1931, pp. 286-7; Calkins, 1933, pp. 406-10, 566; Brumpt, 1936, pp. 424-38.

Trophozoites ring-shaped, amœboid. Schizogony in the peripheral blood. Schizonts, compact or irregular, may completely fill the infected red blood-corpuscle, which may or

may not be altered in size. Hæmozoin pigment present. Gametocytes more or less resemble the schizonts in form; in the fully grown condition are usually circular discs which more or less completely fill the infected red blood-corpuscle. Parasites of man and other mammals.

Remarks.-Coatney and Roudabush (1936) have given a list of all the known species of the genus Plasmodium together with the hosts from which each was described.

## 232. Plasmodium vlvax (Grassi & Feletti). (Fig. 123 (Pl. I.)).

Plasmodium var. tertiana, Golgi, 1889, p. 173.

Plasmodium malariæ var. tertianæ, Celli & San Felice, 1891.

Hæmamæba febris tertiana, Marchiafava & Bignami, 1891.

Hæmamæba vivax, Grassi & Feletti, 1892, p. 10.

Hæmamæba laverani var. tertiana, Labbé, 1894, p. 170, pl. ix.

Plasmodium malariæ tertianum, Labbé, 1899, p. 82, fig. 147 a.

Hæmamæba malariæ var. magna, Laveran, 1900.

Plasmodium vivax, Lühe, 1900, p. 460; Neveu-Lemaire, 1900, p. 9, pl. i, fig. 2.

 p. 5, pl. 1, hg. 2.
 Hæmamæba malariæ var. tertiznæ, Laveran, 1901.
 Plasmodium vivax, Schaudinn, 1902, pp. 169-250, pls. iv-vi;
 Minchin, 1903, pp. 243-54, 267, 332, 351; Doflein, 1909, pp. 670-3, figs. 612-15; Minchin, 1912, pp. 137, 358, 359, 360, figs. 72, 156.

+Plasmodium vivax, Row, 1917, p. 392, pls. xx-xxii.

- Plasmodium vivax, Castellani & Chalmers, 1919, pp. 510-12, figs. 166, 169; pl. i, figs. 1 b-8 b; Mühlens, 1921, pp. 1495-9; pl. xxx, figs. 1-36; pl. xxxi, figs. 1-6; pl. xxxii, figs. 1-15; pl. xxxii, figs. 1-36; pl. xxxii, figs. 1-8; pl. xxxii, figs. 1-15; pl. xxxiii, fig. 4; Thomson & Woodcock, 1922, pp. 1517-29, pl. 1xiii, figs. 1-37; pl. 1xiv, figs. 38-47, 50-7; text-figs. 540-4; Hegner & Talia-ferro, 1924, pp. 317-26, 329-30; pl. i, fig. 124; Craig, 1926, pp. 393-413, figs. 67-70; Wenyon, 1926, pp. 925-34, pl. xii, forg 1-35; fig. 400; Hebir 1927, pp. 164, 6 pl. 925-34, pl. xii, forg 1-35; fig. 400; Hebir 1927, pp. 164, 6 pl. 925-34, pl. xii, figs. 1-35; fig. 400; Hehir, 1927, pp. 164-6, pl. x, figs. 1-37; pl. xi, figs. 38-47, 50-7; text-fig. 581.
- †Plasmodium vivax, Knowles, 1927, pp. 7-11, pl. i, figs. 1-31; 1928, pp. 383-9, pl. xi.
- Plasmodium vivax, Reichenow, 1929, pp. 998-1004, fig. 975, f-l, figs. 976-86; Kudo, 1931, p. 286, figs. 120, 121, a-g; Calkins, 1933, p. 238, fig. 124, A, B; pp. 406-10, pl. i, figs. 1-6; Brumpt, 1936, pp. 432-8, figs. 190-2; Coatney & Roudabush, 1936, p. 340.

The Cycle in Man.-Sporozoites are long and slender,  $10-12\,\mu$  by  $1-2\,\mu$ , and are introduced into the blood through the bite of an infected mosquito. After entering the corpuscle the sporozoite contracts to form a disc of cytoplasm with a single nucleus, and soon develops a vacuole, thus resembling a signet-ring. The rings are comparatively large, not as a rule at the edge of a red blood-corpuscle, and usually not more than one in each corpuscle. A ring occupies about one-third the diameter of the corpuscle. In blood-films of heavy infections there may also be young forms at the edge of the red corpuscles (marginal forms). They may be seen in stained specimens, either as little streaks of blue cytoplasm with a red nucleus

(flattened marginal forms) or disc-shaped bodies with a large central vacuole and with an arched appearance (bridge forms). In living blood, in the warm stage, the trophozoite may be seen actively throwing out and withdrawing pseudopodia within the corpuscle: this special activity earned the name P. vivax for the species. After further growth the trophozoite has a more irregular shape (amœboid form) or the ring form continues, the vacuolc becomes larger, and a few yellow or light brown pigment granules are deposited in the cytoplasm. Growth is accompanied by three distinct changes in the red blood-corpuscle; it becomes larger, slightly paler, and shows on its surface a number of fine granules, which stain bright pink by Romanowsky stains. These granules are known as Schüffner's dots : they are not visible in poorly stained films. The schizont has the form of a circular plate of cytoplasm, which almost completely fills the enlarged red blood-corpuscle. Nuclear multiplication takes place, producing twelve to twenty-four nuclei. The cytoplasm gradually segments into as many portions as the nuclei, and the merozoites separate, leaving the pigment in the residual cytoplasm. The merozoites are liberated into the plasma by the bursting of the corpuscle and enter other corpuscles. The majority of parasites complete schizogony at or near the critical period of forty-eight hours, though some may do so before or after. Sometimes double infections take place, sporozoites having been introduced on two successive nights. The schizonts of one batch will attain maturity one day and those of the other twenty-four hours later. Thus there will be daily attacks of fever instead of every other day. The initial attack of fever takes place about ten to twelve days after the introduction of the sporozoites by a mosquito.

After schizogony has taken place a number of times some of the merozoites develop into gametocytes. This phase takes place mostly in the blood-vessels of the spleen or bonemarrow, as the red blood-corpuscles containing the developing gametocytes are held up in these organs. The gametocytes attain maturity in about ninety-six hours, and the effect produced on the red blood-corpuscles is the same as in the case of the schizonts. The gametocytes are circular in outline. more regularly circular than the schizont; each contains a larger number of pigment granules, more or less uniformly distributed in the cytoplasm, and a single nucleus. The male gametocyte has hyaline cytoplasm which stains a pale blue, and the nucleus is larger and contains fine chromatin granules. The female gametocyte has denser cytoplasm, which stains more deeply blue, has a smaller nucleus, with a single karyosome or a group of granules. Both contain yellowish-brown pigment granules distributed in the cytoplasm.

The Cycle in the Mosquito.—The development in the mosquito is on the same lines as in Laverania malariæ. As the gametocytes of P. vivax are the largest, the zygotes and oöeysts of this species are also probably larger than in the other species. The pigment produced in this species is of a light brown colour, while that in the other two species of human malarial parasites is dark brown or black: thus zygotes and oöcysts with light brown pigment will be those of P. vivax.

Remarks.—Grassi (1900) and Schaudinn (1902 a) described certain changes in the female gametocyte which they interpreted as parthenogenesis. J. D. Thomson (1917) and Wenyon (1926) have refuted the theory and explained that the appearances observed may be due to a red corpuscle being infected with two or more parasites, which may both be schizonts, or a schizont and a gametocyte, and thus combinations of all sorts of appearances may result.

Chalmers and Archibald (1920) described what they call the "tenue" phase of P. vivax, having the same relation to this species as P. tenue Stephens has to Laverania malariæ.

James, Nicol, and Shute (1933) gave a description of the life-cycle of *Plasmodium ovale* Stephens, with different characters to separate the species from the other malarial parasites. According to them the parasite is identical with that described by Craig in 1900 and 1914, but is distinct from *P. camarense* Emin. Giovannola (1935) believes that *P. ovale* is a modification of *P. vivax* after a long residence in the human host, and cites historical, clinical, and morphological evidence to support his claim.

Habitat.—Blood of man in almost all parts of INDIA (vide map on p. 279), and in the body of several species of Anopheles, such as A. annularis Van der Wulp, A. culicifacies Giles, A. fluviatilis James (=A. listoni Liston), A. maculatus Theo., A. minimus Theo., A. philippinensis Ludl., A. stephensi Liston, A. sundaicus Rodenwaldt, and A. varuna Iyengar.

## 233. Plasmodium malariæ (Laveran). (Fig. 124 (Pl. I).)

Malarial parasites, Laveran, 1880 a, p. 158; 1881, pp. 627-30; 1882, p. 737.

Oscillaria malariæ (part), Laveran, 1883, p. 113.

Plasmodium var. quartana, Golgi, 1885; 1886; 1890.

Amæba malariæ febris quartanæ, Celli, 1891.

Plasmodium malariæ quartanæ, Celli & San Felice, 1891.

Hæmamæba febris quartanæ, Marchiafava & Bignami, 1891.

Hæmamæba malariæ, Grassi & Feletti, 1892, p. 10.

Hæmamæba laverani var. quartanarum, Labbé, 1894, p. 170.

†Hæmamæba malariæ, Ross, 1898, pp. 133-6.

Plasmodium malarise quartanum, Labbé, 1899, p. 82, fig. 147 b.

Plasmodium malariæ, Lühe, 1900, p. 460; Neveu-Lemaire, 1900, p. 8, pl. i, fig. 1.

Plasmodium malariæ, Minchin, 1903, pp. 243, 267; 1912, p. 358. †The Quartan Parasite, Row, 1917, p. 392, pls. xviii, xix.

- The Quartan Parasite, Row, 1917, p. 392, pis. XVII, XX.
  Plasmodium malariæ, Castellani & Chalmers, 1919, pp. 512-13;
  Mühlens, 1921, pp. 1499-502, pl. xxxi, figs. 9-19; pl. xxxi, figs. 16-25; pl. xxxiii, figs. 1, 3; Hegner & Taliaferro, 1924, pp. 326-7, 330, 331, pl. ii, figs. 1-8; Craig, 1926, pp. 419-31, figs. 72, 73; Wenyon, 1926, pp. 942-4, pl. xiii, figs. 1-15; Hehir, 1927, pp. 162-4, pl. ix, figs. 1-3; pl. xi, figs. 58-75. *Plasmodium malariæ*, Knowles, 1927, pp. 12-14, pl. ii, figs. 1-22; 1028 pp. 98, 92 pl. xiii
- 1928, pp. 389-92, pl. xii.
- Plasmodium malariæ, Reichenow, 1929, pp. 1004-6, fig. 975, m-p, 987-90;
   Kudo, 1931, p. 287, fig. 121, o-u;
   Calkins, 1933, pp. 238, 406-10, fig. 124; pl. i, figs. 7-12;
   Brumpt, 1936, pp. 424-30, pl. ii, figs. 19-36;
   Coatney & Roudabush, 1936, p. 339.

The Cycle in Man.-The cycle in man is very similar to that of Laverania malariæ and Plasmodium vivax. "Rings" are of the same size as of P. vivax and have a diameter of about one-third to a half that of the red blood-corpuscle. Cytoplasm is denser and stains more deeply, and shows little amœboid activity during growth. Pigment granules are coarse and of a darker brown colour. The organism often shows a tendency to be stretched as a band across the diameter of the corpuscle. These "band forms" are more frequently seen in this species. The band may be narrow or almost as broad as long. The infected red corpuscle is no larger, and may even be smaller than the normal corpuscle, and may also be deeper in colour in stained films; and as a rule no Schüffner's or Maurer's dots are found. Schizont reaches its full size in about sixty hours and schizogony occurs every seventy-two hours. fully grown schizont is circular in outline, and almost completely fills the corpuscle, the dark pigment granules forming a central mass. Schizogony results in the production of six to twelve merozoites that are arranged in a single ring or "rosette." All phases of schizogony take place in the peripheral blood. The gametocytes are oval or spherical in outline, and completely fill the red blood-corpuscles. The distinguishing characteristics of microgametocytes and macrogametocytes are similar to those in P. vivax, the female staining more deeply blue and having a more compact nucleus. The pigment granules are scattered irregularly in the cytoplasm.

The Cycle in the Mosquito.-The development in the Anopheline mosquito is very similar to that of P. vivax, but the various stages can be distinguished from those of P. vivax by the pigment being dark brown or black. In this respect they resemble those of Laverania malariæ.

Remarks .- Two "rings" or partially developed trophozoites may sometimes be found in the same corpuscle, but are less common than in P. vivax infections. Two mature gametocytes in an enlarged cell, as also a gametocyte and a schizont, have also been found to occur in the same corpuscle.

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Double and triple infections sometimes occur owing to inoculations by mosquitoes on different days. Each group of parasites undergoes schizogony independently, thus attacks of fever occur in cases of single infection every seventy-two hours, that is, on the fourth day (hence *quartan*). Double infection may cause the attacks to appear on two successive days, and then again on two successive days after an interval of twenty-four hours. In cases of triple infection attacks of fever take place every twenty-four hours.

Mixed infection of more than one species may also occur. Very often P. vivax and Laverania malariæ are found in the same blood-film, as both these species are of commoner occurrence than P. malariæ. But the latter may also be met with in association with either of the others, and occasionally all three have been seen in the same blood-film. The diagnosis of these mixed infections will depend upon the finding of undoubted forms of each species.

Habitat.—Blood of man in almost all parts of INDIA (vide map on p. 279) and in the body of several species of Anopheles, such as A. annularis Van der Wulp, A. culicifacies Giles, A. fluviatilis James (=A. listoni Liston), A. maculatus Theo., A. minimus Theo., A. philippinensis Ludl., A. stephensi Liston, A. sundaicus Rodenwaldt, and A. varuna Iyengar.

## Distribution of the Human Malarial Parasites.

The climatic features of the different part of India being so diverse, greatest diversity of distribution of species of malarial parasites is found in the country. Knowles, Senior White, and Das-Gupta (1930) have come to the following general conclusions on the basis of the available data :---

(1) In the north-west of India P. vivax tends to predominate during the spring and early summer and L. malarix in the late autumn. P. malarix is quite unimportant.

(2) The importance of *P. malariæ* becomes greater and greater as one passes from the N.W. Frontier Province to the east and south-east. In brief we may trace two chains of increasing quartan prevalence, as follows :---

(a) Across Northern India the figures run :

N.W.F.P., 0.3 per cent.; Punjab, 1; Delhi Province, 1.2; U.P., 4.6; Bengal, 12.3; Assam, 9.9; Burma, 21.2.

(b) Down the west coast of India :

Sukkur and Sind, 15-2 per cent.; Bombay City, 6-6; Goa, 9; Kanara, 2-1; Mysore State, 15-7; Coorg, 24; Ceylon, 33 (in some places up to 71 per cent.). Other hilly areas where the incidence is high are the Central Provinces, 19 per cent., and the Madras Agency Tracts, 41.

Differential Chara	cters of the Malarial Parasi	tes occurring in Man. (Ron	nanowsky's stain.)
	Laverania malariæ (=Plasmodium falciparum).	Plasmodium vivax.	Plasmodium malariæ.
Type of fever Period of schizogony Barly trophozoite, " rings ."	Malignant tertian. 24 to 48 hours. Smallest, one-sixth of red blood-corpuscle. Accolé formas. Often two or moro in each red blood-cor- puscle. Rings thin and nair - like. Cytoplasm scanty. Chromatin deeply stained granule or bar.	Benign tertian. 48 hours. Relatively largo, one-fourth to one-third of red blood- corpuscle. Usually one in each red blood-corpuscle. Round or oval. Chromatin a round dot in thin part of ring.	Quartan. 72 hours. Smaller and denser than <i>P. vivax</i> , one-fourth to one- third of red biod-corpuscle. Usually one in each red blood-corpuscle. Round or oval. Cytroplasm deep blue. Chromatin a large deep red dot. Ring often very nar-
Growing trophozoite	Actively ameboid. Very small, up to one-fourth of red blood-corpuscie. Ctyo- plasm little. Ofromatin putch. often end	Actively ameboid. Flimay. Fine streaming pseudopo- dia through red blood-cor- puscle.	row on each side of chromatin. Very slowly ameboid. Com- pact, frequently rectangular.
Hæmozoin pigment	dividing early. Granular. Brown.	Scanty, fine grains or tiny rods. Yellowish-brown.	Much. Formed early in coarse, dense masses.
Infected red blood-corpuscies.	Normal, or even shrunken in size. Maurer's dots.	Enlarged and pale. Schüff- ner's dots.	Brown-black. Normal. No dots.
Schizont	Not seen in peripheral blood except in very serious cases. Circular, small, occupies one-half to two-thirds of red blood-corpuscle; 8 to 12 chromatin dots.	In peripheral blood. Flimsy, irregularly circular, large; 2, 4, 8, etc., rounded chromatin masses. Fine hæmozoin granules.	Iu peripheral blood. Elon. gate, quadrilateral, medium; later circular, large, com- pact; occupies most of the red blood-corpuscle. Cyto- plasm deep blue; 6 to 8
			pigmented.

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Sporulating schizont,	Small; 8 to 18 merozoites	Large, almost fills the en-	Tvnical resette: 6 to 12 mero-
" rosette."	arranged in a grape-like	larged and pale red blood-	zoites symmetrically ar-
	cluster. Central residual	corpuscie; 12 to 24 mero-	ranged round deeply pig-
	ment.	Zoites in an irregular grape- life chinter Deciding mace	mented central residual
		with hemozoin nigment	- Septit
		eccentrically situated.	
Microgametocyte	Male " crescent." Shorter	Large, circular or ovoid, fills	Smaller than in P. vivax,
	and stouter than the female.	the enlarged pale and al-	circular or ovoid, almost
	Occupies whole of red blood-	most invisible red blood-	fills the red blood-corpuscle,
	corpuscie, traces of which	corpuscle. Smaller in size	which is not enlarged.
	on concave side of crescent.	and lighter in colour than	Smaller in size and lighter
	Cytoplasm clear pale blue.	the femalc, and with a larger	in colour than the female,
	Nucleus large, oval, central,	chromatin mass near centre.	and with the chromatin
	diffuse. Hæmozoin as	Pigment light and yellow-	mass near the centre. Cen-
	scattered fine grains in	brown.	tral cluster of pigment
1	middle two-thirds.		masses.
Macrogametocyte	Female " crescent." Longer	Large, circular or ovoid, fills	Smaller than in $P$ . vitax,
	than male, and with more	the enlarged pale and al-	circular or ovoid, almost
	pointed ends. Cytoplasm	most invisible red blood-	fills the red blood-corpuscie,
	deeply blue. Nucleus com-	corpuscle. Larger in size	which is not enlarged.
	pact, stains intensely.	and darker in colour than	Larger in size and darker
	Hæmozoin as a central	the male, and with a smaller	in colour than the male,
	dense cluster.	chromatin mass at one	and with a chromatin mass
		side. Considerable granular	near one end. Abundant
		hæmozoin.	deep brown-black pigment.
General character of peri-	Kings and crescents only	All stages of schizogony,	All stages of schizogony, as
pheral blood-films.	usually seen. Multiple in-	as well as gametocytes.	well as game to ytes. Multi-
	fection of red blood-cor-	Multiple infection of red	ple infection of red blood-
	puscles common. Infection	blood-corpuscles not un-	corpuscles very rare.
	much more intense than in	common.	
	outler two species.		
A fourth species of malarial Paratick vol. xvi. p. 383. 1922	parasite in Man has been de ), but so far has not heen recor	sscribed, <i>Plasmodium ovale</i> St ded from IndiaEntrop	ephens (Ann. Trop. Med. and
and a start of the second formation of the	toos a manage and and and and an and all	WATER TOTAL WEINTER TIO IT NON	

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(3) Taking all-India generally, P. vivax is the predominant species, but this only applies to the spring and early summer months. In autumn and early winter L. malariæ usually becomes predominant.

(4) Benign tertian (P. vivax) and quartan malaria (P. malarix)are almost equally common in Ceylon, although in certain parts of the island—and especially in the hilly districts—quartan malaria predominates. The figure for malignant tertian (L. malarix) is extremely low throughout the island.

Taking the figures for India as a whole, it is clear that benign tertian and malignant tertian infections share the main incidence of malaria in India about equally between them, while quartan malaria is responsible for less than 7 per cent. The relative distribution of the different species in various localities will be seen from the map reproduced from Knowles, Senior White, and Das-Gupta on p. 279.

# Distribution of Mosquito-carriers of Malaria.

Covell (1931) has summarized the available information concerning the mosquito carriers of malaria in India, Burma, and Ceylon as follows :---

"The chief malaria carriers in Northern and Peninsular India are A. culicifacies, A. stephensi, and A. listonii. A. culicifacies is the most important vector in rural areas, whilst A. stephensi is notorious as being the great urban malaria carrier of India, it being the only malaria-carrying species capable of adapting itself to the conditions obtaining in cities.

"A. maculatus and its variety willmori are considered to be important carriers in submontane areas, chiefly on epidemiological grounds. A. maculatus has been found naturally infected (gut only) by Feegrade (1927 a) in Burma.

"In Eastern India A. minimus is probably the most important carrier. Ramsay (1930) has shown this species to be the principal vector in the Cachar District of Assam. Iyengar (1927) considers A. minimus var. varuna to be the chief carrier in Lower Bengal. Sur and Sur (1929), as the result of their dissections under natural conditions, conclude that A. philippinensis plays an important part in malaria transmission in Bengal. This species has also been found naturally infected in Burma (Feegrade, 1926). A. fuliginosus has also been found infected in nature in Bengal and in Madras, but this species is not generally considered to play an important part in transmission.

"A. ludlowii is an important carrier in the Andamans and on the coasts of Burma and Bengal. In the Andamans it is considered to be the only vector of any practical importance.



"Various other species have been suspected to be carriers of malaria in India, but there is no direct evidence that they play any important part in the transmission of the disease.

"In Ceylon Carter (1927) considers that A. culicifacies and A. listonii are the chief carriers, whilst the position of A. maculatus is uncertain. The last-named species, although prevalent in certain districts where malaria is severely endemic, is relatively more abundant at somewhat higher elevations (1500 feet and over), where the incidence of the disease is low, but where the factors of temperature and atmospheric humidity are favourable for transmission. Senior White (1920), however, is of opinion that this species is 'the malaria carrier par excellence of the Ceylon hill country,' on epidemiological grounds."

Christophers (1933) has summarized the available data as follows :---

"1. Important carriers wherever found :

A. culicifacies ; A. fluviatilis ; A. stephensi ; A. sundaicus ; A. minimus.

- "2. Less important, but proved carriers in some areas :— A. varuna ; A. philippinensis.
- "3. Species that are important carriers in other countries, but of too limited distribution to be important in India:

A. superpictus; A. multicolor.

"4. Species that have been found infected in nature or experimentally within or without the area, but which are probably not of importance as carriers:

a. Found infected in nature in Indian area :

A. maculatus; A. maculatus var. willmori; A. fuliginosus; A. pulcherrimus; A. maculipalpis; A. pallidus; A. ramsayi; A. vagus.

b. Infected experimentally only in Indian area :

A. theobaldi; A. subpictus; A. turkhudi.

c. Found infected in nature outside Indian area only :

A. hyrcanus; A. barbirostris; A. karwari;

A. leucosphyrus; A. sergenti; A. umbrosus;

A. tassellatus; A. aconitus; A. kochi.

"The extremely common species A. subpictus and A. vagus appear to have little or no relation to the incidence of malaria."

Iyengar (1934) found five species of Anopheles, viz., A. jeyporiensis var. candidiensis Koidz, A. varuna Iyengar, A. fluviatilis James, A. listoni (Liston), and A. culicifacies Giles, infected in nature in Travancore.

### 234. Plasmodium cynomolgi Mayer. (Fig. 125 (Pl. II).)

Plasmodium cynomolgi, Mayer, 1908, pp. 314-22, pl. xxi; Flu, 1908, pp. 323-30, pl. xxii; Blanchard & Langeron, 1913, pp. 529-42, pls. viii-ix; pp. 599-607, pl. x; Castellani & Chalmers, 1919, p. 515.

+Plasmodium cynomolgi (?), Donovan, 1920, p. 721.

Plasmodium cynomolgi, Mühlens, 1921, p. 1612-14, fig. 32; Wenyon, 1926, p. 969.

Plasmodium inui, Wenyon, 1926, p. 971; Knowles, 1928, p. 439; Reichenow, 1929, p. 1007, fig. 993.

Plasmodium cynomolgi, Reichenow, 1929, pp. 1007-8, fig. 994.

Plasmodium inui (?), Green, 1931, pp. 649-50; 1932, pp. 455-77, 7 figs.

†Plasmodium kochi (?) (part), Napier & Campbell, 1932, pp. 246-9. †Plasmodium sp. (part), Knowles & Das-Gupta, 1932, pp. 300-20,

7 pls. & 6 charts : Sinton & Mulligan, 1932, p. 324.

<sup>+</sup>Plasmodium inui var. cynomolgi, Sinton & Mulligan, 1932 a, pp. 396-405.

\*Plasmodium cynomolgi, Sinton, 1934 a, pp. 48-50; 1934 b, pp. 392, 399-400; Mulligan, 1935, pp. 288-300, pl. iv, figs. 1-35. Plasmodium cynomolgi, Coatney & Roudabush, 1936, p. 339.

Young rings,  $2-5\mu$  in diameter, round or oval, with prominent chromatin dot and well-developed vacuole. Two or three rings sometimes seen in same red corpuscle; infested corpuscle usually unaltered; no stippling. Growing forms often show marked amœboid movement, resembling that seen in P. vivax. Pigment scanty, appears later, and is darker and coarser than in P. inui: vacuolation, especially in large forms, not marked; infected red corpuscles appreciably enlarged and pale; stippling resembling Schüffner's dots, conspicuous. Schizonts common in peripheral blood; mature schizonts with eight to sixteen merozoites irregularly scattered; pigment in small dense clump; infected red corpuscles with marked pallor and prominent stippling. A minute accessory chromatin dot frequently seen in merozoites and young rings. Schizogony cycle takes forty-eight hours to complete. Gametocytes intra-corpuscular or free, larger than the normal red corpuscle. Female gametocytes round or oval; cytoplasm deep blue, chromatin small, compact, and eccentric; pigment not very abundant, and granules darker and coarser than in P. inui. Male gametocytes smaller, cytoplasm staining less deeply, chromatin central or peripheral, larger and less deeply staining than in the female gametocytes. Sporogony cycle in mosquitoes.

Remarks.—P. inui was described by Halberstädter and Prowazek (1907) from monkeys (Macaca) from Borneo. Mayer (1908) described P. cynomolgi, which was generally considered as identical with P. inui. He (1908) could obtain no development in Culex pipiens nor in Aëdes (Stegomyia) ægypti (Linn.) [= Aëdes argenteus]; he observed small oöcysts in Anopheles maculipennis, but could not follow the complete development. Léger and Bouilliez (1913) inoculated the parasite into a number of monkeys, including *Cercopithecus* sp. (the common host of *P. kochi*), with the result that many of them died of heavy infections. Quinine, even in large doses, appeared to have no influence. Donovan (1920) examined a large number of specimens of *Silenus sinicus* and *Pygerythrus priamus* (=*Presbytis priamus*) in the Nilgiri Hills, South India, but with negative results. Subsequently he found a *Plasmodium* in a blood-film from a specimen of *S. sinicus* from the same region, which he considered as morphologically identical with that found in *S. irus* (=*M. cynomolgus*). He also mentions having seen *Plasmodium* in several other apes and monkeys, but does not give any description or cite localities.

Franchini (1927) found in *Macacus cynomolgus* a parasite which showed many resemblances with *P. malariæ*. Macfie (1928) found *P. inui* in a young baboon in Africa. Reichenow (1929) observed that *P. inui* does not show enlargement of the corpuscles and Schüffner's dots, whilst *P. cynomolgi* shows these, but still came to the conclusion that these forms are probably identical.

Sinton and Mulligan (1932), in a specimen of Silenus irus, obtained an infection which later work showed was a mixed one. They succeeded in isolating in S. rhesus, by blood inoculation from the same naturally infected host, two morphologically distinct types of *Plasmodium*, which they regard as P. inui var. cynomolgi and P. knowlesi. They obtained the sporogony cycle in a number of mosquitoes, and succeeded in transmitting the infection to a healthy specimen of Silenus rhesus by the bites of infected A. annularis Van der Wulp. Napier and Campbell (1932) and Knowles and Das-Gupta (1932) also recorded a malarial infection in S. irus, said to have been imported from Singapore. Sinton and Mulligan (1932 a) gave a critical review of the literature relating to malarial parasites in lower monkeys, and came to the conclusion that the majority of species of Plasmodium described from the lower monkeys fall into two main divisions, those found in the African monkeys (P. kochi group) and those found in the Oriental ones (P. inui group). According to them the latter include P. inui Halb. & Prowazek (sensu restr.), P. inui var. cynomolgi Mayer, P. inui var. gonderi, and P. knowlesi, and the various previous records were arranged according to this scheme. Later on, Sinton (1934 a, 1934 b) and Mulligan (1935) have come to the definite conclusion that P. cynomolgi is a distinct species and not a variety of P. inui, as the duration of schizogony in the former is forty-eight hours and in the latter seventy-two hours.

Habitat.—Blood of Silenus sinicus (Linn.) Nilgiris; blood of Silenus irus (Cuv.), said to have been imported from Singapore, and in inoculation infections in S. irus (Cuv.), S. rhesus (Audeb.), and S. sinicus (Linn.); sporozoites in the salivary glands of Anopheles annularis Van der Wulp (=A. fuliginosus Giles), A. splendidus Koidzumi (=A. maculipalpis James & Liston), A. maculatus Theobald, and A. culicifacies Giles: BENGAL, Calcutta; PUNJAB, Kasanli.

## 235. Plasmodium inul Halberstädter & Prowazek. (Fig. 126 (Pl. II).)

Plasmodium inui, Halberstädter & Prowazek, 1907, pp. 37-43, pl. vi; Castellani & Chalmers, 1919, p. 515; Mühlens, 1921, p. 1612; Wenyon, 1926, pp. 971-2, pl. xv, figs. 8-14; Knowles, 1928, p. 439; Reichenow, 1929, pp. 1007-8, fig. 993.

Plasmodium inui, sens. restr., Sinton & Mulligan, 1933, pp. 389-95.

†Plasmodium inui, Sinton, 1934 b, pp. 393–410, pls. iii, iv; Mulligan, 1935, p. 310.

Plasmodium inui, Coatney & Roudabush, 1936, p. 339.

Young rings about one-fifth to one-fourth diameter of infected red cell; marked vacuolated appearance, amœboid movement of lobose nature. Older trophozoites more rounded. mature forms do not fill infected red cell. Mature schizonts with sixteen merozoites which often form a rosette. Chromatin relatively large, prominent, and usually excentric at all stages; may be divided into two equal or unequal masses in young forms. Pigment yellow to brown, becoming darker with age; appears early; fine and abundant with peripheral distribution. Infected red cells slightly enlarged with older forms; stippling less conspicuous, scantier than with P. cynomolgi. Duration of schizogony cycle seventy-two hours. Gametocytes rounded, about the size of normal red cells, with yellowish-brown to brown pigment, scattered and abundant. Female gametocyte with cytoplasm stained deep blue with Romanowsky's stain, with a small chromatin mass, and with scattered and coarser pigment; when mature, larger than normal red corpuscle. Male gametocyte with large loose chromatin mass and abundant scattered pigment, lighter brown than in the female.

*Remarks.*—Sinton (1934 b) has come to the conclusion that the duration of schizogony is seventy-two hours, and not fortyeight hours as reported by Mulligan (1935), to which paper he had access prior to publication.

Habitat.—Blood of Silenus irus (Cuv.) from Malaya, and in inoculation infections in S. irus (Cuv.) and S. rhesus (Audeb.): PUNJAB, Kasauli.

## 236. Plasmodium knowlesl Sinton & Mulligan. (Fig. 127 (Pl. II).)

Plasmodium sp., Franchini, 1927, pp. 467-71, 2 figs.

Plasmodium kochi (?) (part), Napier & Campbell, 1932, pp. 246-9.
Plasmodium sp. (part), Knowles & Das-Gupta, 1932, pp. 301-20,
7 pls. & 6 charts; Sinton & Mulligan, 1932 a, p. 324.

+Plasmodium knowlesi, Sinton & Mulligan, 1932 b, pp. 379-80, 409-21, pl. v, figs. 1-35.

Plusmodium knowlesi, Coatney & Roudabush, 1936, p. 339.

Youngest rings closely resemble those of Laverania malariæ and measure one-fourth to one-half of the infected corpuscle. Cytoplasm shows slight but definite thickening on the side opposite the chromatin, and a well-developed vacuole. Chromatin prominent, and occurs as a single round, oval or elongate mass or in two (rarely three) smaller masses. One or, rarely, two minute accessory chromatin dots may be present. Older trophozoites rounded or only very slightly amæboid, vacuole very inconspicuous or absent. Mature schizonts of the same size, or smaller than the normal red corpuscles. Number of merozoites commonly about ten. Schizogony cycle takes twenty-four hours. Pigment appears early, is relatively abundant, and granules are fairly coarse, varying from greenish-brown to almost black. Infected red corpuscles not enlarged, but often show characteristic distortion, and stippling usually absent with ordinary stains. Gametocytes spherical, like those of P. malariæ, about the size of normal red corpuscle, and with abundant coarse and dark pigment. Cytoplasm of the female gametocyte stains deep blue with Romanowsky's stain, and its chromatin is dense and compact, is usually situated peripherally, and frequently shows a more deeply staining inner area; the pigment is scattered irregularly. The male gametocyte stains poorly, and its chromatin is large and diffuse, appearing to merge into the cytoplasm. Dark brown or almost black granules of pigment occur in both the female and the male gametocytes. Sporogony not known to take place in mosquitoes.

Remarks.—Napier and Campbell (1932) found a Plasmodium in the blood of a specimen of Silenus irus in Calcutta, said to have been imported from Singapore. Knowles and Das-Gupta (1932) described it in greater detail after inoculating S. rhesus with the strain, in which host it multiplied enormously. Sinton and Mulligan (1932) came to the conclusion that the original infection in S. irus was a mixed one of Plasmodium inui var. cynomolgi and of P. knowlesi, and by inoculating S. rhesus an almost pure infection with P. knowlesi was the result. A large number of specimens of Silenus rhesus from Northern India, examined by Knowles and Das-Gupta (1932) as well as by Sinton and Mulligan, have not shown any malarial infection, which suggests that this species of monkey rarely, if ever, suffers from natural infection with malaria in this region.

Knowles and Das-Gupta (1932) were also successful in infecting a man with a *Plasmodium* from the lower monkeys.

Habitat.—In natural infections of blood of Silenus irus (Cuv.) believed to be imported from Singapore, and experimental infections of S. rhesus (Audeb.): BENGAL, Calcutta.

## 237. Plasmodium kochl Laveran. (Fig. 135.)

Hæmamæba kochi, Laveran, 1899, p. 124.

Plasmodium kochi, Léger & Bouilliez, 1914, pp. 954-85.

\*Plasmodium kochi, Castellani & Chalmers, 1913, pp. 387-8; 1919, p. 515.

Plasmodium kochi, Mühlens, 1919, pp. 1608-11, fig. 31.

†Plasmodium kochi, Donovan, 1920, p. 719.

Plasmodium kochi, Wenyon, 1926, p. 971, pl. xv. figs. 22-8; Knowles, 1928, p. 439, fig. 102, 11-12; Reichenow, 1929, p. 1006, fig. 991.

†Plasmodium kochi, Sinton & Mulligan, 1933. p. 424.

Plasmodium kochi, Coatney & Roudabush, 1936, p. 339.

Resembles P. vivax. Ring-forms large. The young rings occupy a smaller proportion of the corpuscle than in P. vivax, but as growth proceeds resemblance becomes marked. The infected red blood-corpuscles become enlarged, parasite becomes irregular in shape, pigment granules are of a light brown colour, and sometimes Schüffner's dots may be found



Fig. 135.—*Plaemodium kochi* (Laveran). A, ring-form; B, schizont. (From Reichenow, after Gonda and Berenberg-Gossler.)

to occur. Schizogony is completed in forty-eight hours. Schizont produces eight to fourteen merozoites, and bears a striking resemblance to that of *P. vivax*. Gametocytes are large round bodies which can be distinguished as male and female, as in *P. vivax*.

*Remarks.*—The species is a common parasite of monkeys in tropical Africa, but shows little sign of pathogenicity in natural infections or in inoculated animals. Gonder and Rodenwaldt (1910) noted, however, that if splenectomy is previously performed infections are much more severe, temperature rises, and the parasites continue in the blood for many months. They were unable to inoculate the parasite

into man. Further development could not be achieved in Anopheles maculipennis Meig. or in Aëdes (Stegomyia) ægypti (Linn.) [= Stegomyia fasciata]. Castellani and Chalmers (1913) stated that illness and death may occur among the monkeys in Ceylon, due to a malarial parasite which they refer to P. kochi. No description of the parasite is given, and according to Sinton and Mulligan (1932) this report needs confirmation.

Habitat.-Blood of monkeys : CEYLON ; Cercopithecus sp. ? : Nilgiris.

#### 238. Plasmodium pitheel Halberstädter & Prowazek. (Fig. 136.)

†Plasmodium sp., Laveran, 1905 (as noted in Wenyon, 1926, p. 1363). Plasmodium pitheci, Halberstäder & Prowazek, 1907, pp. 37-43, pl. vi; Shibayama, 1910, pp. 189-91, 1 pl.

†Plasmodium pitheci, Donovan, 1920, p. 719.

Plasmodium pitheci, Reichenow, 1920 c, pp. 207-16, 1 pl.; Mühlens, 1921, pp. 1611-12.
 †Plasmodium pitheci, Donovan (first recorded in Wenyon, 1926,

- p. 1363).
- Plasmodium pitheci, Wenyon, 1926, p. 972, pl. xv, figs. 29-35; Knowles, 1928, fig. 102, 13, 14; Reichenow, 1929, pp. 1006-7, fig. 992; Coatney & Roudabush, 1936, p. 340.

Young rings resemble those of Laverania malariæ. Fully developed trophozoite shows dark brown or black pigment. Schüffner's dots present. Schizogony resembles that of P. vivax. Gametocytes resemble those of P. malariæ.



Fig. 136.—Plasmodium pitheci Halb. & Prow. A, ring-form, double infection; B, older ring; C, schizont; D, macrogamete; E, microgametocyte. (From Reichenow, after Halberstädter and Prowazek.)

*Remarks*.—The parasite resembles *P. inui*, but is distinguished by its dark brown or black pigment. Shibayama (1910) states that Schüffner's dots are not present, but Wenyon thinks that his staining was not sufficiently intense to show them. Dodd (1913) recorded an infection of an orang-utang with this parasite, which proved fatal. Reichenow considers it possible that what has been described as P. pitheci may have been one of the human parasites.

Habitat.-Blood of orang-utang, Simia satyrus Desmarest : Asia (? exact locality-not given by Laveran); an Indian menagerie (Donovan).

### 239. Plasmodium semnopitheci Knowles. (Fig. 137.)

†Plasmodium semnopitheci, Knowles, 1919, pp. 195-202, pls. xvixix.

- Plasmodium semnopitheci, Mühlens, 1921, pp. 1614-15.
- +Plasmodium sp., Chimisso, 1922, pp. 38-54. Plasmodium semnopitheci, Wenyon, 1926, pp. 972, 1363.
- +Plasmodium semnopithecus, Scott, 1926, p. 237; Knowles, 1928. p. 439.
  - Plasmodium semnopithecus, Reichenow, 1929, p. 1008; Sinton & Mulligan, 1933, pp. 382-6, 423-4; Coatney & Roudabush, 1936, p. 340,

Young trophozoite an almost non-pigmented ring. Growing trophozoites large flimsy rings with very large vacuole, very abundant pigment, little amœboid movement, and single, oval or rod-shaped chromatin mass. Absence of segmenting forms in the peripheral blood. Decolorization of infected red corpuscles frequently observed, but stippling never seen. Gametocytes rounded. Female gametocytes often free, and larger than normal red corpuscles; chromatin small, deeply staining, oval or much elongated; pigment very dark brown,



Fig. 137.-Plasmodium semnopitheci Knowles. A, ring-form; B, larger and more flimsy ring; C, extracellular form impinging against a corpuscle; D, gametocyte. (After Knowles.)

very abundant, and in very fine granules or small dark collections. Male gametocytes smaller than female gametocytes, and with very large pink nuclei.

Remarks.--Knowles (1919) discovered this parasite during an experiment conducted for the purpose of transmitting the malignant tertian parasite of man to a monkey. The shock of the operation and the injection of foreign blood were believed to have stimulated a latent infection of a species peculiar to the host, which suddenly became virulent and proved fatal. Nearly every corpuscle was found infected, and there were inumerable free forms, some of them undergoing schizogony whilst still extracellular. Wenyon (1926) thinks this due to intensity of infection and to examination after death: he considered the parasite to be very similar to P. inui, and Reichenow (1929) thought that it was probably identical with Sinton and Mulligan (1932 a) consider it noteworthy it. that, except for Donovan's undescribed parasite, the only

Differe	ntial Characters of ti	he Malarial Parasites occ	urring in Lower Oriental	Monkeys.
	P. semnopitheci.	P. inui.	P. cynomolgi.	P. knowlesi.
Period of schizogony. Chromatin in young ring-forms.	Single, oval or rod- shaned mass	72 hours. Frequently double, and of	48 hours. "Accessory dot" present.	24 hours. "Accessory dot " present.
Trophozoites	Amceboidicity slight, with large vacuole.	Amœboidicity of lobose nature : vacuolation marked up to early segmentation.	Amœboidicity marked, of "vivaz" character; vacuole at first well developed, but not	Amœboidicity slight or absent; vacuole small in older forms.
Pigment in tropho- zoitea.	Very abundant.	Yellow to brown, be- coming darker with age; appears early; fine and abundant with peri-	marked in old forms. Golden-brown; appears later, and is coarser and scantier than in <i>P. inui</i> ; distribution less mark-	Golden-brown to almost black; appears early; abundant.
Mature schizonts	I	pheral distribution. Maximum 16 merozoites.	edly peripheral. Maximum 16 merozoites.	Maximum 11 merozoites.
Gametocytes	Larger than normal red cell; pigment dark hrown very	Often rosette. About size of normal red cell; pigment scattered, vellowith house, to	More regular. Distinctly larger than red	Grape-like cluster. About size of normal red cell; pigment relatively
Infected red cells	abundant. Decolorized. Stip- pling never ob- served.	brown and abundant. brown and abundant. Slightly enlarged with older forms. Stippling less conspicuous, scan-	in P. inut. Much enlarged with old forms. Stippling very conspiencus and dots	coarse, prown to place, and abundant. Not enlarged; showing characteristic distortion. Stippling only shown by
Pathogenicity	Virulent.	tier than with P. cyno- molyi. Fow or no symptoms. Easily inoculable to	very numerous. Usually no severe symp- toms. Fasily inoculable	special stains. Mild in <i>S. irus,</i> but causing vary severe
		outer species of <i>Dutenus.</i> Not inoculable into higher monkeys.	to other specuraens of Silenus.	symptoms, otten harmo- globinuria, when inocu- lated into <i>S. rhaeua.</i> Has been transmitted to man and the gibbon.

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natural malarial infections which have been reported from Indian monkeys, namely, by Knowles (1919), by Chimisso (1922), and by Scott (1926), all appear to be due to P. semno-pitheci.

Habitat.—Blood of the entellus monkey or hanuman, Pygathrix entellus (Dufr.) (=Semnopithecus entellus) : ASSAM; blood of Presbytes pileatus Blyth : ASSAM, in the Zoological Gardens, London. Also blood of Silenus rhesus (Audeb.), believed to be from INDIA, examined in Italy.

## 240. Plasmodlum bubalis Sheather. (Fig. 138.)

*Plasmodium bubalis*, Sheather, 1919 a, pp. 1-5, pls. i, ii ; 1919 b, p. 223.

Plasmodium bubalis, Mühlens, 1921, p. 1616.

†Plasmodium bubalis, Edwards, 1925, p. 50.

Plasmodium bubalis, Wenyon, 1926, pp. 975-6, pl. xvi, figs. 13-17;
 Knowles, 1928, p. 442, fig. 102, 15, 16; Reichenew, 1929, p. 1009;
 Coatney & Roudabush, 1936, p. 338.
 †Plasmodium bubalis, Taylor, 1938, p. 42.

Resembles *P. malariæ* in many respects. Smallest parasites round, oval or pyriform; pigment has appeared in parasites  $3\mu$  in size. Larger parasites,  $6\mu$  in size, generally round,



Fig. 138.—*Plasmodium bubalis* Sheather. A, young trophozoite; B, double infection with two trophozoites; C, large trophozoite; D, schizont; E, merozoites escaping from the corpuscle. (After Sheather.)

sometimes oval, and possess a central vacuole. Chromatin is peripheral, single or double, and the pigment in the cytoplasm is scattered or in clumps. The larger parasites have a regular outline and cause a slight enlargement of the infected red blood-corpuscle. Adult schizont completely fills the corpuscle, produces seven to fourteen merozoites, and the pigment is aggregated in a single clump. Some of the large forms with a single nucleus are probably gametocytes, and many of them possess a vacuole.

Remarks.—Sheather (1919) found the parasite in a buffalo, which died after inoculations made for immunization purposes, and noted that there was a fairly heavy infection, 1.6 per cent. of the red corpuscles containing the parasite. Edwards (1925) noted the parasite again.

Habitat.—Blood of buffalo, Bos bubalus Linn., and of ox, Bos indicus Linn. : UNITED PROVINCES, Muktesar, Izatnagar. SPOR.

## 241. Plasmodium canis Castellani & Chalmers. (Fig. 139.)

†Plasmodium canis, Castellani & Chalmers, 1910; 1919, p. 515, figs. 171, 172.

Plaemodium canis, Mühlens, 1921, p. 1617; Wenyon, 1926, p. 976; Knowles, 1928, p. 442; Reichenow, 1929, p. 1009; Costney & Roudsbush, 1936, p. 338.

Morphologically similar to *P. vivax*. Small round merozoite enters the red cell, and grows into a pigmented trophozoite, finally dividing into a number of merozoites. Schüffner's dots are seen. Female gametocyte has a small rounded nucleus. Male gametocyte has a narrow elongated nucleus. Infected red cells are enlarged.



Fig. 139.—Plasmodium canis Castellani & Chalmers. A, young trophozoite; B, full-grown trophozoite; C, early schizont; D, mature schizont; E, female gametocyte; F, male gametocyte. (After Castellani and Chalmers.)

*Remarks.*—Castellani and Chalmers (1910) described this parasite from dogs in Colombo. Castellani (1924) again refers to it, and states that he saw several cases of infection in dogs in Colombo. No other observer has seen the organism. Wenyon (1926) obtained blood-films from 500 pariah dogs from Ceylon, but in none of them could the parasite be found.

Habitat.—Blood of the dog, Canis familiaris Linn. : CEYLON, Colombo.

## 242. Plasmodium equi Castellani & Chalmers.

†Plasmodium equi, Castellani & Chalmers, 1913; 1919, p. 516. Plasmodium equi, Wenyon, 1926, p. 976; Reichenow, 1929, p. 1009; Coatney & Roudabush, 1936, p. 339.

Very similar to P. vivax of man.

*Remarks.*—Castellani and Chalmers gave no description, but simply stated that the species closely resembled *P. canis.* It has not been recorded since by any other observer.

Habitat.-Blood of the horse, Equus caballus Linn. : CEYLON.

### 243. Plasmodium mackiel de Mello & de Sá. (Fig. 140.)

†Plasmodium mackiei, de Mello & de Sá, 1916, pp. 736-7, pl. lxiv, fig. C, 1-10.

Plasmodium mackiei, Wenyon, 1926. p. 1364; Coatney & Roudabush, 1936, p. 339.

Young trophozoite of a bacillary form. The schizonts show a precocious division of the chromatin, and always form six merozoites in the rosette. Gametocytes spherical or ovoid.



Fig. 140.—*Plasmodium mackiei* de Mello & de Sá. A, young trophozoite; B, schizont; C, gametocyte. (After de Mello and de Sá.)

Habitat.—Blood of Myotis muricola (Hodgs.) (==Vespertilio muricola) : PORTUGUESE INDIA, Arjuna (Bardéz), Santa Cruz.

### 244. Plasmodium narayani de Mello & Dias. (Fig. 141.)

†Plasmodium narayani, de Mello & Dias, 1936, pp. 212-13, pl. iii.

Young trophozoite has an annular form, with its cytoplasm more or less thin or more or less compact. Sometimes two "rings" occur in the same red blood-corpuscle. Parasite



Fig. 141.—Plasmodium narayani de Mello & Dias. A, flimsy "ring";
B, compact "ring"; C, two "rings" in the same corpuscle;
D, full grown trophozoite; E, "rosette" showing schizogony; F, female gametocyte. (After de Mello and Dias.)

not seen in the living condition, but inferred to be strongly amœboid, as many aberrant forms, such as fusiform, crescentic or triangular, were met with. Young schizont occupies one-sixth to one-fifth of the red cell, and sometimes shows one or two granules of almost black pigment. Full-grown schizont round, with many granules of pigment, some of them rodshaped, filling the cytoplasm. Chromatin lodged in a clear, vacuolated zone, in contrast with the cytoplasm, which is more compact. Very rarely "rosettes," with merozoites, found in the lung-smears. Female gametocyte oval, with a compact oval nucleus situated in the middle and some granules and rods of pigment irregularly scattered in the surrounding area. Male gametocytes not found. Infected red cell sometimes dehæmoglobinized, somewhat enlarged; but does not show dots of any kind.

Remarks.-The parasites were very scanty and were found in the blood and lung-smears only.

Habitat.-Blood and smears from the lung of the fish-otter, Lutra lutra Linn. : PORTUGUESE INDIA, Nagoa.

#### 245. Plasmodium pteropi Breinl. (Fig. 142.)

Plasmodium pteropi, Breinl, 1912; Johnston, 1913, p. 35, figs.; A. & M. Léger, 1914, pp. 399-401.

A. K. M. Legel, 1914, pp. 359-401.
 †Plasmodium pteropi, Mackie, 1914, pp. 375-6, pl. xlvii; Wenyon, 1926, p. 974, pl. xvi, figs. 8-12, p. 1362.
 Plasmodium pteropi, Knowles, 1928, p. 442; Reichenow, 1929, p. 1009; Coatney & Roudabush, 1936, p. 340.

Resembles P. vivax of man. Trophozoites ring-shaped, Pigment light showing a circular form when full grown.



Fig. 142.-Plasmodium pteropi Breinl. A, ring-form; B, trophozoite; C, gametocyte. (After Mackie.)

brown. Immature schizonts and gametocytes also observed. Infected red blood-corpuscles may be slightly enlarged. Sporogony not known.

Remarks.—Breinl (1912) described P. pteropi from the blood of a flying fox, Pteropus gouldi Peters, in West Australia. Johnston (1913) and A. & M. Léger (1914) described P. pteropi as a new species. Mackie (1914) also described a very similar form from Pteropus medius Temm. (=Pteropus edwardsii Horsfield), and gave it the name P. pteropi without knowing that Breinl had already given the name to the West Australian form. According to Wenyon the two are probably identical. The parasite was also seen in the blood of the flying fox in Cevion, and Wenyon (1926) has drawn the figures from these films.

Habitat.-Blood of the flying fox, Pteropus medius Temm. = P. edwardsii Horsfield): INDIA; CEYLON.

## 246. Plasmodium ratufæ Donovan.

+Plasmodium ratufæ, Donovan, 1920, pp. 719-20.

Plasmodium ratufæ, Wenyon, 1926, p. 974; Knowles, 1928, p. 442; Reichenow, 1929, p. 1009.

Very similar to P. vivax of man. Gametocytes predominated, and in a few films the female gametocytes exceeded in number the male forms.

Remarks.—The species is similar to, if not identical with, P. vassali (Laveran, 1905), the parasite first found by Vassal in Sciurus grisemanus in Annam.

Habitat.—Blood of the giant squirrel, Ratufa indica Erxleben: BOMBAY, Malabar.

## 247. Plasmodium tyrio de Mello, Fernandes, Correa, & Lobo. (Fig. 143.)

Plasmodium tyrio, de Mello, Fernandes, Correa, & Lobo, 1928, pp. 513-16, figs. 1-28.

Plasmodium tyrio, Coatney & Roudabush, 1936, p. 340.

Young trophozoitcs usually circular or oval, very regular; aberrant or fusiform forms very rare. Ring-forms show a



Fig. 143.—Plasmodium tyrio de Mello, Fernandes, Correa, & Lobo. A, B, ring.forms; C, D, rosette forms; E, F, male gametocytes; G, H, female gametes. (After de Mello, Fernandes, Correa, and Lobo.)

precocious division of the nucleus into two or three granular masses united together. Schizonts irregular in form, resemble those of P. vivax, and even when fully developed do not cover the entire area of the corpuscle. Large black pigment granules appear irregularly in the cytoplasm of the parasite. Schizogony takes place in the peripheral blood. The number of merozoites varies from four to eight. Gametocytes spherical or ovoid. Male gametocytes almost devoid of pigment; when the pigment is present, it is in the form of fine dark granules dispersed round the nucleus. Female gametocytes have a more deeply staining cytoplasm, smaller nucleus, and large black granules of pigment.

Dimensions.—Schizonts  $1.5-4.5\mu$  by  $1-2\mu$ ; gametocytes, male  $3-4.5\mu$  by  $2.2-5\mu$ , female  $3-4\mu$  by  $2-2.5\mu$ .

Habitat.—Peripheral blood, as also smears from lungs and liver of the ant-eater, *Manes pentadactyla* Linn. (popularly known in Goa as the "Tyrio"): PORTUGUESE INDIA, Nova Goa.

# 3. Family THEILERIIDÆ du Toit, 1918.

Parasites of blood of Mammals, which do not form pigment (hæmozoin). Schizogony takes place in the endothelial cells of the capillaries of the internal organs, and the schizonts (Koch's "blue bodies") produce a number of merozoites. The parasites finally invade the red corpuscles, within which they occur as round, ovoid, rod-like or irregular forms. Show no tendency towards a paired arrangement. The forms in the red corpuscles do not reproduce, and are possibly gametocytes.

The family includes the genus *Theileria*, the best-known species of which, *T. parva*, causes the East Coast fever of cattle in Africa and elsewhere. Species of *Theileria* have also been recorded from sheep and goats in other countries. Du Toit (1918) recognizes *Rangelia* Carini & Maciel (1914) as a distinct genus, but Wenyon (1926) has advanced convincing reasons for merging the two genera into one.

Remarks.—There is no teneral agreement as regards the position of this family and the following one in the system of classification. Wenyon (1926) places the two families Theileriidæ and Babesiidæ in a suborder PIROPLASMIDEA. The suborder includes parasites which inhabit red blood-corpuscles of Mammals, but do not form pigment (hæmozoin). In films stained with Romanowsky's stain each parasite consists of a blue-staining cytoplasm and a red-staining nucleus, the latter generally consisting of a granule of chromatin with a string of finer granules extending from it. If a vacuole is present it becomes difficult to distinguish the form from the young ring-form of a malarial parasite. The organisms included in the suborder reproduce by division into two or four individuals only.

Reichenow (1929) places the family Theileriidæ under HÆMOSPORIDIA and the family Babesiidæ as an appendix to the same. In his latest work (1935) he places both the families in an appendix after the HÆMOSPORIDIA, and remarks that the life-cycle of the Theileriidæ, so far as is known, shows points of agreement with the typical HÆMOSPORIDIA but the plan of development of the Babesiidæ is quite different from the TELOSPORIDIA. In this work I have included the PIROPLASMIDEA under HÆMOSPORIDIA on grounds of practical convenience, and have divided the order into four families (see p. 209).

The two families, Theileriidæ and Babesiidæ stand in much the same relationship to one another as Hæmoproteidæ to Plasmodiidæ.

## Genus THEILERIA Bettencourt, Franca, & Borges, 1907.

Piroplasma (part), Stephens & Christophers, 1903 b, pp. 335-6; Theiler, 1904, pp. 1-20.

Theileria, Bettencourt, Franca, & Borges, 1907, pp. 341-9; Nuttall, Fantham, & Porter, 1909, pp. 325-40; Gonder, 1910 a, pp. 143-64; 1910 b, pp. 49-52; 1911 a, pp. 222-31; 1911 b, pp. 170-8; Minchin, 1912, pp. 379-80, 382; Nuttall, 1913, pp. 302-20; Hegner & Taliaferro, 1924, p. 305; Wenyon, 1926, pp. 992, 1029-39; Knowles, 1928, pp. 457-9; Reichenow, 1929, pp. 1022-7; Kudo, 1931, p. 289; Calkins, 1933, p. 566; Reichenow, 1935. p. 377.

With the characters of the family.

Remarks.--It is not easy to decide whether a particular form met with in the blood is a Theileria or a Babesia. In the former, schizogony takes place in the endothelial cells of the capillaries of the internal organs, and forms produced there enter the red corpuscles and are seen in the peripheral blood. Unlike Babesia, they do not multiply in the red corpuscles. The blood is consequently not infective when inoculated to healthy animals unless endothelial cells containing schizonts happen to be present.

#### 248. Theileria parva (Theiler). (Fig. 144.)

Piroplasma bigemina, a stage of, Koch, 1898.

Piroplasma kochi, Stephens & Christophers, 1903 b, pp. 335-6, fig. 75.

Piroplasma parvum, Theiler, 1904.

†Piroplasma parvum, Lingard, 1907, p. 274, pl. vii, fig. 1, 8. Theileria parva, Bottoncourt, Franca & Borges, 1907, pp. 341-9, pls. xvii, xviii; Nuttall, Fantham & Porter, 1909, pp. 325-40.

†Piroplasma parvum, Gaiger, 1910, p. 66.

Theileria parva, Gonder, 1910 a, pp. 143-64; 1910 b, pp. 49-52; 1911 a, pp. 222-31; 1911 b, pp. 170-8; Minchin, 1912, p. 382; 1911 a, pp. 222–31; 1911 b, pp. 170–8; Minchun, 1912, p. 382; Nuttall, 1913, pp. 302–20; Castellani& Chalmers, 1919, pp. 498–9, fig. 164; Hegner & Taliaferro, 1924, p. 305; Saceghem, 1925, pp. 651–60; Wenyon, 1926, pp. 1029–34, fig. 428; Sergent, Donatien, Parrot, Lestoquard, & Plantureax, 1926, p. 1362; 1927 a, pp. 489–506, 3 figs.; 1927 b, pp. 721–84, 23 figs.; 1927 c, pp. 161–87, 5 figs.; Knowles, 1928, p. 457, figs. 106, 107; Reichenow, 1929, pp. 1032–4, figs. 1007–11; Kudo, 1931, p. 289, fig. 122, i; du Toit, 1931, pp. 547–8; Beichenow, 1935, p. 377 Reichenow, 1935, p. 377.

The Cycle in Cattle.—The sporozoites injected by the tick collect in the spleen, lymphatic glands, and other organs, penetrate the endothelial cells of the capillaries, grow rapidly,

and form large multinucleate masses or schizonts (Koch's "blue bodies"). They may be discovered by puncture of the enlarged glands. In dried films, stained by Romanowsky's stain, they will be seen as blue masses of cytoplasm with a varying number of red chromatin dots. In films they may be found in the endothelial cells or free, but in sections they are always intracellular. About fourteen days after infection, and simultancously with the attack of fever, the schizonts may be recognized as of two kinds, some containing a smaller number of larger nuclei and others containing a larger number



Fig. 144.—Theileria parva (Theiler). A-D, stages in the red bloodcorpuscles; E, free schizont; F, intracellular schizont which gives rise to merozoites; G, free gametocyte-producing body; H, intracellular gametocyte-producing body; I, young gametocytes in a blood-corpuscle; J, ripe microgamete; K, ripe macrogamete; L, zygote; M, oökinete; N, multiplication cysts in the salivary glands of the tick; O, free sporozoites. (From Reichenow; A-D after Theiler's preparations; E-O, after Gonder.)

of smaller nuclei. The former break up into a number of minute bodies (merozoites), which enter other cells, grow, and reproduce by schizogony. The latter give rise to gametocytes which enter the blood and penetrate the red corpuscles. These forms in the blood are very small, ring-, comma-, pearshaped or rod-like; in the latter the nucleus lies at one end

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Sometimes cross-shaped forms are met with, but these are regarded as aggregates of four parasites, and not fission stages. The forms in the blood do not multiply by fission, and are consequently assumed to be gametocytes. They grow into adult gametocytes of two kinds—male gametocytes, which are long, slender, "bacillary" forms, and female gametocytes, which are plump, rounded or pear-shaped forms. The gametocytes can only develop further in the tick, *Rhipicephalus*.

Dimensions.—Schizonts  $3-10\mu$  in size; gametocytes, rodlike forms  $2.5\mu$  in length by  $1.2\mu$ .

The Cycle in the Tick.—In the gut of the tick the gametocytes leave the red corpuscles and develop into gametes. The macrogamete is stationary, whilst the microgamete is motile, seeks out the macrogamete, and is said to unite with it, forming the zygote. The latter develops into a worm-like oökinete, and no further development is known to take place till after the moulting of the tick. After the moulting of the tick, large cyst-like structures containing numerous nuclei are met with in the salivary glands. Each cyst develops a number of sporoblasts, and numerous very small sporozoites are ultimately set free, which are introduced into the cattle when the tick again sucks the blood.

*Remarks.*—The organism produces a serious disease of cattle. known as East Coast fever, in various parts of Africa. It has also been met with in Transcaucasia, Macedonia, and India. The disease differs from that caused by Babesia bigemina in that hæmoglobinuria, jaundice, and progressive anæmia are absent. Theiler (1904) demonstrated that East Coast fever was a distinct disease, and that cattle which had recovered from hæmoglobinuric fever due to Babesia were not immune to it. He also found that the disease could not be transmitted by inoculation of infected blood into a healthy animal. This was explained as being due to the fact that the blood-forms do not multiply. Experiments by Ed. Sergent and his coworkers (1926) have, however, shown that this view is incorrect, though, as Wenyon (1926) remarks, the occasional positive results obtained by blood inoculations of healthy animals may be due to the presence of endothelial cells in the peripheral blood.

The blood forms were carefully described by Nuttall, Fantham, and Porter (1909) in artificial infections of animals in England by means of ticks, *Rhipicephalus evertsi*, imported from South Africa; and also by Nuttall (1913). The lifehistory was studied by Gonder (1910, 1911 a, 1911 b), whose account has been followed above, but certain points lack confirmation.

Lingard (1907) and Gaiger (1910) recorded this species from India, but no one seems to have seen it since.

Habitat.—Blood of ox, Bos taurus Linn. (?): INDIA.

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## 249. Theileria mutans (Theiler). (Fig. 145.)

Piroplasma mutans, Theiler, 1906, pp. 292-300; 1909, pp. 115-33. Piroplasma mutans, Gaiger, 1900, pp. 202-300, 1900, pp. 110-30.
Piroplasma mutans, Theiler, 1911, pp. 202-3.
Babesia mutans, Minchin, 1912, pp. 380, 382.
Gonderia mutans, du Toit, 1918, p. 86, fig. 8.
Theileria mutans, Castellani & Chalmers, 1919, p. 499; Brumpt, 1923, pp. 54-64, 6 for . Dovla

1923, pp. 16-53, pls. i, ii ; Velu, 1923, pp. 54-64, 6 figs. ; Doyle, 1924, pp. 18-27.

1522, pp. 16-21.
†Theileria mutans, Edwards, 1925, pp. 48-9; 1926, p. 43; Cooper, 1926 a, pp. 96-7; 1926 b, pp. 315-16, pl. xviii, fig. 2.
Babesia mutans, Wenyon, 1926, pp. 1001-2, 1035, fig. 413; Knowles, 1928, pp. 451, 453, fig. 106.
Theileria mutans, Reichehow, 1929, p. 1025, fig. 1012; Du Toit,

1931, pp. 551-2.

†Theileria mutans, Cooper, 1931, pl. i, figs. 2, 3; Achar, 1935, p. 9. Theileria mutans, Reichenow, 1935, p. 377.

Blood-forms and schizonts very similar to those of T. parva. The blood-forms are exceedingly minute, comma-shaped, bacilliform or coccal; dumbbell-shaped, ring or even crossshaped forms occur, the largest not exceeding  $l \mu$  in diameter. Schizonts very similar to Koch's "blue bodies"; they are larger than those of T. parva, and the nuclei tend to be ovoid



Fig. 145.—Theileria mutans (Theiler).  $(\times c. 4000.)$  A-D, different forms in the blood-corpuscles. (From Wenyon, after Gonder.)

and not spherical as in the latter. They occur in small numbers in the internal organs as well as in the peripheral blood.

Transmission occurs by ticks of the genus Rhipicephalus; ticks fed at the nymphal stage transmitted the infection when feeding as adults.

Remarks.—Compared with T. parva, T. mutans is a benign parasite. The infections are mild in character, and the parasite does not cause hæmoglobinuria, though it may cause a certain amount of anæmia. Theiler (1906) showed that inoculation of infected blood readily conveys infection to healthy animals, and this was regarded as the chief distinction between T. parva and T. mutans, so much so that cross-forms which occur in both species were differently interpreted, in T. parva as being due to aggregation of four parasites, and in T. mutans as binary fission, and the latter organism was then

referred to *Babesia*. Brumpt (1923) showed that usually the infections are mild and the schizonts are not apparent, but in intense infections they occur in large numbers in the internal organs as well as in peripheral blood. This was confirmed by Velu (1923), Doyle (1924), and Edwards (1925). Ed. Sergent and his co-workers (1924) disagreed with Brumpt, and stated that T. mutans never produces schizonts, but later (1928) found a few of them. T. mutans and T. parva cannot be differentiated morphologically, and Brumpt (1924) differentiates them biologically as follows :---

plete immunity) ..... T. parva.

Du Toit (1931) recommends the provisional recognition of four species of *Theileria* in cattle, viz., T. parva, T. dispar, T. annulata, and T. mutans, and these show a gradual transition from the most virulent T. parva to the avirulent T. mutans.

Cooper (1926 a) remarked that T. mutans was almost universally present in Indian cattle, and showed the occurrence of Koch's "blue bodies" in this species. Datta (1938) has found that the parasite multiples in the lymphoid tissue generally, including that of glands, spleen, etc.

Habitat.—Blood of bulls, Bos indicus Linn. (?), used for Rinderpest hyper-immunisation: PUNJAB; UNITED PRO-VINCES, Muktesar; MYSORE, Bangalore.

#### 250. Theileria sp.

†Theileria sp., "types A & B," Sen & Srinivasan, 1937, pp. 15–37, pl. vii.

Usually round or slightly oval forms, having the appearance of signet-rings, and uniformly  $1-2\mu$  in diameter. Stained with Giemsa's or Leishman's stain, the cytoplasm takes a pale blue colour; the chromatin stains well, two chromatin spots being sometimes connected together. Koch's "blue bodies" present.

*Remarks.*—Sen and Srinivasan (1937) studied Theileriasis of cattle in a large number of Indian hill bulls artificially infected with a strain isolated from a fatal case at Muktesar. They came to the conclusion that in the present state of our knowledge it is impossible to rely on any morphological features as an indication of the extent of the pathogenicity of the organism. According to them, the diagnosis of the species of *Theileria* would appear to depend on the quantitative

combination of a number of morphological and clinical features. They consider the forms studied by them to be two types of a new species and give their differential characters alongside those given in du Toit's table of previously known species. The examination of blood taken at the first rise of temperature frequently reveals the presence of few parasites and Koch's " blue bodies " or both, but, as the disease advances, the parasites rapidly increase in number and may eventually invade 50 to 100 per cent. of the red blood-corpuscles, although Koch's "blue bodies" may vary in number from "rare" to " numerous."

Sen and Srinivasan also came to the conclusion that the infection in imported Friesian bulls is exotic in origin, being probably acquired by the animals during the course of their voyage to India, and the parasite concerned is Theileria annulata.

Habitat.-Blood of Indian hill bulls, Bos indicus Linn.: UNITED PROVINCES, Muktesar.

## 251. Thelleria cellii (Castellani & Chalmers).

*†Babesia cellii*, Castellani & Chalmers, 1910. Theileria cellii, Castellani & Chalmers, 1919, p. 500. Babesia cellii, Wenyon, 1926, p. 1027.

Bacillary and pear-shaped forms, lying side by side in the red blood-corpuscles. Development not known.

Remarks.-This form was originally described as Babesia cellii by Castellani and Chalmers, but in the third edition of their work they substituted the name Theileria cellii without giving any further details. It is by no means clear from the original description whether the form should be referred to Babesia or to Theileria.

Habitat.-Blood of Macacus pileatus (Shaw) : CEYLON.

## 252. Theileria hirci Dschunkovsky & Urodschevich. (Fig. 146.)

Theileria hirci, Dschunkovsky & Urodschevich, 1924, pp. 108-10, fig. A, 1-24; Wenyon, 1926, pp. 10326-8, fig. 430; Reichenow, 1929, p. 1026; Thomson & Hall, 1933, p. 228. Theileria ovis, Lestoquard, 1924, pp. 122-8, 15 figs. †Theileria ovis, Edwards, 1926, p. 43.

Theileria ovis, du Toit, 1931, p. 556. †Piroplasma taylori, Sarwar, 1935, pp. 172-5, pls. v, vi, vi a, vii. Theileria hirci, Reichenow, 1935, p. 378; Bhatia, 1936, pp. 151-4.

Parasites mostly small and variable in shape, some being distinctly ring-shaped, others bacillary or nail-like, oval or pyriform; occurring singly, in twos or, rarely, in threes in the centre of the red blood-corpuscle. Cross-like forms also occur, each member being pyriform. Extracellular forms, similar to Koch's "blue bodies," encountered in peripheral blood, were round or oval, at times as large or larger than a blood-

#### BABESIIDÆ.

corpuscle, and contained chromatin granules of varying size and shape. Internal organs of host contain the schizonts.

*Remarks.*—Sarwar (1935) described a form from a goat in the Punjab in which the parasites were mostly ovoid or round, pear-shaped forms being rare. Single parasites found in a corpuscle measured  $2\mu$  by  $1.5\mu$ . Although single parasites were frequently seen, it was not uncommon to find two, four, eight or sixteen individuals in one red cell. He also



Fig. 146.—Theileria hirci Dseh. & Urod. A-E, intra-corpuscular forms; F-G, extra-corpuscular bodies showing multiple division. (After Sarwar.)

found extracellular forms showing multiple division, but did not regard them as Koch's "blue bodies," nor did he find them in the internal organs of the host. He named the form *Piroplasma taylori*. I (1936) thought that the form described by Sarwar was identical with *T. hirci* Dsch. & Urod., 1924\*.

Habitat.—Blood of goat, Capra hircus Linn.: UNITED PROVINCES, Muktesar; PUNJAB, Malwale (Sheikhupura).

# 4. Family BABESIIDÆ Poche, 1913.

Non-pigmented parasites of the red blood-corpuscles of Mammals, which multiply in the corpuscle by division into two or four. They are of varying size and shape, and usually arrange themselves in pairs of pear-shaped individuals. The forms in the corpuscles are individuals reproducing asexually, but probably some are gametocytes.

Rémarks.—Franca (1917, 1918) gave a detailed classification of the family and recognized a number of genera. His classification was modified by du Toit (1918), who recognized six genera, viz., Babesia Starcovici, Nicollia Nuttall, Nuttallia Franca, Smithia Franca, Rossiella Nuttall, and Gonderia du Toit. Wenyon (1926), Reichenow (1929), and many other

<sup>\*</sup> Through his courtesy I have now examined his preparations, and the extra-corpuscular bodies (fig. 146, F, G) in the peripheral blood which I interpreted as Koch's "blue bodies" cannot be regarded as such. So his form ought to be transferred to *Babesia* and known as *B. taylori* (Sarwar).

writers, place all the species in a single genus, Babesia, with the characters of the family. Yakimoff (1931) retains the generic name Piroplasma and divides the genus into two subgenera, Piroplasma s. str. and Babesiella. Sometimes as many as a dozen or more of these parasites occur together in a mammalian red corpuscle. They produce no pigment, but destroy the corpuscle in which they are contained and set free the hæmoglobin, which is then excreted by the kidney of the host. Hence a characteristic symptom of the diseases produced by these parasites, generally termed "piroplasmoses" (or "babesioses"), is a great reduction of the corpuscles and a red coloration of the urine (hæmoglobinuria or "red-water").

## Genus BABESIA Starcovici, 1893.

(Syn. PIROPLASMA Patton, 1895.)

Hæmatococcus (non Agardh, 1828), Babes, 1888, p. 692; 1890, pp. 800, 975; 1891, p. 81; 1892, p. 359. Babesia, Starcovici, 1893, pp. 1–8. Pyrosoma (non Péron, 1804), Smith & Kilborne, 1893, p. 67.

Babesia, Kroguis & Van Hellens, 1894, pp. 353-64; San Felice & Loi, 1895, pp. 295-6.

Apiosoma (non Blanchard, 1885), Wandolleck, 1895, pp. 554-6.

Piroplasma, Patton, 1895, p. 498; Labbé, 1899, p. 124.

Babesia, Labbé, 1899, p. 125.

Piroplasma, Laveran, 1901, pp. 385-8; Minchin, 1903, pp. 254, 255, 265, 269, 309.

Babesia, Minchin, 1903, p. 269.

Piroplasma, Lingard & Jennings, 1904, pp. 161-5; Raymond, 1904, p. 272; Stephens & Christophers, 1904, pp. 332-7; Theiler, 1904, pp. 401-5; 1906 a, pp. 283-92; 1906 b, pp. 292-300; 1907, pp. 1-18; Dreyer, 1910, pp. 37-45; Nuttall & Strickland, 1910, pp. 524-5; 1912, pp. 65-96.

Babesia, Minchin, 1912, pp. 379-86. Piroplasma, du Toit, 1918, pp. 84-104; Castellani & Chalmers, 1919, pp. 492-8.

Babesia, Wenyon, 1926, pp. 992–1028; Cooper. 1926 a. pp. 314–15; Knowles, 1928, pp. 445–57; Reichenow, 1929, pp. 1032–40.

Piroplasma, Yakimoff, 1931, pp. 372-400.

Babesia, Kudo, 1931, p. 289; Calkins, 1933, p. 566; Reichenow, 1935, p. 378.

Intra-corpuscular parasites, without pigment. Pyriform (lancet-shaped), mostly in pairs, ring-shaped or elliptical.

The genus includes parasites from dogs, cattle, pigs, sheep, horses, rats, mongooses, monkeys, etc., which are transmitted by ticks.

Remarks.--Species belonging to this genus have been described in India or Ceylon from dogs, jackals, horses, cattle, goats, mongooses, monkeys, etc. Raymond (1904) was the first to draw attention to the presence of indigenous Piroplasma in Bovines in India in 1898. Lingard and Jennings (1904) described and figured specimens of Piroplasma from a large number of Indian animals and referred them all to a single

species, *Piroplasma tropicus*, instead of to several previously described species, such as P. bigeminum, P. canis, P. bovis, P. ovis, etc. Yakimoff (1931), after reviewing the previous classifications of Piroplasma, divides the genus into two subgenera, as follows :----

(1) Of typical pyriform shape, often in pairs. Length of pyriform organisms greater than the radius of the erythrocyte. Situation in the corpuscle central. Pyriform individuals of a pair form an acute angle. Number of chromatin masses in budding pear-shaped forms not less than two. Pear-shaped forms more numerous than the round forms. Trypan blue effective. Species included are P. bigeminum, P. caballi, P. canis, P. motasi [=P. ovis]. Subgenus Piroplasma s. str.

(2) Form pear-shaped (lancet-shaped), annular or elliptical. Length of the pear-shaped forms smaller than or equal to the radius of the erythrocyte. Situation in corpuscle peripheral or central. Pyriform individuals of a pair form an obtuse angle. In the pear shaped forms there is a single chromatin Round forms predominate over the pear-shaped ones. mass. Trypan blue may or may not be effective. Subgenus Babesiella Mesnil, 1919.

The subgenus Babesiella is further subdivided into species groups as follows :---

- Group 1. Babesiella s. str. Form smaller than the radius of the erythrocyte; situation peripheral; trypan blue effective ..... B. bovis.
- Group 2. Françaiella. Form smaller than or equal to the radius; situation central; trypan blue not effective.
  - (a) Form equal to radius .. F. colchica, F. major.
  - (b) Form less than the radius. F. argentina, F. berbera,

F. caucasica. F. occidentalis.

This scheme is convenient for diagnostic purposes, but obviously the names Babesiella and Françaiella cannot be used in a generic sense.

253. Babesia bigemina (Smith & Kilborne). (Fig. 147.)

Pyrosoma bigeminum, Smith & Kilborne, 1893 a, p. 67; 1893 b,

Piroplasma bigeminum, Lingard, 1993.
 Piroplasma bigeminum, Lingard, 1993.

- †Piroplasma sp. (probably bigeminum), Donovan, 1903, p. 1401.
- *Piroplasma bovis*, Stephens & Christophers, 1904, pp. 333-4, pl. iii.

†Piroplasma bigeminum, Holmes, 1904, pp. 317-26, 1 pl.; Lingard & Jennings. 1904, pp. 161-5, pl. i, figs. 4, 5; 1907, pl. vii, fig. G, 9. Piroplasma bovis, Nuttall & Graham-Smith, 1908, pp. 138-9, diagram ii.
- *Piroplasma bigeminum*, Gaiger, 1910, p. 66; Baldrey, 1910, pp. 569-79.
- †Piroplasma (Babesia) bigeminum, Edwards, 1925, pp. 48-9.
- Piroplasma bigeminum, Minchin, 1912, p. 379, fig. 160; pp. 384-5, fig. 162; Castellani & Chalmera, 1919, p. 497; Velu, 1922, pp. 133-45, fig. 21; Hegner & Taliaferro, 1924, p. 304.
- +Piroplasma bigeminum, Cooper, 1926 a, p. 96; 1926 b, pp. 314-
- 15, pl. xviii, fig. 1. Babesia bigemina, Wenyon, 1926, pp. 993-8, figs. 408, 410 A-J, pl. xviii, figs. 6-10; Knowles, 1928, p. 451, figs. 105, 106; Reichenow; 1929, pp. 1034-5, fig. 1019; Dennis, 1930, pp. 179-92, 2 pls.; Kudo, 1931, p. 289, fig. 122, g.
- *Babesia bigemina*, Cooper, 1931, pl. i, fig. 1; Ware, 1932, p. 31; Ray, 1938, p. 265.

Largest of the known Piroplasms of cattle. Round, oval or irregular, or pyriform and occurring in pairs, individuals of the pair lying close together : the pear-shaped forms extend across the diameter of the red corpuscle, which in cattle measures from 5 to  $6\mu$ . Occasionally four pear-shaped forms are arranged in a fan-like manner. Multiplication takes place



Fig. 147.—Babesia bigemina (Smith & Kilborne).  $(\times c. 3000.)$ A, pear-shaped form; B-F, multiplication by budding; G, two pear-shaped forms in a corpuscle; H, four pearshaped forms in a corpuscle. (From Wenyon, after Nuttall and Graham-Smith.)

by a characteristic budding process, the buds remaining attached by their pointed ends. Transmission by ticks. Dimensions.—Round forms  $2-3\mu$  in diameter; pyriform

examples 2-4  $\mu$  in length by 1.5-2  $\mu$  in width.

Remarks.-It was in the case of B. bigemina, the cause of red-water fever in cattle in Texas, that Smith and Kilborne (1893) demonstrated the possibility of transmission of protozoal parasites by Arthropod hosts. Not only did they discover that infection is transmitted by the tick, but that the eggs of an infected tick are also infected, so that the second generation of ticks hatched from such eggs may also be infective. This was the first time that the rôle of Arthropods in the transmission of protozoal disease was demonstrated-several

years before Ross's discovery of the transmission of Plasmodium præcox by Culex mosquitoes. Not much, however, is known of the actual development in the tick. Koch (1906 a) observed that in the stomach of the tick the pear-shaped forms escaped from the blood-corpuscles, became amœboid, threw out long spiky pseudopodia, associated in pairs, and gave rise to elongate bodies with two nuclei and spiky pseudopodia at both extremities. Fusion of the nuclei then took place, along with the withdrawal of the pseudopodia. Christophers (1907), who studied the development of B. can is in the tick, doubted if the amœboid stages were essential in the development, and described motile club-shaped forms, which he referred to as "zygotes" or "oökinetes." These give rise to a globular stage, which divides by multiple fission into sporoblasts and sporozoites. The sporozoites collect in vast numbers in the salivary glands of the tick, and thence pass into the Vertebrate when the tick feeds. Observations on the development in cultures were published by Kleine (1906) and by Nuttall and Graham-Smith (1908), and certain stages in the tick were also described by Dschunkovski and Luhs (1909).

The development of the parasite in the Vertcbrate host consists of multiplication by binary or quadruple fission within the corpuscle. After destroying the corpuscles in which they are lodged, the parasites become free in the blood and penetrate other red corpuscles. The stages in the blood were studied in great detail by Nuttall and Graham-Smith (1906, 1907, 1908) and by Christophers (1907) in *B. canis*, and a summary of their observations will be found under that species.

The organism remains in the blood of animals, that have recovered from acute symptoms, for many years after its apparent disappearance, as tested by microscopic examinations, and inoculation of blood can produce infection in a healthy animal.

Cooper (1926 a) is of the opinion that infection with this parasite is so widespread in India that probably in most localities cattle become infected as young calves, at which age they possess a high degree of resistance and recover readily, and are subsequently immune, but continue to act as carriers. Acute disease is liable to occur when adult cattle are imported from countries where the species does not exist, or when the immunity is broken down through the effect of intercurrent disease conditions.

Dennis (1930) described the structure of the nucleus of B. bigemina and even noted the presence of a blepharoplast.

Ray (1938) found the nuclear chromatin to be confined to the apical portion of the parasite, with a circular row of fine SPOR. X chromatin beads connected with the main mass of chromatin. He did not find any evidence of a blepharoplast.

Habitat.—Blood of ox, Bos indicus Linn. (?): PUNJAB, Lahore; UNITED PROVINCES, Muktesar; MADRAS.

## 254. Babesia bovis (Babes). (Fig. 148.)

Hæmatococcus bovis, Babes, 1888, p. 692; 1890, pp. 800, 975.
Babesia bovis, Starcovisi, 1893, pp. 1-8; Labbé, 1899, p. 125.
Piroplasma divergens, MacFadyean & Stockman, 1911, p. 340; Nutall, 1913, p. 305, fig. 2.
Piroplasma bigeminum (Babesia bovis), Minchin, 1912, p. 379.
Microbabesia divergens, Sohns, 1918.
Babesiella bovis, Meenil, 1919.
Babesia bovis, Wenyon, 1926, pp. 998-1000, figs. 411, 412; Reichenow, 1929, pp. 1035-6, fig. 1021; 1935, p. 378.
†Babesia bovis, Idnani, 1938, p. 265; 1938, p. 42.

Smaller than B. bigemina. Ameeboid forms chiefly rounded, often ovoid, pear-shaped, or rod-shaped forms also occur.



Fig. 148.—Babesia bovis (Babes). (After Nuttall.)

In films stained with Romanowsky's stain the nucleus is seen as a red dot, with a string of fine granules extending from it. Two pear-shaped individuals often lie with their pointed ends together and forming an obtuse angle, or even in a line. The forms are smaller than the radius of the corpuscle, usually lie near the margin of the corpuscle, and trypan blue is usually effective.

Dimensions.—Round forms  $1-1.5\mu$  in diameter; pyriform individuals  $1.5-2\mu$  in length.

*Remarks.*—The organisms were marginal in position and the pointed ends of a pair of individuals included an obtuse angle, or the individuals were in a straight line. The host had shown acute piroplasmosis associated with hæmoglobinuria, and trypan blue had been administered, but without producing any effect.

Habitat.—Blood of an Indian buffalo, Bos bubalus Linn.: MADRAS, Belgaum.

## 255. Babesia caballi (Nuttall & Strickland). (Fig. 149.)

Piroplasma sp., Marzinowsky & Bielitzer, 1909, pp. 28-31, pls. vvii.

- Piroplasma caballi, Nuttall & Strickland, 1910, pp. 524-5; 1912, pp. 65-96, pl. iii, 8 diagrams and 5 charts; Dschunkovski & Luhs, 1913, pp. 289-302, pls. xiv, xv : Darling, 1913, pp. 197-
- & Luns, 1913, pp. 209-302, pp. xv.; Darning, 1913, pp. 10-7-203, pl. iii; Carpano, 1913 b, pp. 13-41, 3 pls. *†Piroplasma caballi*, Valladares, 1914, pp. 88-94. *Piroplasma caballi*, Yakimoff, Schokhor, & Koselkine, 1917, pp. 302-11; Velu, 1922, pp. 197-212, fig. 27. *Babesia caballi*, Wonyon, 1926, p. 1008, pl. xviii, 21-25, & fig. 415; Kosenka, 1052, p. 452, 66, 1052, 22-30.
  - Knowles, 1928, p. 453, fig. 105, 22-30; Reichenow, 1929, pp. 1036-8.

Parasites relatively large, and resemble B. bigemina found in cattle. Multiplication takes place by budding, and pearshaped forms occur in pairs in the red blood-corpuscles.



Fig. 149.-Babesia caballi (Nuttall & Strickland). A, pear-shaped form; B-D, stages in the process of budding; E, a pair of pear-shapod individuals. (After Nuttall and Strickland.)

*Remarks.*—This form, like *B. equi* (Laveran, 1901), causes hæmoglobinuric fever in horses. Nuttall and Strickland (1910) showed that two distinct species occur in horses. They placed the small form, previously known as *Piroplasma* equi Laveran, in a new genus under the name Nuttallia equi, and described the larger form as Piroplasma caballi. Later (1912) they described the method of multiplication of the latter in the blood.

Marzinowsky and Bielitzer (1909) showed that the tick, Dermacentor reticulatus Koch, was the vector in South Russia, and Carpano (1913 b) thought that Margaropus annulatus (Sav) was the vector in Italy.

Habitat.-Blood of the horse, Equus caballus Linn.: MADRAS, Madras.

#### 256. Babesia canis (Piana & Galli-Valerio). (Figs. 150, 151.)

Pyrosoma bigeminum var. canis, Piana & Galli-Valerio, 1895, p. 345.

Piroplasma canis, Labbé, 1899, p. 124; Lounsbury, 1901, p. 714; Minchin, 1903, pp. 242, 270, 337, 350; Lounsbury, 1904, p. 113; Nuttall, 1904, pp. 219-57, 7 figs. & 7 charts.

- †Piroplasma canis, Daggetty, 1904, pp. 67-8; Lingard & Jennings, 1904, pp. 161-5, pl. i, fig. 10; Stephens & Christophers, 1904, pp. 334-5, fig. 74.
  - Piroplasma canis, Nuttall & Graham-Smith, 1905, pp. 237-49; 1906, pp. 586-651, pls. xi-xiii and 23 text-figs.; 1907, pp. 232-72, pls. i-iii.
- *†Piroplasma canis*, Webb, 1906, p. 1; Christophers, 1907 a, pp. 76-8; 1907 b, pp. 1-83, 3 pls.
- Piroplasma canis, Nuttall & Graham-Smith, 1909, pp. 211-14, pl. ix.
- \*Piroplasma canis, Baldrey, 1910, pp. 569-79; Gaiger, 1910, p. 66; Brandford, 1912, pp. 643-6.
  - Babesia canis, Schuberg & Reichenow, 1912, pp. 415-34, pl. ii.
- Piroplasma canis, Minchin, 1912, pp. 382-3, 384-5, 387, figs. 161, 162.
- †Piroplasma canis, de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, p. 16.
  - Piroplasma canis, Castellani & Chalmers, 1919, pp. 493-6, fig. 163;
     Velu, 1922, pp. 224-40, figs. 29, 30; Hegner & Taliaferro, 1924,
     pp. 303-4, fig. 114.
- \*Piroplasma canis, Rau, 1926, pp. 243-4.
- Babesia canis, Wenyon, 1926, pp. 1012-20, pl. xviii; figs. 410, 417, 418, 419, 421; Knowles, 1928, pp. 446-50, figs. 103, 104; Reichenow, 1929, pp. 1032-4, figs. 1016, 1018.
- *†Piroplasma (Babesia) canis, Stirling, 1929, pp. 647-53.* 
  - Babesia canis, Belitzer & Markoff, 1930, pp. 498-601; Regendanz, 1932, pp. 745-8; Regendanz & Reichenow, 1933, pp. 50-71, 2 pls., 1 fig.; Reichenow, 1935, p. 378, fig. 31.

The Cycle in the Dog.—Parasites typically pear-shaped, rounded and bulbous at one end and pointed at the other, with



Fig. 150.—Babesia canis, development in the dog ( $\times$  c. 3000). A, pearshaped form; B, vacuolated form; C, rounded form; D-G, stages in the process of budding; H-J, ultimate formation of pear-shaped individuals. (After Nuttall and Graham-Smith.)

a vacuole in the cytoplasm;  $4.5-5\mu$  in length. In films stained with Romanowsky's stain the nucleus is seen as a deeply stained granule situated near the pointed end, with a string of fine granules extending from it. This typical BABESIA.

form passes through a series of stages before division takes place. The organism becomes rounded, the vacuole first enlarges, then disappears, and the string of chromatin granules is withdrawn into the larger granule (fig. 150, A-C). It now passes through an amœboid phase, and division of the organism is initiated by the separation from the larger granule of chromatin of a smaller one, which remains connected with the larger by a string of granules. The smaller granule divides into



Fig. 151.—Babesia canis, development in the tick ( $\times$  c. 1500). A, globular forms in the stomach of the tick; B, globular form showing split; C, club-shaped forms; D, "zygote," which passes through the stomach-wall; E-H, globular body in ovum or tissue-cells, which shows multiple fission; I, sporoblasts; J, formation of sporozoites; K, sporozoites in the salivary glands. (From Nuttall, after Christophers.)

two, and the cytoplasm forms two buds, each containing one of these granules; thus finally a trilobed form results, in which the three chromatin granules are connected with a Y-shaped or V-shaped chromatin strand (fig. 150, D-G). The larger chromatin granule also now divides, followed by the division of the cytoplasm, and two pear-shaped merozoites result (fig. 150, H-J). These merozoites may now leave the corpusele to invade other corpuseles or may further divide within the same corpusele so as to produce multiple-cell infections. Schizogony is thus by budding, and is not comparable with that of the malarial parasites. Sometimes there may be as many as sixteen of these organisms in a single blood-corpusele.

The Cycle in the Tick.—When ingested into the stomach of the tick the parasites leave the blood-corpuscle, increase in size, and become globular bodies,  $4-5\mu$  in diameter. A split appears in the globular body, a portion swings round, and the globular body is changed into a club-shaped body. The clubshaped bodies are motile and Gregarine-like. Whether any sexual process is involved is not known. The club-shaped bodies, after passing through the wall of the gut, enter the ova and later are found in the tissue-cells of the embryo developed from the egg. There they again become globular, and increase in size up to a diameter of  $25\,\mu$ . This globular stage, termed "zygote" by Christophers, according to Minchin probably corresponds to the occust of the Plasmodiidæ. The globular body divides up by multiple fission into a number of "sporoblasts," which do not remain aggregated together, but scatter themselves through the tissues of the tick, larva, nymph, or adult as the case may be. The nucleus of each sporoblast divides into a large number, and then the sporoblast segments into an equal number of sporozoites, which are small bodies with a single nucleus similar in appearance to the forms in the blood. The sporozoites collect in the salivary glands of the tick and pass into the blood of the dog when the tick next feeds on it.

Remarks.—The parasite was first discovered as the cause of malignant jaundice of dogs in Italy, and has since been shown to have a wide distribution. It has long been known to yield to treatment with the aniline dye, trypan blue. Lingard and Jennings (1904), James (1905), Webb (1906), and Christophers (1907 a, 1907 b) were the earliest to study it in India. Christophers showed that in India Rhipicephalus sanguineus (Latreille) was the transmitting tick, and described the cycle in the tick as summarized above. Christophers also proved that eggs laid by an adult which had fed on infected dogs gave rise to larvæ which were not infective, but that the nymphs, and probably the resulting adults, were infective. James took specimens of R. sanguineus to England and succeeded in infecting English dogs with them. Baldrey (1911) gave a useful summary of the observations on piroplasmosis in India up to that date.

Lounsbury (1901, 1904 a) was the first to demonstrate experimentally that infection is transmitted from dog to dog by the tick *Hæmaphysalis leachi* (Audouin) in South Africa. Nuttall (1904) and Nuttall and Graham-Smith (1905, 1906) described the structure and life-history of the parasite in the dog. Breinl and Hindle (1908) described a biflagellate stage of the parasite, but Wenyon (1926) and Knowles (1928) are of the opinion that this must have been a species of Bodo (Mastigophora) from some extraneous source, and was not part of the life-cycle of B. canis. Schuberg and Reichenow (1912) concluded from their observations that the amœboid forms are extra-corpuscular, become rounded, and produce buds, and these buds pass into the red corpuscles, producing there the intra-corpuscular pairs of pear-shaped forms. They also studied the details of nuclear division in wet fixed films.

Nawrotzky (1912) infected dogs by introducing infected blood into the stomach by means of a stomach tube. Laveran and Nattan-Larrier (1913) adduced evidence to show that the viruses of France and North Africa were different, dogs which had recovered from infections with French virus, and were immune, could be infected with the North African one.

Several workers have attempted to infect animals other than dogs, but without success. Dunsbury (1903) failed to infect a jackal. Nuttall and Graham-Smith (1909 a) failed to infect foxes. Contrary to these negative results, Rau (1926) has succeeded in experimentally infecting a jackal with *B. canis* from a dog. He also found the parasite in a bloodsmear from a wild jackal.

Regendanz and Reichenow (1933) have restudied the development of Babesia canis in Dermacentor reticulatus. They find that most of the parasites when sucked in with the dog's blood by the tick die, but some produce small vermiform bodies that infect the cells of the tick's midgut and multiply there asexually. The daughter cells enter the body-cavity and proceed to the eggs, where, after a few divisions, they decrease in size and remain dormant. Later, in the nymph or the adult developing from the egg, they enter the salivary glands, multiply and produce vermiform cells that escape into the salivary ducts; thence they are passed into the dog. In the dog's blood they reproduce asexually. No sexual stages were found by them at any stage of the life-history, and they believe that the sexual stages previously reported by others are misinterpretations of the observations. They further claim that piroplasms have no close relationship with either Sporozoa or Flagellata, but have their nearest affinities with Sarcodina.

Habitat.-Blood of the dog, Canis familiaris Linn. : MADRAS, Madras; PUNJAB; PORTUGUESE INDIA, NOVA GOA; CENTRAL **PROVINCES**; the jackal, Canis aureus Linn. : MADRAS, Madras; and the blood of the transmitting tick, Rhipicephalus sanquineus Koch.

## 257. Babesla equi (Laveran). (Fig. 152.)

Piroplasma equi, Laveran, 1901, pp. 385-8, text-figs.; Minchin, 1903, pp. 242, 270, 350; Stephens & Christophers, 1904, p. 337.

†Piroplasma sp., Lingard & Jennings, 1904, pp. 161-5. pl. i, figs. 2,

Plasmodium equi, Axe, 1906, pp. 222-5; Joliffe, 1907, pp. 51-66. Piroplasma equi, Gaiger, 1910, p. 66.

Nuttallia equi, Franca, 1910, p. 14; Nuttall & Strickland, 1910, pp. 524-5; Minchin, 1912, p. 380; Dschunkvoski & Luhs, 1913, pp. 289-302, pls. xiv, xv. Piroplasma equi. Carpano, 1913 b, p. 845; 1914 a, pp. 13-41.

3 pls.; 1914 b, pp. 42-53, 1 pl. †Piroplasma equi, Valladaros, 1914, pp. 88-94; Williams, 1914,

pp. 1-6.

Nuttallia equi, Schein, 1917, pp. 871-2; Yakimoff, Schokhor, & Koselkine, 1917, pp. 302-11; Du Toit, 1919, pp. 84-104; Castellani & Chalmers, 1919, p. 500.

Piroplasma equi, Velu, 1922, pp. 197-212, fig. 28.

Babesia equi, Wenyon, 1926, pp. 1009-10, fig. 416, pl. xviii, figs. 26-30; Knowles, 1928, p. 453, fig. 106, 26-30; Reichenow, 1929, pp. 1036-7, fig. 1022.

Very small, nearly rounded and actively ameboid, ovoid or pear-shaped, not exceeding  $2\mu$  in diameter. Division



Fig. 152.—Babesia equi (Laveran). (After Wenyon,)

gives rise to four individuals, which are arranged in a crosslike manner. The four daughter individuals eventually separate, escape from the red corpuscle, and infect other corpuscles. Transmission by ticks.

Remarks .-- Round or ring-shaped, oval or rod-shaped piroplasms showing division rosettes of four, arranged in the form of a cross, were placed by Franca (1910) in a separate genus called Nuttallia: but Wenyon (1926) thinks that differences in size and in the number of daughter individuals produced are not sufficient grounds for recognition of new genera.

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Nuttall and Strickland (1910) have described the growth and division stages in this species. The species produces a disease like that caused by *B. caballi*, but is much more widely distributed. Theiler (1905 a) showed that *Rhipi*cephalus evertsi Neumann is the vector in South Africa, and according to Carpano (1913 b), *Rhipicephalus bursa* Canestrini & Fanzega is the vector in Italy.

Young animals are not so seriously affected as older ones. Immunity is acquired. The blood remains infective for many years after a clinical recovery.

Habitat.—Blood of the horse, Equus caballus Linn.: PUNJAB; UNITED PROVINCES, MURTESAR; RAJPUTANA; MADRAS.

## 258. Babesia felis Davis. (Fig. 153.)

*Pyroplasma* sp., Lingard & Jennings, 1904, p. 161. *Babesia felis*, Davis, 1929, pp. 304, 523-34. *Babesia felis*, Mangrulkar, 1937 a, p. 15; 1937 b, pp. 243-6, pl. xvi.

Small, rounded, non-pigmented, intra-corpuscular parasite multiplying by division into four in a cross-like arrangement,



Fig. 153.—Babesia felis Davis. Various forms seen in Leishmanstained films. Light blue cytoplasm is indicated by stippling and the dark red chromatin by black. Outlines of the corpuscles are represented schematically. (After Davis.)

Schizonts do not occur in the internal organs. Trypan blue not effective.

Dimensions.— $1-2\cdot25\mu$  in diameter, the majority being about  $1\cdot25\mu$ .

Remarks.—Lingard and Jennings (1904) recorded piroplasmosis in wild and tame cats, but did not describe the form found. Davis (1929) described Babesia felis from the Sudanese wild cat, Felis ocreata, and found that the parasite is readily transmissible to the domestic cat by means of blood inoculation, and that the progress of infection in inoculated cats follows a constant but benign course. Splenectomy prior to or after inoculation results in an intense infection characterized by anæmia and hæmoglobinuria. Mangrulkar (1937) has recently found the form in Indian cats, but has not come across any division into four or cross-forms.

Habitat.—Blood of cat, Felis domesticus Linn.: UNITED PROVINCES, Bareilly, Muktesar.

#### SPOROZOA.

#### 259. Babesla gibsoni (Patton). (Fig. 154.)

†Piroplasma tropicus (part), Lingard & Jennings, 1904, pp. 161-5. †Piroplasma gibsoni, Patton, 1910, pp. 274-81, fig. 1.

†Piroplasma tropicus (part), Baldrey, 1910, pp. 572-7. †Piroplasma gibsoni, Patton, 1911, pp. 615-21, 1 fig.; Symons & Patton, 1912, pp. 361-70.

Achromaticus gibsoni, Franca, 1917, pp. 221-9.

- Piroplasma gibsoni, Castellani & Chalmers, 1919, p. 497. Babesiella gibsoni, Velu, 1922, p. 131. Babesia gibsoni, Wenyon, 1926, pp. 1020–1, fig. 422. Piroplasma gibsoni, Symons, 1926, pp. 293–315. †Piroplasma gibsoni, Rau, 1927, pp. 785–800, pls. xxxi, xxxii, 4 text-figs.; Stirling, 1929. pp. 647-53. †Babesia gibsoni, Swaminath & Shortt, 1937, pp. 499-503; Ray &
- Idnani, 1938, p. 265.

Smaller than B. canis; young trophozoites rounded, not pear-shaped, and appear as small ring-like forms occupying about one-eighth of the infected corpuscle. Parasites (in films stained with Leishman's or Gienisa's stain) appear as small rings, somewhat resembling the rings of the malignant tertian malarial parasites but very much smaller and staining



Fig. 154.—Babesia gibsoni (Patton). (From Wenyon, after Patton.)

less deeply. They also differ from B. canis in not showing reticular structure of protoplasm or vacuoles. The protoplasm takes a very faint blue stain, the ring being sometimes made up of a blue-staining periphery and a clear centre. Generally there are two masses of compact chromatin, staining red, one larger and the other smaller, which is situated either near the larger or on the periphery of the ring. Larger ovoid or elongate forms are also met with, though in smaller numbers. Parasite usually excentrically situated in the bloodcorpusele. Usually only one, but sometimes up to five, may be found in a single corpuscle. The infected host-cell generally not altered in shape or size. Sometimes the parasite escapes from the corpuscle, becomes disc-shaped, and after a short while enters another corpuscle. In spleen-puncture smears similar forms are met with, and also very large rings with their chromatin divided into two, three or even four parts. Schizogony takes place in the spleen. The parasite, having penetrated a red cell in the spleen, grows and divides into two, four, etc. The daughter parasites are somewhat pear-shaped, and, when

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present in large numbers in a cell, the shape varies from a long oval to pyriform or circular. The merozoites are set free by the rupture of the corpuscle and infect other corpuscles.

Remarks.—Patton (1910) believed that the hounds got the infection from the jackal. He demonstrated the same parasite in the blood of the jackal, and showed that the infection could be transferred to dogs my means of inoculation of blood from the infected animal. Dogs which had recovered from *B. canis* were inoculable with *B. gibsoni*. He suspected *Rhipicephalus* sp., a species related to *Rhipicephalus* simus (Koch) which occurs on the jackal, as the transmitting agent.

Baldrey (1911) thought that the outbreak described by Patton as due to this species was of the same nature as that previously described by Pease and Gunn (1908), and that the form is not morphologically distinct from *Babesia tropicus* (Lingard & Jennings, 1904). Lingard and Jennings studied piroplasmosis in different animals, but probably confused a number of species, and *B. tropicus* is not now recognized as a distinct species.

Rau (1927) observed that smears from engorged larvæ of the tick, *Hæmaphysalis bispinosa* Neumann, showed some parasites with fibrillar prolongations and a few free large rings. Transmission experiments with *H. bispinosa* were inconclusive. Cultivation of the parasite *in vitro* was also not successful. The blood from an infected jackal when injected into a dog gives the disease to it and vice versa. In the dog, however, the disease takes a more acute form than in the jackal.

According to Stirling (1929) the infection caused by this much smaller piroplasm differs clinically from that caused by P. canis in being usually more prolonged in its effects. The symptoms are generally those of a progressive anæmia with frequent tendency to relapses at prolonged intervals. It does not respond to treatment with trypan blue, but good results have been obtained by repeated treatment with certain arsenical preparations, notably tryparsamide.

Swaminath and Shortt (1937) have shown that the jackaltick, *Hæmaphysalis bispinosa* Neumann, is a vector of B. gibsoni, and that all stages of the tick can transmit. Hereditary transmission through the egg also occurs.

Ray and Idnani (1938) have recently made a detailed study of *B. gibsoni* in the Vertebrate host. They found two types of parasites, viz., ring forms and thin elongate forms, in smears from the peripheral blood as well as internal organs. The ring forms were found to multiply by repeated binary fission until 12 to 16 merozoites were formed; while the thin elongate forms were observed to multiply in a manner which suggested a process of schizogony and gave rise to more than 30 merozoites in a corpuscle. Sometimes corpuscles infected with both forms were seen in smears from peripheral blood, and they regard this as suggestive of sexual dimorphism.

Habitat.—Blood of the jackal, Canis aureus Linn.: CEYLON (?); MADRAS, Kurnool; blood of the dog, Canis familiaris Linn.: CEYLON (?); MADRAS; CENTRAL PROVINCE. Body of Hæmaphysalis bispinosa Neumann: MADRAS, Madras.

## 260. Babesia motasi Wenyon. (Fig. 155.)

Babesia bigemina, Motas, 1903 a, pp. 1522-4; 1903 b, pp. 501-4. Piroplasma ovis, Dschunkovski & Luhs, 1909, pp. 149-51; Lestoquard, 1925, pp. 140-5.

Babesia motasi, Wenyon, 1926, pp. 1005-6, fig. 414, 9-12.

\*Babesia motasi, Achar & Srikantiah, 1934, pp. I-3, 2 pls. & 2 charts. Babesia motasi, Sarwar, 1935, pp. 171-2; Thomson & Hall, 1933, pp. 218-220, figs. 1-6.

Parasites are most frequently pear-shaped, though ovoid, round or irregularly shaped forms are also met with. They occur singly or in pairs, and the pairs always meet at an acute



Fig. 155.—Babesia motasi Wenyon. (×3000.) (From Wenyon, after Lestoquard.)

angle at their narrower ends. Chromatin often appears as double.

Dimensions.  $-2.5-4\mu$  in length by  $1.2-3\mu$  in breadth.

Remarks.—Motas (1903) found this parasite in sheep and considered it to be identical morphologically with *B. bigemina* of cattle. He also described the transmission of the infection through the agency of the tick, *Rhipicephalus bursa* Canestrini & Fanzega. The disease caused by this parasite also resembles red-water fever of cattle, and is said to occur in acute or chronic form. Lestoquard (1925) observed this parasite in the blood of goats in Algeria, and showed that the parasites were readily inoculable from one goat to another. He referred to it as *Piroplasma ovis*, but as the name *Babesia ovis* is already preoccupied for a piroplasm of sheep of intermediate size, Wenyon (1926) re-named it *B. molasi*.

Achar and Srikantiah (1934) have recorded the finding of this species in sheep. They have conducted a few transmission experiments, and report that the parasite was not transmissible to goats.

Habitat.-Blood of sheep, Ovis sp. : MYSORE STATE.

## 261. Babesia ninense (Yakimoff). (Fig. 156.)

Piroplasma ninense, Yakimoff, 1909, pp. 472-7, 1 pl. †Nuttallia ninense, Sinton, 1922, pp. 359-63, pls. xxvi, xxvii. Babesia ninense, Wenyon, 1926, p. 1358. Nuttallia ninense, Reichenow, 1929, p. 1039.

Parasites occur in the red blood-cells, usually one in a cell, occasionally more. Stained with Giemsa's stain the cytoplasm is blue and the nucleus a deep carmine-red. The cytoplasm is condensed and deeply stained at the periphery, with usually a lighter area in the neighbourhood of the nucleus. Cytoplasm is voluminous as compared with the nucleus, which is a solid mass of chromatin, usually round, but elongated in dividing forms, and is usually peripheral, rarely central. The intracellular parasites may be ring-shaped, oval or elongate rod-like in form. The oval or oat-shaped form is the common type in division rosettes. Division rosettes consisting of four small parasites found in smears from blood, spleen, and bonemarrow. Cross-form of division also met with.



Fig. 156.—Babesia ninense (Yakimoff). (×c. 2500.) (After Sinton.)

Dimensions.—Intracellular parasite, small form  $0.7-1\mu$ , large form  $2-3\mu$  in length.

*Habitat.*—Red blood-corpuscles in smears from the blood, liver, spleen, and bone-marrow of the hedgehog, *Erinaceus* sp. : N.W.F. PROVINCE, Kohat.

#### 262. Babesia sergenti Wenyon. (Fig. 157.)

Piroplasma ovis (part), Ratz, 1913, pp. 194-200. Theileria ovis, Yakimoff, 1916, p. 201. Gonderia ovis, Sergent, Parrot & Hilbert, 1922, pp. 789-92, 1 fig. Piroplasma ovis, Velu, 1922, pp. 216-23. Gonderia ovis, Lestoquard, 1924, pp. 122-8, 15 figs. Babesia sergenti, Wenyon, 1926, pp. 1004, 1007-8, fig. 414, 1-4. Babesia ovis, Reichenow, 1929, p. 1036. †Babesia sergenti, Krishna Iyer, 1933, p. 33.

Morphologically the parasite corresponds very closely to B. mutans. Similar rounded and bacillary forms occur. Reproduction is by budding into two or four, so that the characteristic cross-forms are produced. Nothing is known of the method of transmission.

Remarks.—The piroplasms of sheep and goats continue to be referred to *Piroplasma ovis* Laveran & Nicolle (1909), but Wenyon (1926) has restricted that name to the organisms of intermediate size, and named the larger form as *B. motasi* and the smaller form as *B. sergenti*. Thus *B. motasi*, *B. ovis*,



(From Wenyon, after Lestoquard.)

and B. sergenti of sheep and goats correspond to B. bigemina, B. bovis, and B. mutans of cattle. B. sergenti produces no recognizable symptoms.

Habitat.—Blood of goat, Capra hircus Linn.: UNITED PROVINCES, Muktesar.

#### 263. Babesla soricls (Christophers).

<sup>†</sup>*Piroplasma soricis*, Nuttall, 1908. *Babesia soricis*, Hoare, 1930, p. 246.

Parasites of moderate size, occupying one-third of the corpuscle in the larger forms. Did not show typical binary forms.

Remarks.—Nuttall (1908) stated that "Christophers has found a parasite in musk rats in India which he names P[iroplasma] soricis." Hoare (1930), failing to trace the original publication, referred to Christophers, who gave him the meagre information quoted above and remarked that he had found the parasite, but never described it or referred to it in print.

Habitat.—Red blood-corpuscles of the musk rat, Crocidura cærulea (Kerr): MADRAS, Guindy.

264. Babesla tropicus (Lingard & Jennings). (Fig. 158.)

†Piroplasma tropicus (part), Lingard & Jennings, 1904, pp. 161-5; Baldrey, 1910, pp. 572-7, pl. xxxix.

Small ring-forms, with central position of the chromatin in many of the infected red corpuscles. Chromatin in the form of two dots and a connecting bar between them. Pear-shaped forms rare in corpuscles, but occasionally seen free in the plasma.

Remarks.—Lingard and Jennings (1904) found piroplasms in bovines, buffaloes, equines, elephants, camels, goats, sheep,

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dogs, cats, monkeys, deer, fowls, guinea-pigs, and lizards, without referring them to different species. Baldrey (1911) not only referred a form found in the blood of cavalry and artillery horses at Mhow and Meerut to this species, but also thought that *B. gibsoni* (Patton) was identical with it. Lingard and Jennings, and Baldrey were wrong in referring the parasite from cattle, dogs, sheep, and horses to *P. tropicus* when *P. bovis*, *P. canis*, *P. ovis*, and *P. equi* were already known from those animals. Since then other species have also been described from these animals, and piroplasms of monkeys



Fig. 158.—Babesia tropicus (Lingard & Jennings). A, from camel; B, from guinea-pig; C, from elephant; D, from the common lizard. (After Lingard and Jennings.)

and deer have been described as P. pitheci and P. cervi. No one else seems to have described piroplasms from elephants, camels, fowls, guinea-pigs, and lizards, and for this residue the name P. tropicus may be retained till the parasites from these animals are examined again and the specific name restricted to one particular form.

Habitat.—Red blood-corpuscles of elephants, camels, guineapigs, fowls, and lizards : UNITED PROVINCES ; Bareilly. 265. Babesla sp.

†Piroplasma sp., Patton, 1910, p. 279; 1911, p. 620. Babesia sp., Wenyon, 1926, p. 1027.

Remarks.—The form may perhaps be the same as Babesia cervi Bettencourt, Franca and Borges (1907) described from Cervus dama Robert of Portugal.

Habitat.-Blood of the spotted deer, Axis axis (Erxl.) (=Cervus axis) : INDIA.

266. Babesia sp.

Babesia sp., Plimmer, 1915, p. 130; Wenyon, 1926, p. 1356.

Remarks.—Wenyon (1926) thinks that this form may possibly be the same as B. gibsoni. He saw the parasite in large numbers in the blood of a Colombo dog.

Habitat.—Blood of the wild dog, Cyon dukhunensis (Sykes), from INDIA, in the Zoological Gardens, London; CEYLON, Colombo.

267. Babesia sp.

†Piroplasma sp., Patton, 1910, p. 279; 1911, p. 620. Babesia sp., Wenyon, 1926, p. 1025.

Remarks.-The form may possibly be the same as Babesia herpestidis Franca (1908) from the mongoose, Herpestes ichneumon Illiger, in Portugal.

Habitat.-Blood of Herpestes edwardii Desmarest [=Herpestes mungo (Gmel.)].

## Doubtful Protozoa.

INCERTÆ SEDIS.

# Family ANAPLASMIDÆ Nietz, Alexander & du Toit. 1933.

#### Genus **GRAHAMELLA** Brumpt, 1911.

(Syn. BARTONELLA Strong, Tyzzer, Brues, Sellards, and Gastiaburu, 1915).

A new Hæmatozoan, Graham-Smith, 1905, pp. 453-9, pls. xili, xiv. Grahamella, Brumpt, 1911, pp. 514-17; 1913.

Graham-Smith bodies, Laveran & Marullaz, 1914, pp. 240-6, pl. ii. Grahamella, Carini, 1915, pp. 103-4.

Bartonella, Strong, Tyzzer, Brues, Sellards, & Gastiaburu, 1915,
 p. 32; Mayer, Borchardt, & Kikuth, 1926, p. 559; Wenyon,
 1926, pp. 1059-60; Noguchi & Battistini, 1926, p. 851; Noguchi,
 1926, pp. 533, 697, 715, 729.
 Grahamella, Wenyon, 1926, pp. 1056-9; Knowles, 1928, pp. 462-3;

Brumpt, 1928, pp. 1079-81.

Bartonella, McCarrison & Mula Singh, 1931, pp. 945-9.

Grahamia, Nietz, Alexander & du Toit, 1933, pp. 263-71.

Bartonella, Donatien & Lestoquard, 1934, pp. 650-2; Adler & Ellenbogen, 1934, pp. 219-21; Kikuth, 1934, pp. 1241-50. Lestoquard, 1934, pp. 650-2; Adler & Ellenbogen, 1934, pp. 219-

21.

(Bartonella), Topley & Wilson, 1936, pp. 710-11. Grahamella, Topley & Wilson, 1936, p. 712.

Parasites minute; rounded, oval or more usually rodlike bodies, showing an irregular staining, a blue cytoplasmic portion being distinguishable from a red chromatinic part. Rods may be straight or slightly curved, and forms with a red-staining granule at each end with a constriction in the middle suggest that reproduction is by binary fission. One, two or as many as fifty may infect the same red corpuscle or an endothelial cell.

Remarks.—Graham-Smith (1905) described these bodies in red blood-corpuscles of moles. Brumpt (1911), who encountered the same structures in the red blood-corpuscles of moles in France, described them as a new genus. Other observers have found similar structures in the red bloodcorpuseles of various other mammals. Wenyon (1926) thought that there was little evidence for regarding them as parasites, and still less that they were PROTOZOA. Structurally they resemble bacilli more than any other organisms. Knowles (1928) supported this view. Reichenow (1929), Kudo (1931), and Calkins (1933) have left them out of consideration, probably because they do not believe them to be PROTOZOA.

Very similar organisms were found in human red bloodcorpuscles and placed in a new genus called Bartonella. These are believed to be the causal agents of two diseases, namely, Oroya fever and Verruga peruviana, occurring in man in Peru. The former is a fever with marked anæmia, and the latter a nodular eruption of the skin. Strong, Tyzzer, Brues, Sellard, and Gastiaburu (1915) established that these diseases are distinct, and gave the name Bartonella bacilliformis to the organism associated with Oroya fever. Noguchi and Battistini (1926) succeeded in cultivating the organism in vitro. They also succeeded in inoculating the form into Macacus rhesus (Audeb.). Noguchi in a series of papers (1926, 1927) has further shown that on subpassage in monkeys the virulence of the organism was increased, and it produced more marked anæmia than at first. The organism was constantly present, as shown by culture, in the lymph glands, less often in the spleen, bone-marrow, and heart-blood. Inoculation of the monkey might produce either Oroya fever or Verruga peruviana, or both the conditions simultaneously. He also established by cross-immunity experiments that the two conditions were caused by the same virus. He further succeeded in transmitting SPOR. Y

the infection from monkey to monkey by the bites of the tick, Dermacentor andersoni.

Mayer (1925) described another species of the genus in mice, and Donatien and Lestoquard (1934) yet another in cattle.

Brumpt (1928) recalls in detail the successive creation of the genera Grahamella and Bartonella, and concludes that there is now insufficient cause for maintaining the genus Bartonella, created on the sole characteristic of pathogenicity. He regards Bartonella muris Mayer, 1921, as synonymous with Grahamella muris Carini, 1915, and combines under Grahamella Brumpt, 1911, all forms described under the genus Bartonella, which decision is followed here.

Nietz, Alexander, and du Toit (1933), however, regard the two genera as distinct, and change the name Grahamella to Grahamia, but their paper is not available to the author. Topley and Wilson (1936) also regard them as distinct, and remark that the evidence in favour of Bartonella being a living reproducible micro-organism, capable of giving rise under favourable conditions to disease, is now very strong, but considerably less is known about Grahamella, and it would be unwise to assert at the moment that these bodies are definite bacteria. According to them, Grahamella, though resembling Bartonella, are much coarser, and more like ordinary bacteria. With Giemsa's stain they take on a blue rather than a reddish tint. Only occasional red cells are affected. Grahamella appear to be non-pathogenic and are not influenced by splenectomy. They have not so far been cultivated, and though it is probable that they are living organisms, the evidence in favour of this is not vet conclusive.

## 268. Grahamella canis (Kikuth).

Bartonella canis, Kikuth, 1928, p. 1729; 1929, pp. 1-7; Topley & Wilson, 1936, p. 711.

†Bartonella canis, Ray & Idnani, 1937, p. 371; 1938, p. 259.

Forms in the red blood-corpuscles very pleomorphic, large or small, coccoid or rod-shaped.

*Remarks.*—Kikuth (1928) described this form and considered it responsible for causing infectious anæmia of dogs. He was unsuccessful in cultivating the organism on artificial media. Ray and Idnani (1937) have recently recorded the occurrence of this parasite in dogs in India, and believe it to be the cause of one of the types of obscure canine fever. Appearance of the parasite in the blood was marked by a distinct thermal reaction associated with a number of pathological changes. Kikuth (1927) and others have stated that *Bartonella* usually appears in the blood of splenectomized dogs, but Ray and Idnani (1938) find the infection to be inoculable into healthy non-splenectomized dogs, producing acute symptoms and ultimate death of the host in several cases.

Habitat.—Blood of the dog, Canis familiaris Linn. : UNITED PROVINCES, Muktesar.

## 269. Grahamella muris Carini.

Grahamella muris, Carini, 1915, pp. 103-4. Bartonella muris, Mayer, 1925; Mayer, Borchardt, & Kikuth, 1926, p. 559; Wenyon, 1926, p. 1060. †Grahamella sp., Knowles, 1928, p. 463. †Bartonella muris, McCarrison & Mula Singh, 1931, pp. 945-9.

Dot- and rod-like bodies in the red blood-corpuscles of mice. Remarks.—Mayer (1925) described this organism from the red blood-corpuscles of mice which had been treated with Bayer 205 for trypanosome infections. Later, Mayer, Borchardt, and Kikuth (1926) showed that the same bodies appear in anæmia caused by splenectomy in rats. They believed that the operation had stimulated a latent infection. McCarrison (1927) and Wills (1930) showed that the same type of anæmia may arise in a proportion of non-splenectomized rats when they are fed on a diet deficient in fat-soluble vitamins and vitamin C. The faulty food would thus appear to induce functional injury to the spleen, thereby lowering resistance to infection, in a manner comparable to, though not so effective as, splenectomy. McCarrison and Mula Singh (1931) have further shown that the blood of one- to four-day-old suckling rats born of splenectomized or non-splenectomized mothers may also show the infection.

Knowles (1928) recorded *Grahamella* in certain films of rats' blood prepared by Parmanand of Bombay. Up to eight forms per red blood-corpuscle were encountered, while a very few lay apparently free and extra-corpuscular.

Habitat.--Blood of rat : MADRAS, Madras; BOMBAY, Bombay.

## 270. Grahamella sp. (Fig. 159.)

"Corps Bartonelliformes," de Mello, 1928, pp. 515-16, fig. B, 29-38.

Intra-corpuscular bodies, bipolar, navicular, fusiform or



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annular, the chromatin being concentrated into an excentric nucleus or at the two poles.

Remarks.—De Mello regards these bodies of doubtful nature, and intermediate between Grahamella and Paraplasma.

Habitat.—Blood of the ant-eater, Manis pentadactyla Linn. : PORTUGUESE INDIA, Nova Goa.

## Genus PARAPLASMA Seidelin, 1912.

"Seidelin bodies," Seidelin, 1911, pp. 282-8.

- Paraplasma, Seidelin, 1912, pp. 503-4; 1914 a, pp. 203-8; Wenyon & Low, 1914, pp. 369-72; 1915, pp. 55-6; Seidelin, 1915, pp. 38-40; Seidelin & Connal, 1915, pp. 427-48.
- "Šeidelin bodies," Thompson, 1915, pp. 479-82; Fowler, Simpson, Ross, Leishman, & Balfour, 1915, 3rd report; Cropper & Drew, 1916, pp. 20-4; de Mello, 1917, pp. 21-4.

Polymorphic intra-corpuscular bodies, ranging from a small chromatic granule to large bodies consisting of a chromatic granule with a shaft of cytoplasm.

*Remarks.*—Seidelin, who first described these bodies, considered them to be the cansative agent of yellow fever, and named the organism *Paraplasma flavigenum*. Seidelin and Connal (1915) considered the organism to be capable of transmission by artificial infection, and claimed to have discovered naturally infected guinea-pigs in West Africa, which were supposed to be reservoirs of the disease. Wenyon and Low (1914, 1915) refuted the parasitic view and showed that similar bodies were found in guinea-pigs in England, and especially in young animals. Finally, this latter view was supported by the Yellow Fever Commission, who reported (1915) that *P. flavigenum* was not an organism, and was not connected with yellow fever. Cropper and Drew (1916) reported the occurrence of similar bodies in anæmic fœtal blood.

## 271. Paraplasma sp. (Fig. 160.)

†" Seidelin bodies" (?), de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, pp. 21–4, pl. ii, figs. 1–20.

Circular, vacualated bodies occurring in groups of two, three or four in red blood-corpuscles, or in the form of thin or thick bacilli or vibrios.



Fig. 160.—Paraplasma sp. (After de Mello and others.)

Remarks.—De Mello found these bodies in the blood-corpuscles of a man who had never been outside India, but it is doubtful if the so-called "Seidelin bodies" are an independent organism. Habitat.-Blood of man : PORTUGUESE INDIA, Nova Goa.

## Genus ANAPLASMA Theiler, 1910.

"Marginal points," Smith & Kilborne, 1893, p. 177.
Anaplasma, Theiler, 1910, pp. 135-7; 1912 a, pp. 319-38; 1912 b, pp. 105-16; 1912 c, pp. 193-207; Minchin, 1912, p. 383; Dias & Aragão, 1914, pp. 231-49; Carpano, 1914, pp. 42-53; Lignières, 1914, pp. 133-62; 1919 a, pp. 558-66; 1919 b, pp. 568-66; 1910 b, pp. 641-51: 1919 c, pp. 774-9; Hegner & Taliaferro, 1924, pp. 305-6; Wenyon, 1926, pp. 1053-6; Knowles, 1928, pp. 466-7.

Spherical body of chromatin less than  $0.5\mu$  in diameter, situated at or near the edge or towards the centre of the red blood-corpuscles. Infection transmitted by a tick.

Remarks.—Smith and Kilborne (1893) in their studies on Texas fever of cattle recognized certain red-staining granules on the margin of red corpuscles ("marginal points") and regarded them as a resistant phase of Babesia bigemina. Theiler (1910) recognized similar bodies in the blood of cattle in South Africa, and referred them to Anaplasma, a new genus of parasitic PROTOZOA. The bodies are commonly found in association with infections of B. bigemina and B. mutans, and Carpano (1914) considered them as resistant forms of Piroplasmata. Dias and Aragão (1914) believed that they are not true organisms at all, but a hæmolytic product. Theiler (1912) and Lignières (1919) have described cases of pure infection with these bodies. According to the latter author, in true Anaplasma infections as many as 50 per cent. of the red corpuscles may be infected, and the infection is readily transmissible to clean cattle. Infected cattle suffer from fever and an intense and progressive anæmia. Laveran and Franchini (1914) discovered them in numerous Mammals, such as rats, mice, rabbits, guinea-pigs, moles, cats, dogs, calves, pigs, and monkeys. De Mello and his colleagues (1917) found them in human blood, and also in the blood of fishes, frogs, lizards, snakes, and birds, but did not regard them as **PROTOZOA**, considering them to be hæmolytic products.

## 272. Anasplasma sp. (Fig. 161.)

†Anaplasma, de Mello, de Sá, de Sousa, Dias, & Noronha, 1917, pp. 17-21, pl. ii, figs. 21-9.

Intra-corpuscular, oval or lanceolate, of small dimensions, and staining like chromatin; apparently not possessing any cytoplasm.

**Remarks.**—De Mello and his colleagues (1917) found Anaplasmosis in the blood of fishes, frogs, lizards, snakes, tortoises, and man. Discussing the origin of the so-called *Anaplasma*, they came to the conclusion that they are not PROTOZOA, but are unquestionably due to hæmolytic alterations. They regarded them as due to nuclear fragmentation,



Fig. 161.—Anaplasma sp. (After de Mello and others.)

which is normal in Vertebrates with nucleated red corpuscles, or pathological, as in anæmias of Mammals.

Habitat.—Blood of Perca fluviatilis Linn., Rana tigrina Daud., Rana esculenta Linn., Hemidactylus brooki Gray, Coluber blumenbachii Schlegel, Naja naja Linn., Chitra indica (Gray), and Homo sapiens Erxleben : PORTUGUESE INDIA, Nova Goa.

#### 273. Anaplasma sp.

†Anaplasma sp., Knowles, 1928, p. 467.

Habitat.-Blood of Indian bats : BENGAL, Calcutta.

## Genus BERTARELLIA Carini, 1930.

Bertarellia, Carini, 1930, pp. 1312–13; de Mello & de Meyrelles, 1937 b, pp. 98–108.

Polymorphic intra-corpuscular bodies, probably of a parasitic nature; usually round or oval, variable in size, smallest of the size of cocci and the largest  $1-2\mu$  in diameter. Stained with Leishman's or Giemsa's stains, in the largest individuals the cytoplasm is stained blue, usually with a chromatic granule stained red. The infected red blood-corpuscles are not altered.

## 274. Bertarellia calotis de Mello & de Meyrelles. (Fig. 162.)

†Bertarellia calotis, de Mello & de Meyrelles, 1937 b, pp. 98-108, pl. xii, 2 text-figs.

Circular bodies, with refringent greenish cytoplasm surrounded by a strong membrane, intra-corpuscular or free, dividing by a process of budding. Stained by Leishman's or Giemsa's stains they show a variety of forms; they appear either as a small round chromatic dot, of anaplasmoid nature,

#### BERTARELLIA.

or like a vacuole devoid of any central granule or a small roundish body surrounded by a more or less strong membrane, taking a chromatic stain, with a central nuclear granule. The cytoplasm is stained blue or greyish-blue, often surrounded by a white circular halo, without any granule at all or with a central chromatic granule or the nuclear mass may be located at the periphery attached to the membrane. The infected red blood-corpuscle may have two or three of these parasites, and does not show any alteration.



Fig. 162.—Bertarellia calotis de Mello & de Meyrelles. A, anaplasmoid ; B, surrounded by a vacuole ; C, cytoplasm surrounded by a strong membrane ; D, cytoplasm containing a central chromatic granule ; E, cytoplasm with peripheral chromatic mass : F, with central and peripheral chromatic granules ; G, chromatic granule showing budding. (After de Mello and de Meyrelles.)

*Remarks.*—Blood-smears fixed with Bouin's or Schaudinn's fluid and stained with Heidenhain's iron-hæmatoxylin are said to have confirmed the structure as revealed by Romanowsky's stain, and have thrown light on the process of division. The chromatic granule or nucleus becomes enlarged and takes a ring form. Later it becomes compact and gives origin to a small bud, which protrudes to the exterior, becomes covered by membrane and separated from the main cell.

Habitat.—Blood-corpuscles of Calotes versicolor Daud. subspecies major Blyth : PORTUGUESE INDIA, Nova Goa.

## 275. Bertarellia sp.

†Bertarellia sp., de Mello & de Meyrelles, 1937 b, p. 107.

Habitat.—Blood of the Indian tortoise, Lissemys punctata granosa (Schoepff): PORTUGUESE INDIA, Nova Goa.

# II. Subclass CNIDOSPORIDIA Doflein, 1901.

This subclass includes organisms which are amœboid during the trophic phase, and of which dissemination takes place through resistant spores, which are provided with one to four polar capsules. Each spore contains one to many sporoplasms or generative cells, and the spore-membrane may be complete



Fig. 163.—Some typical CNIDOSFORDIA and their spores. A, Chloromyxum leydigi Mingazzini; B, C, Leptotheca agilis Thélohan in different stages of locomotion; D, spore of the same; E, spore of Henneguya psorospermica Thélohan, stained section. ect., ectoplasm; end., endoplasm; f.g., fatglobules; n., nuclei; p.c., polar capsules; ps., pseudopodia; s., sutural plane; sp., spores; st.ps., stylar pseudopodium; v., vacuole. (From Reichenow, after Doffein and Thélohan.)

or be bivalved or trivalved. Each polar capsule contains a long coiled filament, which, when extruded, serves to attach the spore to the intestinal wall till the amœboid body can escape from the spore and infect the tissues of the new host.

Schaudinn (1900) divided the SPOROZOA into two subclasses, the TELOSPORIDIA and the NEOSPORIDIA. The TELO-SPORIDIA include the GREGARINIDA, COCCIDIA, and H $\not\equiv$ mo-SPORIDIA, and are a uniform group, characterized by having

definite intracellular stages, schizogony, and, following upon conjugation of gametes, formation of occysts within which the sporozoites are formed. The NEOSPORIDIA, including the CNIDOSPORIDIA, SARCOSPORIDIA, and HAPLOSPORIDIA, do not form a natural group, nor have they much in common with the TELOSPORIDIA. In the NEOSPORIDIA the life of an individual does not come to an end when reproduction takes place, but reproduction continues throughout the trophic phase, the sporoblasts being carried about by the more or less active organism, which may ultimately become a large mass of spores. Among the CNIDOSPORIDIA the parasites may sometimes be intracellular, but they usually reproduce by binary fission. and the zvgotes do not become encysted and do not produce sporozoites. The SARCOSPORIDIA and HAPLOSPORIDIA possess simple spores, and do not seem to be related either to the CNIDOSPORIDIA or to the TELOSPORIDIA.

Wenyon (1926) considers it justifiable to reserve the title SPOROZOA for the TELOSPORIDIA, to consider the CNIDOSPORIDIA as an independent class, and to regard SARCOSPORIDIA and HAPLOSPORIDIA as parasites of undetermined position. Reichenow (1929, 1935) regards TELOSPORIDIA, CNIDO-SPORIDIA, SARCOSPORIDIA, and HAPLOSPORIDIA as independent subclasses of SPOROZOA. Kudo (1931) also follows this arrangement, but places SARCOSPORIDIA and HAPLOSPORIDIA in one subclass with the title ACNIDOSPORIDIA. Calkins (1933) regards TELOSPORIDIA, CNIDOSPORIDIA, and ACNIDOSPORIDIA as classes of the subphylum SPOROZOA.

CNIDOSPORIDIA are exclusively parasites of the Invertebrates and the lower Vertebrates, and are responsible for causing epidemics of infection among animals of economic importance such as fishes, silkworms, and honey-bees. There are no secondary or intermediate hosts.

Following Kudo, the CNIDOSPORIDIA are divided into four orders, as follows :---

1.	Spore large; membrane bivalved; two or	[Bütschli, p. 330.
	four polar capsules visible in vivo	Myxosporldia
<b>2</b> .	Spore large; membrane trivalved; three dis-	[Stole, p. 346.
	tinctly visible polar capsules	Actinomyxidia
3.	Spore small; membrane in one piece; one	[Balbiani, p. 346.
	(rarely two) polar filaments; invisible in vivo.	Microsporidia
4.	Spore small, barrel-shaped; a thick filament	-
	coiled beneath the spore-membrane; three	[Kudo, p. 360.
	sporoplasms	Hellcosporidia

# I. Order MYXOSPORIDIA Bütschli, 1881.

The MYXOSPORIDIA are distinguished by their characteristic spores, which are of various shapes and dimensions. Each spore is covered by a bivalved chitinous membrane called the spore-membrane, the two valves of which are united in a sutural plane, which may be more or less straight or irregularly



Fig. 164.—Some characteristic forms of spores of various MXXOSPORIDIA. A, Ceratomyxa truncata Thélohan; B, Chloromyxum leydigi Mingazzini; C, Sphærospora rostrata Thélohan; D, Myxidium procerum Auerbach; E, Zschokkella acheilognathi Kudo; F, Myxobolus carassii Klokačewa; G, Henneguya gurleyi Kudo. (×1500.) (After Kudo.)

curved. The surface of the valves may be smooth or marked with ridges, and the form of the spore depends upon the shape of the valves and the presence of accessory appendages.

Within the shell are polar capsules, which may be one, two or four in number and are usually situated at what is described as the anterior end of the spore. In the family Myxidiidæ there are two polar capsules, one near each pole of the spore. Each polar capsule contains a coiled filament, which can be extruded through its pore. The substance contained in the spore, apart from the polar capsules, is designated the sporoplasm. It usually contains two nuclei, and in the family Myxobolidæ an iodinophilous vacuole containing glycogenous substance which stains mahoganyred with iodine.

When introduced into the digestive tract of a fish, the sporoplasm leaves the spore as amœboid "planonts" or wanderers. These probably fuse together and grow into the characteristic trophic phase, the so-called multinucleate plasmodium. Passing through the epithelium of the gut, it enters the tissues of specified organs, grows into a schizont, and its nucleus divides repeatedly. Some of the nuclei become surrounded by cytoplasm and form sporonts. The sporont may be monosporous, disporous or polysporous, according as it produces one, two or many spores.



Fig. 165.—Development of the spore from the pansporoblast in Myxobolus pfeifferi Thélohan. A, single propagative cell from multinucleate plasmodium; B, division to form one large and one small cell; C, association of two pairs to form a group of two large and two small cells; D, formation of sixcelled stage; E, stage with fourteen nuclei, two of which are the nuclei of the original small cells; F, division into two bodies, each with six nuclei, while the nuclei of the small cells take up a position at the angles between them; G, each sporoblast now divides into three cells, two of which, each with a single nucleus and a vacuole, are the capsulogenous cells containing the polar capsules, one with two nuclei forms the binucleate amœbula, while the remaining two nuclei become peripherally arranged and form, together with some cytoplasm, the valves of the spore; I, J fully developed stage of one of the developing spores; I, J fully Sporulation begins with a peculiar process of endogenous or internal budding. An island of protoplasm or pansporoblast is formed round two of the nuclei, which are usually dimorphic. Each of these nuclei undergoes division until fourteen are present, seven from each of the original nuclei. The bud then divides into two cells, each of which is a sporoblast containing six nuclei, after one has been extruded. Two of these six nuclei form the valves of the capsule (sporocyst), two form the polar capsules, and two remain as pronuclei, which subsequently fuse when the spore becomes mature. Thus the endogenous bud represents a zygote, and the two original nuclei of the pansporoblast the progametic nuclei. The details of the process of sporulation



Fig. 166.—External appearance of Myxosporidian infection in fish. A, head of the short-headed red-horse, showing the cysts of Myxobolus conspicuus Kudo,  $\times \frac{1}{2}$ ; B, the river chub, with cysts of Myxobolus squamosus Kudo,  $\times \frac{3}{4}$ ; C, a bluntnosed minnow, with numerous cysts of Myxobolus aureatus Ward on the fins,  $\times \frac{2}{3}$ . (After Kudo.)

differ in different species. Neville (1931) has reviewed the previous literature and made an independent study of five different species. He concludes that there are two types of nuclei, germinal and vegetative. The germinal nuclei contain the diploid number of chromosomes, but after several divisions undergo reduction, whereby the number of chromosomes is reduced to one-half.

The simpler members of the order occur in various cavities in the bodies of their host, such as the gall-bladder, uriniferous tubules of the kidney or urinary bladder, and do not cause much harm to the host. In other cases the parasites invade the tissues, causing them to degenerate, and give rise to tumours in muscles, central nervous system, etc., which are referred to as Myxosporidian cysts and are visible to the naked eye, or the parasites may spread through tissue, giving rise to diffuse infiltration.

The MYXOSPORIDIA are parasites of cold-blooded Vertebrates, more particularly fishes. Each species attacks one or several species of fish. In the case of freshwater fishes they usually invade the gills and the gall-bladder, while in the marine fishes they are usually found in the gall-bladder or the urinary bladder. Whitish pustules visible to the naked eye may indicate the infection in the gills, and in the case of internal organs there may be abnormal changes of form or colour, but ordinarily infection can only be detected by microscopical examination.

Doflein (1901) divided the order into two suborders, DIS-POREA and POLYSPOREA, but Kudo (1920) considers such a division an artificial one, as the number of spores produced by any species is not always constant.

Following Kudo (1920, 1933) the MYXOSPORIDIA are divided into three suborders, as follows .---

1.	Largest diameter of the spore at right angles to the sutural plane; one polar capsule on each side of the plane; sporoplasm without iodinophilons vacuole	[Kudo, p. 333. Eurysporea,
2.	Spore spherical or subspherical, with one, two or four polar capsules : sporoplasm without	[Kudo, p. 335.
	iodinophilous vacuole	Sphærosporea,
3.	Sutural plane of the spore coincides or forms an acute angle with the longest diameter; one	
	or two polar capsules; sporoplasm with or without an iodinophilous vacuole	[Kudo, p. 336. Platysporea,

# I. Suborder EURYSPOREA Kudo, 1920.

The largest diameter of the spore is at right angles to the sutural plane, with one polar capsule on each side of the plane. Sporoplasm without iodinophilous vacuole. Vegetative form found in body-cavity. The great majority are parasites of marine Fish. Monosporous, disporous or polysporous.

## Identification Table of Families.

1.	Typically cœlozoic, in marine Fish	Ceratomyxidæ Doflein, p. 334.
2.	Histozoic or cœlozoic, in fresh-	
	water Fish	Wardiidæ * Kudo.

# Family CERATOMYXIDÆ Doflein, 1899.

Typically cœlozoic parasites of marine Fish.

## Genus CERATOMYXA Thélohan, 1892, emend.

Ceratomyza, Thélohan, 1892, pp. 169, 171, 175; Labbé, 1899, pp. 89–91; Minchin, 1903, pp. 289, 293, 295, 298; 1912, p. 408; Kudo, 1920, pp. 61–80; Wenyon, 1926, p. 725; Knowles, 1928, p. 324; Roichenow, 1929, pp. 1953–7; Kudo, 1931, p. 306; 1933, pp. 196, 199–201; Calkins, 1933, p. 567; Reichenow, 1935, p. 386.

Spore arched, breadth more than twice the sutural diameter. Valves of spore extended into long, lateral, conical, and hollow processes. Sporoplasm asymmetrically placed, usually not filling the intrasporal cavity. Monosporous, disporous or polysporous. Generally in the gall-bladder or urinary bladder of marine Fish.

## Key to Indian Species.

1 (2). Spore crescent-shaped,  $14-15\mu$  by  $4-5\mu$ . C. gobioidesi Chak., [p. 334. 2 (1). Spore elliptical,  $30-40\mu$  by  $10\mu$  ..... C. hilsæ Chak., p. 335.

## 276. Ceratomyxa gobioidesi Chakravarty.

†Ceratomyxa sp., Ray, 1933 a, p. 259; 1933 b, p. 349. †Ceratomyxa gobioidesi, Chakravarty, 1938.

Trophozoites more or less spherical, not showing sharp demarcation between ectoplasm and endoplasm, pseudopodia short and blunt. Disporous; spores crescent-shaped, valves symmetrical and terminating in blunt points, sutural plane distinct. Extra-capsular cavity filled with finely granular sporoplasm. Polar capsules equal and spherical, situated on each side of the sutural plane, and provided with distinct coiled filaments.

Dimensions.—Trophozoites  $500-650 \mu$  in diameter; spores  $14-15 \mu$  in breadth,  $4-5 \mu$  in sutural diameter; polar capsules  $2 \cdot 5 - 3 \mu$ ; polar filament  $15 \mu$ .

*Remarks.*—Ray (1933 a, b) reported this species from a number of estuarine fish, but did not publish any description. Chakravarti (1938) has recently restudied the form and described it as a new species. He has kindly allowed me to see the manuscript of his paper prior to its publication, and to abstract the description of this and several other species.

Habitat.—Liver, gall-bladder, kidney, ovary, etc., of Gobioides rubicundus Hamilton; gall-bladder of Trichogaster fasciatus Schneider and Macrones gulio (Ham.-Buch.): BENGAL, Calcutta.

## 277. Ceratomyxa hilsæ Chakravarty.

†Certaomyxa hilsæ, Chakravarty, 1938.

Trophozoites in large numbers in the gall-bladder of the host. Disporous, spores often seen lying side by side in a common envelope. Spores elliptical, valves tapering to blunt ends, sutural plane prominent, dividing the spore into equal parts. Extra-capsular cavity filled with finely granular sporoplasm. Polar capsules spherical, of equal sixe, provided with well-marked coiled polar filaments.

Dimensions.—Spores  $\hat{3}0-40\,\mu$  in breadth,  $10\,\mu$  in sutural diameter; polar capsules  $5\,\mu$  in diameter; polar filament  $35-40\,\mu$ .

*Remarks.*—The above description is based on Chakravarty's paper in manuscript.

Habitat.—Gall-bladder of Hilsa ilisha (Ham.-Buch.): BENGAL, Calcutta.

# II. Suborder SPHÆROSPOREA Kudo, 1920, emend.

Spore spherical, with one, two or four polar capsules. Sporoplasm without iodinophilous vacuole. Vegetative form found in body-cavity and tissues. Monosporous, disporous or polysporous. Parasites of marine and freshwater Fish and Amphibia.

Identification Table of Families.

1.	With four polar capsules	Chloromyxldæ Thélohan,
2.	With two polar capsules	Sphærosporidæ Davis,
3.	With one polar capsule	IP. 336. Unicapsuiidæ * Kudo.

## 1. Family CHLOROMYXIDÆ Thélohan, 1892.

Spore with four polar capsules. Monosporous, disporous or polysporous.

## Genus CHLOROMYXUM Mingazzini, 1890.

Chloromyxum, Mingazzini, 1890, p. 160; Labbé, 1899, pp. 94-6;
Minchin, 1903, p. 295; 1912, p. 409; Kudo, 1920, pp. 87-99;
Wenyon, 1926, p. 726; Reichenow, 1929, pp. 1058-9; Kudo, 1931, p. 308; 1933, pp. 197, 202-3; Calkins, 1933, p. 568;
Ray, 1933, p. 259; Reichenow, 1935, p. 386.

Spore with four polar capsules, grouped at the anterior end; surface often striated or with ridges; sutural line often obscure. Monosporous, disporous or polysporous. Histozoic, or cœlozoic in freshwater and marine Fish and also in Amphibians.

## 278. Chloromyxum amphipnovi Ray.

†Chloromyxum amphipnovi, Ray, 1933 a, p. 259. †Chloromyxum sp., Ray, 1933 b, p. 349.

This parasite was reported by Ray (1933) as a new species, but no account of it has as yet been published.

Habitat —Gall-bladder of Amphinous kuchia (Ham.-Buch): BENGAL, Calcutta

# 2. Family SPHÆROSPORIDÆ, Davis, 1917.

Spore with two polar capsules. Monosporous, disporous or polysporous.

## Genus SPHÆROSPORA Thélohan, 1892.

Sphærospora, Thélohan, 1892, p. 167; Labbé, 1899, p. 86; Minchin, 1903, pp. 286, 293, 295, 298; Kudo, 1920, pp. 100-4; Wenyon, 1926, p. 726; Reichenow, 1929, p. 1059; Kudo, 1931, p. 308; 1933, pp. 197, 203; Calkins, 1933, p. 568; Reichenow, 1935, p. 387.

Spore with two polar capsules at anterior end. Sutural line straight. Monosporous, disporous, or polysporous. Bodycavity and tissue parasites of freshwater and marine Fish.

## 279. Sphærospora sp.

\*Sphærospora sp., Southwell & Prashad, 1918, pp. 347-8. Sphærospora sp., Kudo, 1920, p. 103 ; 1933, p. 203.

*Remarks.*—The poor condition of the material did not allow of a complete account of its structure being given, but the bicapsulate, rounded structure of the spore places it in this genus.

Habitat.—Cysts occurred in very large numbers, one under each scale of *Barilius barna* (Ham.-Buch.), from the vicinity of the Ruby Mines : BURMA

# III. Suborder PLATYSPOREA Kudo, 1920.

Sutural plane coincides or forms an acute angle with the longest diameter. One, two or four polar capsules. Sporoplasm with or without an iodinophilous vacuole.

## Identification Table of Families.

1 (5).	Without iodinophilous vacuole.	2-4.
2.	Two polar capsules, one at each	
	pole	Myxidiidæ Thélohan, p. 337.
3.	One polar capsule	Coccomyxidæ * Léger & Hesse.
4.	Two or four polar capsules at	
	anterior end	Myxosomatidæ * Poche.
5 (1).	With an iodinophilous vacuole .	Myxobolidæ Thélohan, p. 340.

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# 1. Family MYXIDIIDÆ Thélohan, 1892.

Spore fusiform or semicircular. One polar capsule at each end. Sporoplasm without iodinophilous vacuole.

#### Key to Indian Genera.

1.	Polar filaments long and fine	2.
2 (3).	Spores fusiform, with pointed ends;	[p. 337.
	polar capsules oppositely directed	MYXIDIUM Bütschli,
3 (2).	Spores fusiform, usually with truncated	[bach, p. 339.
	ends; polar capsules obliquely directed.	ZSCHOKKELLA Auer-

## Genus MYXIDIUM Bütschli, 1882.

Myxidium, Bütschli, 1882, p. 593.

Cystodiscus, Lutz, 1889, pp. 84-8.

Myxidium, Labbé, 1899, pp. 91-2; Minchin, 1903, pp. 286, 288, 295; 1912, p. 409; Kudo, 1920, pp. 108-18; Wenyon, 1926, p. 726; Reichenow, 1929, pp. 1060-2; Kudo, 1931, p. 309; 1933, pp. 197, 204-6; Calkins, 1933, p. 568; Reichenow, 1935, p. 387.

Spore fusiform, with pointed or rounded ends. Polar capsules typically pyriform. Polar filament comparatively long and fine. Monosporous, disporous or polysporous. Typically cœlozoic, but also histozoic in marine or freshwater Fish, Amphibians or Reptiles.

## Key to Indian Species.

1	(2).	Spore elongated, fusiform, $10-12\mu$ in	[p. 337.
		length	M. danilewskyi Lav.,
2	(1).	Spore not fusiform	3.
3	(4).	Spore oval, tapering at each end to	[p. 338.
		a blunt point, $16\mu$ by $5\mu$	M. mackiei Bosan.,
4	(3).	Spore ovoidal, $12-15\mu$ by $8\cdot 5-10\mu$	M. glossogobii Chak.,
			[p. 337.

#### 280. Myxidium danilewskyi Laveran.

Myxidium danilewskyi, Laveran, 1897, p. 725; 1898, pp. 27-30, 30 figs.; Labbé, 1899, p. 92.

†Myxidium danilewskyi, Laveran & Mesnil, 1902, p. 609.

Myxidium danilewskyi, Minchin, 1903, pp. 275, 294, 346; Kudo, 1920, pp. 109-10, figs. 255-7; 1933, p. 204; Reichenow, 1935, p. 387.

Vegetative phase not known. Disporous. Spores elongated, fusiform, sometimes a little incurved. Polar capsules small, situated at the extremities.

Dimensions.—Length of spore  $10\mu$ .

Habitat.—From the kidney of the tortoise, Chinemys reevesi (Gray): CEYLON.

## 281. Myxidium glossogobil Chakravarty.

†Myxidium glossogobii, Chakravarty, 1938.

Vegetative form not observed. Spores elongate oval, spor. z

#### SPOROZOA.

valves not striated. Space outside the capsule filled entirely with uniformly granular sporoplasm, containing several black dots in the fresh condition. Polar capsules one at each end of the spore, spherical at first, but ovoidal after the extrusion of the polar filament. Openings through which filaments are extruded are marked by elevated areas of the shell just in front of the polar capsules.

Dimensions. Spore  $12-15\mu$  in length,  $8.5-10\mu$  in breadth; polar capsules  $3 \cdot 1 - 4 \cdot 1 \mu$ ; polar filament  $40 - 50 \mu$ . Remarks.—Description is based on Chakravarty's paper in

manuscript.

Habitat.—Gall-bladder of Glossogobius giuris (Ham.-Buch.): BENOAL, Calcutta.

#### 282. Myxldium mackiei Bosanquet. (Fig. 167.)

†Myxidium mackiei, Bosanquet, 1910, pp. 436-8, figs. 7-13.

The vegetative phase does not ordinarily exhibit a distinction between ectoplasm and endoplasm; in a few cases where the



Fig. 167 .- Myxidium mackiei Bosanquet. A, vegetative phase in contact with cells of a renal tubule; B, spores. (After Bosanquet.)

whole organism was converted into spores there was a cuticle or cyst-wall. Cytoplasm contains a large number of nuclei, apparently of two varieties, some vesicular, others smaller and compact. Spores formed in pairs, at many points simultaneously, ultimately the whole mass being converted into spores. A portion of the cytoplasm becomes rounded off and lies in a definite space within the parasite. This portion

(disporoblast ?) contained in one instance 10 nuclei, but it seems likely that 12 is the full number of nuclei in the disporoblast, as the final sporoblasts into which this divides appear to contain usually 6 nuclei. Of the 6 nuclei of the sporoblast 2 go to form the valves of the sheath, 2 attach themselves to the polar capsules, and 2 remain in the sporoplasm. The two last-mentioned nuclei may fuse into one. In the sporoplasm two large vacuoles are often seen, one near each polar capsule. Both ends of a spore taper to a blunt point and the spore-coat is markedly striated.

Dimensions.—Spores average  $16 \mu$  in length by  $5 \mu$  in breadth.

*Remarks.*—The description is based on sections of the kidney of the host prepared by J. Percival Mackie in the Bombay Bacteriological Laboratory.

Habitat.—Kidney of the tortoise, Trionyx gangeticus Cuvier : BOMBAY, Bombay.

#### 283. Myxidium sp.

†Myxidium sp., Ray, 1933 b, p. 349.

Habitat.—Gall-bladder of Clarias batrachus Cuvier & Valenc., Saccobranchus fossilis (Bloch.); Ophiocephalus punctatus Bloch: BENGAL, Calcutta; gall-bladder of Kachuga smithi (Gray), Lissemys punctata granosa (Schoepff): UNITED PROVINCES, Allahabad, and Geomyda trijuga (Schweigger): MADRAS, Madras.

## Genus ZSCHOKKELLA Auerbach, 1910.

Zschokkella, Auerbach, 1910; Kudo, 1920, pp. 122-3; Wenyon, 1926, p. 727; Reichenow, 1929, p. 1062; Kudo, 1931, p. 310; 1933, pp. 198, 206; Calkins, 1933, p. 568; Reichenow, 1935, p. 387.

Spore semicircular in front view; ellipsoidal in profile; ends pointed. Sutural line much curved. Polar capsules large and spherical; polar filaments long and fine. Monosporous, disporous or polysporous. Typically cœlozoic parasites of fresh- and salt-water Fish.

## 284. Zschokkella prashadi Ray.

†Zschokkella prashadi, Ray, 1933 a, p. 259. †Cystodiscus (Zschokkella) sp., Ray, 1933 b, p. 349.

This parasite was reported by Ray (1933) as a new species, but no description has so far been published.

Habitat.—Gall-bladder of Bufo melanostictus Schneid., Rana tigrina Daud.: BENGAL, Calcutta; also gall-bladder of Lissemys punctata granosa (Schoepff): UNITED PROVINCES, Allahabad.
# 2. Family MYXOBOLIDÆ Thélohan, 1892.

Spore with one or two polar capsules at the anterior end, with or without posterior processes. Sporoplasm with an iodinophilous vacuole. Majority polysporous in freshwater Fish.

#### Key to Indian Genera.

1 (2).	Each valve of spore prolonged into a	[p. 344.
• •	long process	HENNEGUYA Thélohan,
2 (1).	Valves without posterior processes	3. [p. 340.
3 (4).	Spores ovoidal; two polar capsules	MYXOBOLUS Bütschli,
4 (3).	Spores pyriform ; one polar capsule	THELOHANELLUS Kudo,
		[p. 342.

#### Genus MXYOBOLUS Bütschli, 1882.

Myxobolus, Bütschli, 1882, pl. xxxviii, figs. 6-10; Labbé, 1899, pp. 97-101; Kudo, 1920, pp. 58, 128-58; Wenyon, 1926, p. 727; Reichenow, 1929, pp. 1668-72; Kudo, 1931, p. 312; 1933, pp. 198, 208-10; Calkins, 1933, p. 568; Reichenow, 1935, p. 388.

Spore ovoidal, flattened. Shell without posterior processes. Two pyriform polar capsules at the anterior end. Sporoplasm with an iodinophilous vacuole. This genus includes many species; the majority are polysporous and tissueparasites of freshwater Fish, but a few are known from marine Fish, an Annelid, and an Amphibian.

#### Key to Indian Species.

1 (4).	Spore with unequal polar capsules	2. р. 340.
2 (3).	Spores oval, $12.4 - 15\mu$ by $8.2 - 10\mu$	M. calbasui Chak.,
3 (2).	Spores spherical to oval, $7 \cdot 21 - 8 \cdot 24 \mu$ in	[p. 341.
	length	M. mrigalæ Chak.,
4 (1).	Spore with equal polar capsules	5. [& Prash., p. 341.
5 (6).	Spores ovoidal, $9\mu$ by $7\cdot 2\mu$	M. nodularis Southw.
6 (5).	Spores spherical, $13 \mu$ in diameter	M. sp., Southw.,
		[p. 342.

#### 285. Myxobolus calbasui Chakravarty.

†Myxobolus sp. (part), Ray, 1933 b, p. 349. †Myxobolus calbasui, Chakravarty, 1938.

Trophozoites spherical to oval. Polysporous. Spores oval, rounded at the posterior and pointed at the anterior end, sutural plane distinct. Sporoplasm occupying the posterior region of the spore. Polar capsules pyriform, one smaller than the other. Coiled polar filaments well marked, unequal in length when extruded. A spherical iodinophilous vacuole present.

Dimensions.—Spore  $12\cdot4-15\mu$  in length,  $8\cdot2-10\mu$  in breadth; polar capsules  $6\cdot18\mu$  by  $4\cdot12\mu$  and  $4\cdot12\mu$  by  $3\cdot09\mu$ ; polar filaments  $125\mu$  and  $60\mu$  respectively; iodinophilous vacuole  $4\cdot1\mu$  in diameter.

*Remarks.*—Description is based on Chakravarty's paper in manuscript.

Habitat.—Ovary and liver of Clarias batrachus Cuvier & Valenc.; gills of Katla katla (Ham.-Buch.); liver of Cirrhina mrigala (Ham.-Buch.); and gall-bladder of Labeo calbasu (Ham.-Buch.), Labeo rohita (Ham.-Buch.), and Cirrhina mrigala (Ham.-Buch.): BENGAL, Calcutta.

#### 286. Myxobolus mrigalæ Chakravarty.

†Myxobolus mrigalæ, Chakravarty, 1938.

Cysts oval, on the scales of the host, perforated, and containing large number of mature spores. Polysporous. Spores spherical to oval, valves thick, exhibiting several triangular markings in front view. Sporoplasm situated at the posterior end of the capsules. Polar capsules pyriform, unequal in size, divergent, and containing distinct coiled filaments. An iodinophilous vacuole is present.

Dimensions.—Cysts  $\cdot 75-1 \cdot 5$  mm. in length and  $\cdot 75-1$  mm. in breadth; spores  $7 \cdot 21-8 \cdot 24 \mu$ ; polar capsules  $5 \cdot 15 \mu$  by  $3 \cdot 09 \mu$  and  $3 \cdot 09 \mu$  by  $2 \cdot 06 \mu$ ; polar filaments  $41 \cdot 2 \mu$  and  $20 \cdot 6 \mu$ respectively.

*Remarks.*—Description is based on Chakravarty's paper in manuscript.

Habitat.—Scales of Cirrhina mrigala (Ham.-Buch.): BENGAL, Calcutta.

#### 287. Myxobolus nodularis Southwell & Prashad. (Fig. 168.)

†Myxobolus nodularis, Southwell & Prashad, 1918, p. 347, pl. xi, figs. 32-6.

Myxobolus nodularis, Kudo, 1920, p. 153; 1933, p. 209.

Cyst rounded or slightly elongated, creamy-yellow in colour. Spore ovoidal. Suture between valves thick. Two polar capsules. Polar filament very much coiled.



Fig. 168.—Spore of Myxobolus nodularis Southwell & Prashad. (After Southwell and Prashad.)

Dimensions.—Cysts from 3.5 to 3.8 mm. in length and 2.3-2.8 mm. in breadth; spores  $9\mu$  by  $7.2\mu$ ; polar capsules  $3.4\mu$  in length, filament  $18.3\mu$  in the extruded condition.

Habitat.—Muscles of Rasbora daniconius (Ham.-Buch.): BENGAL, Mirpur, Dacca.

#### 288. Myxobolus sp.

\*Myxobolus sp., Southwell, 1915, pp. 312-13, pl. xxvi, fig. 3. Myxobolus sp., Kudo, 1920, p. 151; 1933, p. 209.

Cyst soft, flattened, roughly oval or lenticular, milky-white in colour. Spore with two equal polar capsules, with a very short anterior tail-like process. Iodinophilous vacuole present.

Dimensions.—Cysts up to 1.1 mm; spores  $13\mu$  by  $13\mu$ ; polar capsule  $4\mu$  by  $4\mu$ .

Habitat.—Subcutaneous intermuscular tissue of Rasbora daniconius (Ham.-Buch.) from a stream near Katiwan: UNITED PROVINCES, Mirzapore.

#### Genus THELOHANELLUS Kudo, 1933.

Myxobolus (part), Bütschli, 1882, pl. xxxviii; Labbé, 1899, pp. 97–101; Kudo, 1920, p. 132; Wenyon, 1926, p. 727; Reichenow, 1929, pp. 1068-72; Kudo, 1931, p. 312; Calkins, 1933, p. 568.
 Thelohanellus, Kudo, 1933, pp. 198, 210.

Spore pyriform; flattened. One polar capsule at the anterior end. Sporoplasm with an iodinophilous vacuole. Histozoic in freshwater Fish.

#### Key to Indian Species.

1 (2).	Cyst oval to cylindrical; spore $30-32 \mu$	[Prashad), p. 342.
	by $7-8\mu$	T. rohitæ (Southwell &
2 (1).	Cyst elongated, ellipsoidal; spore	[Prashad), p. 344.
. ,	$13 \cdot 2 - 13 \cdot 6 \mu$ by $10 \cdot 1 - 10 \cdot 3 \mu \dots$	$T.\ seni$ (Southwell &

# 289. Thelohanellus rohitæ (Southwell & Prashad). (Fig. 169.) †Myxobolus rohitæ, Southwell & Prashad, 1918, pp. 344-7, pl. xi, figs. 1-27. Thelohanellus rohitæ, Kudo, 1920, p. 132; 1933, p. 210.

Cysts oval to cylindrical, with the long axis of the cyst parallel to the gill-filaments, creamy-yellow in colour. The wall of the cyst is formed of a vertically striated portion showing no nuclei, covered externally by a membrane, two to three layers thick, and lined internally by an endoplasmic layer showing a coarse granular structure and with scattered nuclei. In the endoplasm two kinds of nuclei are found, viz., the vegetative and the generative. The latter occur in rounded cells called pansporoblasts or propagative cells. They are rounded in shape, with a marginally situated nucleus, and vary in size from 6 to  $11\mu$ . The nucleus in the pansporoblast divides mitotically. Finally ten fully formed nuclei can be distinguished in the mother-pansporoblast, besides two nuclei for the pansporoblast mother-cell, and reduction nuclear chromatin particles lying free in the cytoplasm of the mother-cell. The pansporoblast ultimately divides into two daughter-cells or sporoblasts, each with five nuclei; two of these unite later to form the nucleus of the sporoplasm, one forms the nucleus of the polar capsule and the other two are for the spore-membrane. A fully formed spore is an elongated pear-shaped body, rounded posteriorly and acutely pointed anteriorly. The spore-wall consists of



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Fig. 169.—*Thelohanellus rohitæ* (Southwell & Prashad). A, gill with cysts; B, young spore; C, mature spore. (After Southwell and Prashad.)

two valves; the suture is a distinctly thickened ridge lying parallel to the long axis of the spore. There is only one polar capsule in each spore, with a long and much-coiled polar filament. An iodinophilous vacuole is present. *Dimensions.*—Cysts 3.1-3.8 mm. by 0.8-1.2 mm.; spores

Dimensions.—Cysts  $3\cdot1-3\cdot8$  mm. by  $0\cdot8-1\cdot2$  mm.; spores  $30-32 \mu$  by  $7-8 \mu$ ; polar capsule  $22-23 \mu$  in length.

Habitat.—Gills of Labeo rohita (Ham.-Buch.), from Turag River : BENGAL, Mirpur, Dacca.

### 290. Thelohanellus senl (Southwell & Prashad). (Fig. 170.)

*Myxobolus seni*, Southwell & Prashad, 1918, p. 347, pl. xi, figs. 28-31.

Thelohanellus seni, Kudo, 1920, pp. 132-3; 1933, p. 210.

Cyst elongated, ellipsoidal, whitish, with black scattered granules on the surface. Spore oval, pointed at the anterior end and much wider behind. Two valves, suture slightly thickened. Polar capsule single, with a much-coiled filament. Iodinophilous vacuole present.



Fig. 170.—Spore of *Thelohanellus seni* (Southwell & Prashad). (After Southwell and Prashad.)

Dimensions.—Cysts 4.7-5.4 mm. by 2.9-3.7 mm.; spores  $13.2-13.6\,\mu$  by  $10.1-10.3\,\mu$ ; polar capsule about  $4\,\mu$  in length, filament  $43\,\mu$  in the extruded condition.

Habitat.—On the median and caudal fins of Labeo rohita (Ham.-Buch.): BENGAL, Mirpur, Dacca.

#### Genus HENNEGUYA Thélohan, 1892.

Henneguya, Thélohan, 1892, pp. 167, 176; 1895, p. 352.
Myxobolus (part), Gurley, 1893, p. 418; Cohn. 1896, pp. 227-72.
Henneguya, Labbé, 1899, pp. 101-4; Minchin, 1903, pp. 286, 296; 1912, pp. 409, 426; Kudo, 1920, pp. 158-73, 198-9; Wenyon, 1926, p. 728; Reichenow, 1929, pp. 1059, 1072; Kudo, 1931, pp. 312-13; 1933, pp. 198, 210-12; Calkins, 1933, p. 568; Keichenow, 1935, p. 388.

Spores ovoidal; flattened. With a single or double caudal prolongation. Two pyriform polar capsules at the anterior end. Monosporous, disporous or polysporous. Sporoplasm contains an iodinophilous vacuole. Mostly histozoic (a few cœlozoic) parasites in freshwater Fish; one species in a marine Fish.

#### Key to Indian Species.

1 (2).	Cysts attached to the gill-filaments of	[p. 345.
	the host	H. ophiocephali Chak.,
2 (2).	Cysts in the bulbus arteriosus of the	[p. 345.
	host	H. otolithi Ganapati,

#### 291. Henneguya ophlocephali Chakravarty.

†Henneguya sp., Ray, 1933 b, p. 349.
†Henneguya ophiocephali, Chakravarty, 1938.

Cysts spherical, attached to the gill-filaments of the infected host. Polysporous. Spores ovoidal or oblong, with the anterior end rounded and broader and the posterior tapering and forming a tail which is bifurcated along its entire length. Polar capsules elongate, with anterior end pointed and provided with a coiled filament, one capsule slightly smaller than the other. An iodinophilous vacuole is present.

Dimensions.—Cysts about 2 mm. in diameter; spore  $16\cdot4-20\cdot5\,\mu$  in length,  $6\cdot15\,\mu$  in breadth, tail  $28-32\,\mu$ ; polar capsules  $6\cdot18\,\mu$  by  $2\cdot06\,\mu$  and  $5\cdot18\,\mu$  by  $2\cdot06\,\mu$  respectively; polar filaments  $26-32\,\mu$ .

Remarks.--Description is based on Chakravarty's paper in manuscript.

Habitat.—Gills and muscles of Ophiocephalus punctatus Bloch : BENGAL, Calcutta.

### 292. Henneguya otolithi Ganapati.

*†Henneguya* sp., Ganapati, 1936, p. 204; 1938, p. 155.

The affected area presents numerous white pustules, which are cysts containing the spores. Trophozoite shows a clear differentiation into ectoplasm and endoplasm, and vegetative and generative nuclei are present. Pansporoblasts originate by divisions of single generative cells. Each pansporoblast gives rise to two spores. Autogamy occurs. The phenomenon of diffuse infiltration seems to be much pronounced, bringing about considerable pathological changes.

*Remarks.*—This is the second record of a Myxosporidian infecting the heart, the previous one being that by Keysselitz of *Myxobolus cordis* from the ventricle of *Barbus fluviatilis*.

Habitat.—Tissue parasite in the bulbus arteriosus of Otolithus ruber (Bl. Schn.) and O. maculatus (Kuhl & Hass.): MADRAS, Madras.

# II. Order ACTINOMYXIDIA Stole, 1899.

The organisms included in this order are characterized by spores of complicated structure. Each spore has its membrane or shell composed of three valves, which may be drawn out into simple or bifurcated processes. It has three polar capsules, and the polar filaments are visible *in vivo*. Several sporoplasms occur in each spore.

They are parasitic in the body-cavity or the gut-epithelium of freshwater or salt-water Annelids, but have not been studied in India.

#### Identification Table of Families.

1 (2). Spore with a double membrane;	
single piece. A single binucleate	[= Tetractinomyxidae.
sarcoplasm	Hapioactinomyxidæ* Granata
2 (1). Spore-membrane a single trivalve shell. A single eight-nucleate or	{= Triactinomyxida

shell. A single eight-nucleate or [= Triactinomyxidæ. eight-uninucleate sporoplasm... Euactinomyxidæ\* Granata

# III. Order MICROSPORIDIA Balbiani, 1882.

Intracellular parasites typically of Arthropods and Fishes. Pseudopodia and amœboid movements have rarely been observed. Multiplication takes place by schizogony, and the resulting agametes are small, uninucleate bodies with indefinite outlines. Successive nuclear divisions, not accompanied by cell-division, lead to chain formation. These ultimately give rise to sporulating individuals or sporonts. Sporont develops into one to numerous spores. The spores are relatively small and less complex than those of MYXOSPOBIDIA. They are ovoidal or bean-shaped. The spore-membrane is usually of a single piece and envelops the sporoplasm, and the polar filament is very long and fine. The filament may be contained in a polar capsule or lie coiled in the spore. In rare cases there may be two capsules and filaments in a spore. Owing to their small size, the polar capsules and filaments are invisible or obscure in the living spore, but can be rendered visible upon treatment with alkalies. Intermediate hosts are unknown.

The life-history of Stempellia magna, as described by Kudo, may be regarded as typical. When the spore reaches the mid-gut of the Culicine host, the polar filament is extruded, the unicleate sporoplasm creeps out, enters a fat-cell, and reproduces by division (fig. 171, A). The products of this division become multinucleated, with four to eight nuclei (B). These chains then break up into binucleated cells, the nuclei of which discard some chromatin and fuse together in pairs (C). The sporonts thus formed may give rise to a single spore (D), or divide to form two (E), four (F) or eight (G) sporoblasts.



Fig. 171.—Diagram showing the probable development of a typical Microsporidian, Stempellia magna Kudo, × 800. A, amebula coming out from the spore in the mid-gut of Culicine larva; B, stages in schizogony or nuclear increase; C, sporont formation; D-G, formation of one, two, four or eight sporoblasts; H, transformation of a sporoblast into a nucleated spore with polar capsule. (After Kudo.)

Each of these forms a single spore after chromidia formation and reconstitution of small nuclei has taken place (H).

The MICROSPORIDIA are responsible for causing devastating epidemics in silkworms, honey-bees, and certain fishes. All the tissues of the host may become infected, and the tissuecells destroyed on an extensive scale. Many species cause tumour-like masses to be formed in which the organisms may be encapsuled in a membrane derived from the host; in others the membrane may be wanting.

Pérez (1905) subdivided the MICROSPORIDIA into two suborders, SCHIZOGENEA (or OLIGOSPOREA) and BLASTOGENEA (or POLYSPOREA). In the former the principal trophic phase is a uninucleate meront which multiplies by fission, and finally gives rise to a sporont; and in the latter there is a multinucleate plasmodium producing sporonts by internal cleavage. As the exact structure of the trophic phase in many forms



Fig. 172.—Effects of Microsporidian infection upon certain hosts. A, central nervous system of Lophius piscatorius infected by Nosema lophii (Doflein); B, fish showing a heavy infection by Glugea hertwigi Weissonberg; C, a Culicine larva infected by Thelohania opacita Kudo,  $\times 8$ ; D, a Simulium larva infected by Thelohania multispora (Strickland),  $\times 6$ ; E, F, normal and hypertrophied nuclei of the adipose tissue of larvæ of Culex pipiens, the lattor as affected by Stempellia magna Kudo,  $\times 750$ . (After Kudo.)

is not known with certainty, the MICROSPORIDIA are now generally divided, following Léger and Hesse (1922), into two suborders, as follows :--

Spore with a single polar filament.
 Spore with two polar filaments.

Monocnidea Léger & Hesse, p. 348. Dicnidea Léger & Hesse, p. 360.

# I. Suborder MONOCNIDEA Léger & Hesse, 1922.

MICROSPORIDIA in which the spore is provided with one polar filament typically coiled in a polar capsule.

Identification Table of Families.

1.	Spore oval, ovoid or pyriform; if sub-	
	cylindrical, length less than four times	[p. 349.
	the breadth	Nosematidæ Labbé,
2.	Spore spherical or subspherical	Coccosporidæ* Kudo.
3.	Spore tubular or cylindrical, length greater	[Hesse.
	than five times the breadth	Mrazeklidæ* Léger &

# Family NOSEMATIDÆ Labbé, 1899.

Spores oval, ovoid or pyriform; if subcylindrical, length is less than four times the breadth.

#### Key to Indian Genera.

1 (2).	Sporont becomes a single sporoblast	[p. 349.
	forming a single spore	Nosema Nägeli,
2 (1).	Sporont develops into eight sporoblasts	[neguy, p. 356.
	and ultimately into eight spores	THELOHANIA Hen-

#### Genus NOSEMA Nägeli, 1857, emend. Pérez, 1905.

Nosema, Nägeli, 1857, p. 760.
Glugea (part), Thélohan, 1895, p. 356.
Myxocystis, Mrázek, 1897, pp. 1-5.
Nosema (part), Labbé, 1899, pp. 105-8.
Nosema, Minchin, 1903, p. 297; Pérez, 1905, p. 17.
Myxocystis (part), Mrázek, 1910, pp. 245-59.
Nosema, Minchin, 1912, p. 418; Castellani & Chalmers, 1919, p. 529; Kudo, 1924 c, p. 65; Wenyon, 1926, p. 738; Knowles 1928, pp. 324-30; Reichenow, 1929, p. 1088; Kudo, 1931, p. 320; Calkins, 1933, p. 569; Reichenow, 1935, p. 389.

The vegetative form divides by multiple or binary fission into uninucleate rounded bodies or sporonts, each of which gives rise to a single ovoid or pyriform spore, which is not enclosed in a capsule.

#### Key to Indian Species.

Spores oval, $3 \mu$ by $1.7 \mu$ . In bed-bugs.	N. adiei (Christophors), p. 349.
Spores egg-shaped, $3-4 \mu$ by $1-2 \mu$ . In	
silkworms	N. bombycis Nägeli, p. 351.
Spores oval, up to $1.5 \mu$ . In dog-fleas.	N. ctenocephali Kudo, p. 354.

#### 293. Nosema adiei (Christophers). (Fig. 173.)

†Leishmann-Donovan bodies, Adie, 1922 a, p. v.

- Bodies found by Mrs. Adie in salivary glands of bed-bug, Christophers, 1922, p. v.
- \*Bodies observed in Cimex rotundatus, Adie, 1922, pp. 236-8, pl. iv, figs. 1-4.
- Nosema adiei, Short & Swaminath, 1924, pp. 181-4, pl. xiii, figs. 1-3.
- Nosema adisi, Knowles, 1928, pp. 225, 330; Iyengar, 1929, p. 140.

Spores elliptical or oval, with sharp and distinct boundary wall. They stain a rather pale, washed-out blue, with a central dot which may be of a deeper blue or red colour, and both kinds may occur in the same intracellular group. Planonts irregular in shape. Cytoplasm stains a bright shade of slateblue, and possesses a large mass of chromatin, usually excentrically placed. Usually seen singly, and not definitely intracellular. Planonts come to rest in some suitable nidus, round off, forming meronts, which undergo schizogony. Meronts rounded or ovoid. Cytoplasm stains a cobalt blue and there may be one or two comparatively large masses of chromatin which may be central or excentric.



Fig. 173.—Nosema adiei (Christophers). A, spores; B, planonts; C, meronts. (After Short and Swaminath.)

Dimensions.—Planont about 1.6  $\mu$  in diameter; meront 3.2  $\mu$  by 2.7  $\mu$ ; spore 3  $\mu$  by 1.7  $\mu$ .

Habitat.—Gut, salivary glands, and ovaries of Cimex rotundatus Linné : ASSAM.

#### 294. Nosema bombycis Nägeli. (Fig. 174.)

Nosema bombycis, Nägeli, 1857, p. 760.

Panhistophyton ovatum, Lebert, 1858, pp. 149-86.

Pébrine corpuscles, A. de Quatrefages, 1860, pp. 1-638; Pasteur, 1870, pp. 1-327.

Microsporidie, Balbiani, 1884, pp. 150-68.

Pébrine corpuscles, Wood-Mason, 1887, pp. 1-3; Pfeiffer, 1888, pp. 469-86.

Glugea bombycis, Thélohan, 1894, pp. 1425-7; 1895, pp. 357-8.

Nosema bombycis, Labbé, 1899, pp. 106-7.

Glugea bombycis, Toyama, 1900, pp. 1-40.

Nosema bonbycis, Ninchin, 1903, pp. 1-40. Nosema bonbycis, Minchin, 1903, p. 297; Léger & Hesse, 1907, p. 6; Stempell, 1909, pp. 281-358; Minchin, 1912, pp. 411, 413-14, fig. 172; Omori, 1912, pp. 108-22; Kudo, 1913, pp. 368-71; 1916, pp. 31-51; 1918, pp. 141-7; 1924 c, pp. 69-76, figs. 1-39; fig. 757, text-figs. B2, D.

†Nosema bombycis, Hutchinson, 1920, pp. 177-245, pls. i-xxvi.

Nosema bombycis, Wenyon, 1926, p. 736, fig. 313; Knowles, 1928, pp. 324-30, figs. 72-5.

†Nosema bombycis, Iyengar, 1929, p. 140.

Nosema bombycis, Řeichenow, 1929, pp. 1088–90, figs. 1085–9; Kudo, 1931, p. 320, fig. 137, *a*, *b*; Reichenow, 1935, p. 389, fig. 43.

Spore egg-shaped, with the anterior end somewhat narrower than the other. It contains two vacuoles, one near the anterior and the other near the posterior end. The single polar capsule lies axially in the spore, occupying its whole length, and contains a long polar filament wound spirally in its interior. The sporoplasm forms a cytoplasmic girdle round the polar capsule, separating the two vacuoles and placed slightly nearer the anterior pole of the spore. It contains at first a single nucleus which later divides into two and then into four. When the spores are ingested by a silkworm, the polar capsule ruptures in the lumen of its gut, and the polar filament is extruded, attaching the spore to an epithelial cell. This explosion of the polar capsule can be artificially brought about by treating the spores in some infected material with dilute acids or iodine solution, or even by pressure between coverslip and the slide. When germination of the spore takes place, the sporoplasm or amœbula creeps by amœboid movement along the polar filament to an epithelial cell, which it invades. The planonts multiply by binary fission (fig. 174, A), become distributed throughout the body, enter tissue-cells, and become meronts (B). These meronts are spherical or oval in shape, and divide actively by fission (C) or by multiple division (D). The hostcell finally becomes completely filled with schizonts. Each meront ultimately develops into a spore (E). The spores are set free by the disintegration of the host-cells, and are taken with the food into the digestive tract of another host-larva, where the two nuclei of the spore divide once, giving rise to four nuclei (F). The polar filament is extruded (G) and later

becomes detached. The binucleated sporoplasm creeps out through the opening of the capsule (H), and the remaining two nuclei degenerate in the spore. The two nuclei in the amœbula fuse into one and the uninucleate planont is formed. The entire life-cycle may be completed in four days.

Dimensions.—Planont  $0.5-1.5\mu$  in diameter; meront about  $3-5\mu$  in diameter; spore  $3-4\mu$  by  $1-2\mu$ ; polar capsule  $1.5-2\mu$  by  $0.8-1\mu$ ; length of extruded filament  $57-72\mu$ , or even up to  $98\mu$ .



Fig. 174.—Life-cycle of Nosema bombycis Nägeli. I, extracellular stages; II, intracellular stages. A, planonts; B, meront; C, binary fission of meronts; D, multiple division of meronts; E, stages in spore formation; F, ripe spores in the midgut of a new host; G, extrusion of the polar filament; H, amebula leaving the spore. (From Kudo, after Stempell.)

Remarks.—Ohshima (1927), discussing the function of the polar filament of the spore of Nosema bombycis, showed that as soon as filament extrusion is accomplished in the midgut of a silkworm the spore discharges a viscous fluid through the tube of the filament, which remains hanging in a spherical drop at the extremity of the filament. He has now shown (1937) that the polar filament is a germination tubule. In order to germinate the amœbula in the alimentary canal the spore evaginates its filament through the peritrophic membrane

#### NOSEMA.

into the epithelium, and the germ is safely poured out there through the tubule of the filament, protected on the way from the digestive ferment of the canal. The viscous fluid is the amœbula itself. The view previously held of regarding the polar filament as an attachment apparatus is thus contradicted.

Pathogenicity.—The organism eauses a widespread disease, in silkworms, known as pébrine, or by various other names in different countries. In India it is locally known as Cota. The disease spreads among the host-larvæ and to the offspring through the ova, causing disastrous epidemics and entailing huge economic loss to the silkworm industry. The outbreak of such disastrous epidemics in France and other countries of Europe, in the middle of the last century, led Pasteur to make his classical researches into the nature of the disease and the mode of its control.

Lightly infected larvæ do not show definite symptoms, but the heavily infected ones move about sluggishly, lose their appetite, grow slowly, and succumb to death before pupation. The coccoons spun by the infected larvæ are very thin and poor in quality. In advanced stages of the disease irregularshaped dark brown or black spots appear over the surface of the host-body, particularly on the posterior ventral side. The internal organs show a milky-white appearance due to the presence of a large number of spores.

The eggs of the host may become infected while developing in the infected ovaries of the female moth, or the spores may be taken by the host-larvæ into their digestive tract with contaminated food. The fæcal matter of the infected hostlarvæ is the most dangerous source of infection, as the contained spores are easily spread over the mulberry leaves on which the silkworms feed. The disease can, however, be controlled by eliminating both sources of infection. Infected eggs and all stages of the host-insect showing the least sign of infection must be destroyed as soon as possible by frequent and careful inspection.

Hutchinson (1920) has made a study of the disease as it occurs in India. He has shown that pébrine spreads rapidly in Indian silk farms and is a much more acute problem than in Europe. He has further shown that light infections can be identified by dissecting out the gut of the moth and examining the posterior part of the colon, where light infections are usually located. A far greater proportion of infected moths can thus be detected than by the Pasteur method.

Habitat.—All tissues of eggs, larvæ, pupæ, and imagos of Bombyx morii Fabricius.

SPOR.

#### SPOROZOA.

#### 295. Nosema etenocephali Kudo. (Fig. 175.)

Nosema pulicis, Korke, 1916, pp. 725-30, pl. lxiii. Nosema ctenocephali, Kudo, 1924 c, pp. 105-6, pl. v, figs. 178-82; Iyengar, 1929, p. 140.

Spore oval, refractile in fresh state, with one or two vacuolar spaces at the poles, and with an actively mobile sporoplasm.



Fig. 175.—Stages in the life-cycle of Nosema ctenocephali Kudo. A, fully developed sporocyst (C.N., capsulogenous nucleus; P.C., polar capsule; P.F., polar filament; P.N., parietal nucleus; s., spore; s.N., spore-nucleus; s.T., sporocyst); B, germination with extruded filament, with a planont at its extremity (a, sporocyst; b, phront; c, same, highly magnified); C, colony of planonts; T division of meronts to form a chain-like group; E, Unision of meronts to form a cluster; F, capsule and polar filament (After Korke.)

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By addition of weak acetic acid or iodine water the movement within the spore is accentuated and results in the extrusion of the polar filament. The detached filaments are delicate threads, sometimes nodular. The filament issues through the pore of the sporocyst and, as it breaks off, the amœbula emerges through the pore. In stained preparations the amœbulæ are seen as minute bodies with irregular outline, suggesting the possession of pseudopodial movement during life. The planont is a minute globular organism; when active its movements are fairly rapid, a blunt pseudopodium being extruded in one direction only. Division takes place by binary fission. Planonts collect into groups or colonies of varying numbers of individuals.

After penetration into a cell the mobile planont becomes the oval meront. The meront is larger than the previous phases, has homogeneous cytoplasm, with a centrally or excentrically placed refractile spot. It forms generations of merozoites, either by binary fission forming chain-like groups (fig. 175, D) or by multiple fission forming small clusters of cells (fig. 175, E). The final product of the division of the meront is a sporont which forms the sporocyst, polar capsule, and polar filament. The spore is recognized by its egg-shaped outline and by the presence of one or two vacuoles at either pole. In stained films can be seen, in addition, a cystic wall containing two parietal nuclei, a polar capsule and polar filament, and the sporoplasm. The polar capsule is a hollow pear-shaped body containing a spirally twisted thread, and a capsulogenous nucleus situated at its broader end. The sporoplasm contains a single nucleus and no vacuoles.

Dimensions.—Planont 0.75-1 $\mu$ ; spore up to 1.5 $\mu$  in length; length of extruded filament 25 $\mu$ .

*Remarks.*—Heavily infected larvæ are dark and mottled in appearance, owing to the presence of the *Nosema*, and are thus easily distinguished.

Korke (1916) described the species as N. pulicis, probably in ignorance of the fact that Nöller (1912, 1914) had already described a species of that name from the dog-flea in Germany. As the dimensions of the spores are quite different in the two forms, Kudo (1924) has given the name N. ctenocephali to the Indian form.

Habitat.—Digestive tract of the larvæ of the dog-flea, named by Korke Ctenocephalus felis (Bouché), probably Ctenocephalus canis (Curtis): PUNJAB, Kasauli.

#### SPOROZOA.

#### Genus THELOHANIA Henneguy, 1892.

Thelohania, Henneguy, in Thélohan, 1892, p. 174; Gurley, 1893, p. 410; Thélohan, 1895, p. 361; Labbé, 1899, pp. 111-12; Minchin, 1903, pp. 285, 297, 320; Pérez, 1905, p. 17; Minchin, 1912, p. 418; Castellani & Chalmers, 1919, p. 529; Kudo, 1924 c, pp. 67, 130-63; Wenyon, 1926, p. 738; Reichenow, 1929, p. 1102; Kudo, 1931, p. 321; Calkins, 1933, p. 569; Reichenow, 1935, p. 390.

Each sporont develops into eight sporoblasts and ultimately into eight spores. The sporont-membrane may degenerate at different times of development.

#### Key to Indian Species.

1 (4).	Spores oval, with equally rounded ex-	
	tremities	2.
2 (3).	Spore $4-5\cdot 2\mu$ by $2\cdot 4-2\cdot 8\mu$	T. indica Kudo, p. 356.
3 (2).	Spore $4.75-6 \mu$ by $3-4 \mu$	T. legeri Hesse, p. 357.
4(1).	Spores broadly oblong with dissimilar	
	extremities, showing a small knob at	
	one end and a clear oblong space at	
	the other; $4.5-5 \mu$ by $3-3.5 \mu$	T. obscura Kudo, p. 359.

#### 296. Thelohania indica Kudo. (Fig. 176.)

*Thelohania* sp., Iyengar, 1929, p. 138.
 *Thelohania indica*, Kudo, 1929, pp. 3-4, figs. 19-50.

The schizonts are elongated bodies containing four to eight



Fig. 176.—Thelohania indica Kudo. A, tetranucleate schizont; B, octonucleate schizont; C, division into binucleate schizonts; D-F, stages in formation of sporont; G, sporont; H, sporont with eight sporoblasts; I, sporont with mature spores; J, stained spore. (After Kudo.)

#### THELOHANIA.

nuclei arranged in pairs (fig. 176, A, B). They divide finally into two to four binucleate schizonts (C). The two nuclei become enlarged and closely associated, and ultimately fuse into one, thus forming a sporont (D-F). The sporont-nucleus undergoes three successive divisions, producing eight sporo-In each sporoblast the nucleus divides into two blasts. masses. One of them remains as a single nucleus of the sporoplasm, while the other disintegrates and produces the polar filament. The unstained spores are ellipsoidal, with equally rounded extremities; the spore-membrane is moderately thick, and there is no indication of its being composed of two valves; the contents are irregularly vacuolated. The stained spore shows a comparatively large nucleus at one end and a deeply stained polar capsule near the other.

Dimensions.—Spores measure  $4-5\cdot 2\mu$  by  $2\cdot 4-2\cdot 8\mu$ .

Habitat.-Adipose tissue cells of the larvæ of Anopheles hyrcanus (Giles) : BENGAL.

- 297. Thelohania legeri Hesse. (Fig. 177.)
  - Thelohania legeri, Hesse, 1904 a, pp. 570-1, 1904 b, pp. 571-2, 10 text-figs.
  - Thelohania illinoisensis, Kudo, 1921, pp. 167-71, 177; 1922, pp. 74-5.
  - Thelohania legeri, Kudo, 1924 a, pp. 147-62. 1 pl., 9 text-figs.; 1924 c, pp. 143-6. figs. 499-507, 694-727, 764, text-fig. H; Wenyon, 1926, p. 748.
  - †Thelohania sp., lyengar, 1929, p. 138.
  - †Thelohania legeri, Kudo, 1929, pp. 2-3, figs. 1-18.
    - Thelohania legeri, Reichenow, 1929, pp. 1086, 1104; Kudo, 1931, p. 321. fig. 138, a-f; Calkins, 1933, p. 554, fig. 223.

The earliest stages are unknown. The youngest stages found in the infected cells of the "fat-body" of the host are rounded bodies with compact chromatin granules (fig. 177, A). The nucleus becomes vesicular and then divides ; the cytoplasm becomes constricted and two uninucleated daughter-schizonts are formed (B). This division is repeated. Some of the binucleate schizonts do not split, but grow into large elongate bodies with four nuclei (C). Each of these divides into two large binucleate schizonts. These nuclei lose their vesicular nature, become compact, and divide, the daughter halves remaining connected with each other by a strand. By further division two binucleate forms are formed, the nuclei in each being cousin nuclei and not daughter nuclei (D). Sometimes octonucleate schizonts occur, which by division produce four binucleate schizonts. The zygote or sporont is formed by the fusion of the two nuclei (E). The nucleus of the zygote divides three times in succession, resulting in stages with two (F), four (G), and eight nuclei (H). The sporont is then transformed into a pansporoblast with eight sporoblasts, each of which develops into a spore (I). When the spore reaches the gut of a new host-larva the filament is extruded (J) and the sporoplasm emerges as an amæbula, starting the development again.

Dimensions.—Meronts  $3-4\mu$  in diameter; sporonts  $9-10\mu$  by  $4-6\mu$ ; spores  $4.75-6\mu$  by  $3-4\mu$ .

Remarks.—Iyengar (1929) described the life-cycle of Thelohania sp. from Anopheles pseudojamesi Strickland & Chowdhury (=A. ramsayi Covell), and found it similar to T. legeri as described by Kudo. The measurements of the spores as recorded by him also fall within the limits of this species.



Fig. 177.—Life-cycle of Thelohania legeri Hesse. A, youngest stage; B, formation of schizonts; C, large schizont with four nuclei; D, formation of binucleate forms; E, sporont; F, G, H, nuclear division of sporont to form binucleate, tetranucleate or octonucleate stages; I, pansporoblast with eight sporoblasts; J, spore with extruded filament. (After Kudo.)

Habitat.—Adipose tissue cells of larvæ of Anopheles barbirostris Van der Wulp, A. annularis Van der Wulp (=A. fuliginosus Giles), A. varuna Iyengar, A. hyrcanus (Giles), A. ramsayi Covell, and A. subpictus Grassi (=A. rossi Giles): BENGAL.

#### 298. Thelohania obscura Kudo. (Fig. 178.)

†Thelohania sp., Iyengar, 1929, p. 138. †Thelohania obscura, Kudo, 1929, p. 4, figs. 51-61.

Developmental stages not studied. Sporont octosporoblastic (fig. 178, A). Spores, when viewed in water after decolorization, are broadly oblong, often with dissimilar extremities, with a thin membrane containing within a rounded mass of homogeneous and refractile protoplasm showing a small knob at one end and a clear oblong space at the other. The stained



Fig. 178.—*Thelohania obscura* Kudo. A, sporont containing eight sporoblasts; B, young spore; C, mature spore. B and C stained with Giemsa's stain. (After Kudo.)

spores show a nucleus near one end and a coiled polar filament forming an oblong mass (B, C).

Dimensions.—Spores measure  $4.5-5\mu$  by  $3-3.5\mu$ .

Remarks.—The record of T. legeri from India and the descriptions of T. indica and T. obscura are based on material sent to Kudo by M. O. T. Iyengar from Bengal. As remarked by the latter, these parasites form whitish translucent masses beneath the cuticle of the thorax and the anterior abdominal segments of the larvæ. The infected segments show hypertrophy. None of the larvæ showing these symptoms metamorphosed, and all died before pupation. Those that are lightly infected would be able to metamorphose into adults.

Habitat.—Larva of Anopheles varuna Iyengar : BENGAL.

#### 299. Gen. et sp. incert.

Protozoan, Ross, 1906, p. 104. Thelohania (?) sp., Kudo, 1921, p. 71; 1922, pp. 71-2. Gen. et spec. incert., Kudo, 1924 c, p. 194.

Eight spores were seen to be closely packed within an oval envelope. Spores were oval, refractive, apparently hard, with a circular vacuole at one focus of the ellipse.

Habitat.—Nerve-cord of imago of Stegomyia sp. and Culex fatigans Wied. : INDIA.

# II. Suborder DICNIDEA Léger & Hesse, 1922.

MICROSFORIDIA in which the spore is provided with two polar capsules, one at each end, each containing a polar filament.

The suborder includes the single family Telomyxidæ\* Léger & Hesse, 1910, but no representative has been recorded from India up to the present time.

# IV. Order H E L I C O S P O R I D I A Kudo, 1931.

Minute spore, composed of a thin membrane of one piece, containing three uninueleate sporoplasms, around which is coiled a long, thick filament. Young trophozoites are histozoic or cœlozoic, undergo schizogony, and produce uninueleate sporonts. The sporont divides twice and forms four small cells, each of which develops into a spore.

The order has been created by Kudo (1931) to include the species *Helicospiridium parasiticum* Keilin from the body of certain insects. The structure of the spore is quite peculiar, and the species was previously referred to HAPLOSPORIDIA. According to Kudo it seems to be best placed in the CNIDO-SPORIDIA, if it is a Protozoan.

# III. Subclass SARCOSPORIDIA Bütschli, 1882.

These are typically parasites of the muscles of mammals, although birds and some reptiles are also known to harbour them. They usually affect the striped muscles of the skeleton. tongue, larynx, and diaphragm. Lying between the musclefibres are thin-walled, whitish, tubular cysts termed Miescher's tubes, which are the remains of an infected muscle-fibre, the membrane of which is distended over the sac containing the parasites. They are generally visible to the naked eye, and often very large. When examined microscopically each is seen to consist of an envelope, from which partitions run inward, dividing the interior into a number of chambers containing vast numbers of crescent-shaped spores, the so-called "Rainey's corpuscles." The spore is rounded at one end and pointed at the other; it contains a nucleus near the rounded end and a collection of deeply staining chromatinic grains near the pointed end. Laveran and Mesnil described a striated structure at the pointed end which several workers considered as suggesting a rudimentary polar capsule, but recent authorities do not regard it as such.

The spores are taken into the digestive tract of a specific host through the mouth. The spore-membrane ruptures, setting free the sporozoite, which enters the gut-epithelium. After multiplying there the organism makes its way into the muscular tissue. At first there is an elongate multinucleate mass, which may or may not divide into uninucleate bodies and which become the centre of infection in other muscle-fibres. Some of these trophozoites grow in size, and the body becomes divided into chambers, inside which spores are formed.

Development has been studied chiefly in experimentally infected mice by Erdmann (1910, 1914), Negri (1910), and Crawley (1916), and in sheep by Alexeieff (1913). Smith (1901, 1905) was the first to demonstrate that mice could be infected by feeding them with the flesh of other infected mice. Negri (1910) and Darling (1910 a) showed that guineapigs could be infected with the parasite of rats, and Darling also pointed out that the forms in guinea-pigs are morphologically identical with those from man. Erdmann (1910) infected mice with the parasite of sheep. Wenyon (1926) gives a list of thirty-nine species of *Sarcocystis* from a variety of hosts, but agrees with Alexeieff that there is no means of distinguishing the supposed species.

Babudieri (1932) regards the SARCOSPORIDIA as more closely related to the COCCIDIA than the CNIDOSPORIDIA. He divides the subclass into SARCOSPORIDIA s. str. (including the families Sarcocystidæ and Fibrocystidæ) and GLOBIDIA, including the family Globididæ. The Sarcocystidæ includes the genus Sarcocystis with twenty-two species. The Fibrocystidæ includes Fibrocystis and Besnoitia with two species The Globididæ includes Globidium with five species each. and *Ileocystis* with one species. The work should be referred to for the diagnostic features, synonymy, and hosts of various species. Krause and Goranoff (1933) tabulate the morphological differences of forms described from birds. They succeeded in infecting the fowl with the strain from the buffalo.

Pathogenicity.—In many cases, even with fairly heavy infection, the host may not be adversely affected; but in others the host sooner or later dies as the result of muscular degeneration. The organisms are known to produce a peculiar substance called sarcocystin, which is highly toxic when injected into other animals. Teichmann and Braun (1911) showed that rabbits could be immunized against the toxin, and that the serum contained antibodies which could produce passive immunity in other animals.

# I. Order SARCOSPORIDIA s. str., Babudieri, 1932.

Spore banana-shaped; the cyst is wholly or partially of parasitic origin, and the spore produces metachromatic granules and secretes a toxin known as sarcocystin. The spore is produced by the proliferation of the pansporoblast, and the sporoblast has a peripheral situation in the cyst.

### Identification Table of Families.

1 (2	).	Cysts elongated in form, parasitic in the	[Babudieri, p. 363.
		striped muscle-fibres	Sarcocystidæ
2 (1	.).	Cysts rounded in form, parasitic in the	[Babudieri.
		connective tissue or plain muscle fibres .	Fibrocystidæ*

# Family SARCOCYSTIDÆ Babudieri, 1932.

#### Genus SARCOCYSTIS Lankester, 1882.

Sarcocystis, Lankester, 1882, p. 54.

Sarcocystis + Balbiania + Miescheria, Blanchard, 1885, p. 244.

Sarcocystis, Labbé, 1899, pp. 116-19; Minchin, 1903, p. 308: Minchin, 1912, pp. 419-22; Castellani & Chalmers, 1919, pp. 530-3; Wenyon, 1926, pp. 760-9; Knowles, 1928, pp. 333-7; Reichenow, 1929, pp. 1120-7; Kudo, 1931, pp. 326-8; Calkins, 1933, pp. 555-6; Reichenow, 1935, p. 394.

With the characters of the family.

The cysts are to be found in practically all body-muscles; in Ruminants they are found specially in the cosophagus, in other animals in the heart, intercostal or abdominal muscles. The cyst-wall may be produced entirely by the parasite (as in Sarcocystis muris) or in part by the host (as in S. tenella). The cyst is divided into chambers, the peripheral ones containing pansporoblasts or sporoblasts, while the central ones contain mature spores. The mature spores have neither polar capsules nor polar filaments. They show a very granular nucleus, a thin cell-membrane, and many metachromatic granules in the evtoplasm.

Many species have been recognized as occurring in man, sheep, cattle, horses, rats, and mice on the basis of occurrence in different host-species and slight difference in the dimensions of the spore; but they are otherwise morphologically indistinguishable.

#### 300. Sarcocystis blanchardi Doflein, 1901. (Fig. 179.)

Sarcocystis blanchardi, Doffein, 1901, p. 221, figs. 197, 198.

†Sarcocystis tenella, Shipley, 1904, pp. 45-7, pl. i, figs. 10, 17.

†Sarcocystis tenella bubalis, Willey, Chalmers, & Philip, 1904, pp. 65-72, figs. 1, 2.

†Sarcocystis sp., Chatterjee, 1907, pp. 77-8. Sarcocystis tenella bubali, Wenyon, 1926, p. 768.

Sarcocystis blanchardi, Wenyon, 1926, p. 768.

Sarcocystis tenella. Wenyon, 1926, pp. 760, 768, fig. 329; Knowles, 1928, pp. 333-6.

Sarcocystis blanchardi, Reichenow, 1929, p. 1123, figs. 1126, 1127; 1935, p. 392, fig. 46.

Cysts somewhat pointed, though not very sharply, at either end, or both ends truncated, the thickest portion being in the middle. The covering of the cyst consists of two sheaths, the outer one being continuous and the inner forming partitions which divide the cyst into a number of polyhedral chambers with granular contents. The peripheral chambers are completely filled with fine granulations and stain deeply. The central chambers are empty or contain spores. Spores are minute crescent-shaped bodies with granular contents.

Dimensions.-The largest cysts measure 30 mm. in length, 5 mm. in breadth, and 3 mm. in thickness.

**Remarks.**—Shipley (1904) and Willey, Chalmers, and Philip (1905) first described the sarcosporidial infection in buffaloes in Ceylon. According to Wenyon as many as four species have been named by different observers from the buffalo, but these are probably identical, and Reichenow recognizes only *S. blanchardi*. Chatterjee (1907) found the cysts and spores of *Sarcocystis* sp. in the heart-muscle of a cow. He did not find fine radiations round the capsule. The spores



Fig. 179.—Sarcocystis blanchardi Doflein. A, transverse section; B, longitudinal section of the infected muscle-fibres of a young individual (a, muscle-fibre; b, parasite); C, longitudinal section through a portion of the cyst in the æsophagus of cattle (a, muscle-fibre; b, cyst-membrane; c, nucleus of the muscle-fibre; d, spores; e, partition-wells between the chambers). (From Wasielewski, after Van Eecke.)

were grouped in loculi, but distinct alveolar partitions could not be made out.

Habitat.—Muscles of buffalo, Bos bubalus Linn.: CEYLON; heart-muscle of cow, Bos indicus Linn.: BENGAL, Calcutte.

# 301. Sarcocystis lindemanni (Rivolta). (Fig. 180.)

?, Lindemann, 1863, p. 426; 1865, p. 382.

Gregarina lindemanni, Rivolta, 1878, p. 12; Haden, 1883, p. 326; Blanchard, 1885, p. 244; Eve, 1889, p. 444; Target, 1889, p. 444.

Sarcocystis hominis. Rosenburgh, 1892; Baraban & St. Remy, 1894, pp. 79-82, figs. 1-5.



Fig. 180.—Sarcocystis lindemanni (Rivolta). A. B. transverse sections of infected muscle-fibres; C, individual spores. (After Vasudevan.)

Sarcocystis lindemanni, Labbé, 1899, pp. 117-18; Minchin, 1903, p. 351.

†Sarcocystis sp., Darling, 1919, p. 98.

Sarcocystis lindemanni, Hegner & Taliaferro, 1924, pp. 373-4, figs. 139 b, 140; Wenyon, 1926, pp. 767-8.

- †Sarcocystis sp., Vasudevan, 1927, pp. 141-2, pls. xxiii, xxiv, figs. 1-9.
  - Sarcocystis lindemanni, Knowles, 1928, p. 336, fig. 77, 2, 3; Reichenow, 1929, pp. 1123, 1125, fig. 1128; Kudo, 1931, p. 328; Reichenow, 1935, p. 394.

The cysts (the so-called "Miescher's tubes") are just visible to the naked eye as very thin, long, whitish streaks in the muscle-fibres and running parallel to them. The sheath proper is clear and homogeneous. External to this is a false sheath formed by compressed muscle-fibres showing clearly their nuclei and striation. From the true capsule numerous septa run into the cyst, dividing it into a number of partitions and giving a honeycombed appearance. On crushing one of these cysts numerous tiny curved bodies or spores are liberated. These spores are somewhat crescentic and pointed at both ends. No terminal filaments could be seen. The nucleus in each spore is oval, placed along the long axis either in the centre or, more commonly, subterminally. In some spores near one end is a stained area which at one time was supposed to represent a polar capsule. No vacuole is present.

Dimensions.—In the case of human sarcosporidiosis observed by Baraban and St. Rémy Miescher's tubes were from 150 to  $1600\mu$  long and from 77 to  $168\mu$  in thickness; Vasudevan gives 5.3 cm. as length and  $322\mu$  as thickness. Spores are  $8.33\mu$  in length and  $1.66\mu$  in width, the nucleus in the spore measuring  $3.33\mu$  by  $1.66\mu$ .

*Remarks.*—The cysts of this parasite are much thinner and longer than those of *S. miesheriana*, and are just visible to the naked eye. The spores also are smaller and both ends are pointed instead of one end being blunt as in that species.

The parasite described by Vasudevan (1927) is of special interest, as only six or seven cases of sarcosporidial infection have previously been recorded in man.

Habitat.-Muscles of a man : MADRAS, Madras.

# II. Order GLOBIDIA Babudieri, 1932.

Spores fusiform containing large siderophil granules in the centre. Cysts occur exclusively in the intestinal submucosa, inside a hypertrophied host-cell, with a peripheral nucleus. No toxin secreted.

### Family GLOBIDIDÆ Babudieri, 1932.

With the characters of the order.

#### Genus GLOBIDIUM Flesch, 1884.

Globidium, Flesch, 1884, p. 459; Labbé, 1899, p. 72; Minchin, 1903, p. 350.

Gastrocystis, Chatton, 1910, p. 114.

Besnoitia, Franco & Borges, 1916, p. 269.

Globidium, Wonyon, 1926. pp. 769-73 : Knowles, 1928, pp. 337-8 ; Reichenow, 1929, pp. 905-7 ; 1935, p. 394.

Cysts spherical, up to 5 mm. in diameter, embedded in the mucosa of the alimentary canal or skin of mammals. Each is enclosed by a membranous capsule and, when full grown, contains groups of spores which resemble those of SARCOSPORIDIA.

**Remarks.**—Flesch (1883) observed the infection in the horse; Chatton (1910) in sheep and goats; Gilruth and Bull (1912) in the kangaroo, wallaby, and wombat; Kupke (1923) in the horse; Wenyon and Scott (1925) in the wallaby; and Henry and Masson (1932) in the camel. There is great divergence of opinion as regards the systematic position of the genus. Wenyon (1926) considers it as closely related to SARCOSPORIDIA, Reichenow (1929) places it in an appendix after GREGARINIDA, and Henry and Masson (1932) regard the genus as large Coc-CIDIA, while Hobmaier (1922) believed that the parasites included in the genus are fungi, and not PROTOZOA at all. Babudieri (1932) placed it in a separate order of the subclass SAROOSPORIDIA. Enigk (1934) has described the development of macrogametes, microgametes, and schizonts of Globidium cameli and pointed out the similarity to that of CoccIDIA.

#### 302. Globidium fusiformis Hassan, 1935. (Fig. 181.)

†Globidium fusiformis, Hassan, 1935, pp. 1-7, pls. viii-x; Ware, 1937, p. 35.

Spherical protoplasmic bodies, varying in size, the smallest with a single nucleus and the largest with as many as fortyeight or more nuclei in a single optical plane. The parasite is at first intracellular in the gastro-intestinal epithelial cells, but later the tissues form a thin capsule round the mass. The colony becomes intracellular and is pushed towards the subepithelial connective tissue. Ultimately the cysts are

#### SPOROZOA.

liberated in the lumen of the organ. The centrally situated single nucleus grows and undergoes division, and the many nuclei become arranged round the periphery of the cyst. Finally projections representing the extremities of marginally situated spores appear to radiate round the periphery of the spherical body. In a freshly ruptured cyst twenty or more spores are seen lying parallel to one another. Spores are elongate, spindle-shaped, and slightly curved, one end being more finely pointed than the other. The nucleus is oval, vesicular, and situated near the rounded end of the spore. In the centre of the spore is a round chromatin mass which stains intensely, and minute granules of chromatin are also irregularly distributed on both sides of the nucleus.



Fig. 181.—Globidium fusiformis Hassan. A, multinucleated cytoplasmic bodies; B, a stage in the development of the spores; C, cluster of spores from a freshly ruptured cyst; D, individual spores. (After Hassan.)

Dimensions.—Cysts measure  $60-90\,\mu$  in diameter; spores are  $13\,\mu$  by  $2-2\cdot5\,\mu$ , the nucleus measuring  $3\cdot25\,\mu$  by  $2\cdot25\,\mu$ .

*Remarks.*—The parasite was examined in the dried faces of a cow, and it is not certain if the form is morphologically distinct from G. besnoit of the cattle and G. faurei (G. gilruthi) of the sheep and goats.

Habitat.—Fæces of cow, Bos indicus Linn. : PUNJAB, Gurgaon.

#### 303. Globidium sp.

Sarcocystis macropodis, Gilruth & Bull, 1912, pp. 432–50, pls. lxxiilxxxi.

Globidium sp., Wenyon, 1926, p. 773. †Globidium sp., Knowles, 1928, p. 337.

*Remarks.*—Gilruth and Bull (1912) recorded the infection in the kangaroo, wallaby, and wombat in Australia. Knowles (1928) mentions that Acton observed a case of infection in a wallaby in the Zoological Gardens, Alipore, Calcutta, but a description does not appear to have been published.

Habitat.--Alimentary canal of a wallaby : BENGAL, Calcutta.

# IV. Subclass HAPLOSPORIDIA Lühe, 1900.

HAPLOSPORIDIA are characterized by the presence of large spores, each containing a single voluminous nucleus, but no polar capsule, and showing a simple type of development. The young parasite is an amœbula, which at first multiplies by fission. The nucleus of each daughter-cell multiplies repeatedly and produces a multinucleate plasmodium. The plasmodium may divide (plasmotomy) or produce merozoites (schizogony) or form spores. The spores are produced from sporoblasts, each forming a single spore, or from pansporoblasts, each giving rise to a number of spores. Infection is carried by the spores, which produce amœbulæ and start a new generation.

The spore is spherical or ellipsoidal, and is covered by a resistant membrane which may be marked by ridges or tubercles, and may be prolonged into a tail-like process. In a few species the spore-membrane possesses a lid, which opens to allow the uninucleate sporoplasm to emerge as an amœbula.

The subclass includes parasites of certain Invertebrates, chiefly Annelids, and Fish. They float freely in the bodycavity fluid of the Invertebrates or infest the tissuc-cells, such as those of the intestine. In Fish they attack the gills or other tissues, giving rise to white nodules. One species occurs in the Malpighian tubules of the cockroach. The genus *Rhino*sporidium, species of which form little cysts in the nasal cavities of man and horse, used also to be included in this subclass, but the studies of Ashworth (1923) have shown that it is not a Protozoon but a fungus.

Debaisieux (1916, 1920) has shown that certain species of Haplosporidium Caullery & Mesnil and Bertramia Caullery & Mesnil should be included with the MICROSPORIDIA, while Cælosporidium Crawley is not a Sporozoon. In his view HAPLOSPORIDIA should be abandoned as a separate group.

For classification of the HAPLOSPORIDIA, Caullery and Mesnil (1905) and Ridewood and Fantham (1907) may be referred to. The latter divided the HAPLOSPORIDIA into two sections—the OLIGOSPORULEA, in which each pansporoblast produces only a small number of spores or a single spore, and the POLYSPORULEA, in which the pansporoblast gives rise to many spores, either successively or almost simultaneously. No one has as yet studied HAPLOSPORIDIA in India.

#### Doubtful Protozoa.

#### Genus RHINOSPORIDIUM Minchin & Fantham, 1905.

Rhinosporidium, Minchin & Fantham, 1905, pp. 521-32; Minchin, 1912, pp. 424-5; Castellani & Chalmers, 1919, pp. 533-4; Ashworth, 1923, pp. 301-42; Hegner & Taliaferro, 1924, pp. 375-7; Wenyon, 1926, pp. 776-8; Knowles, 1928, pp. 338-42; Reichenow, 1929, pp. 1136-7.

Organisms giving rise to polypi, especially in the nose, of human beings and horses. Believed for a long time to belong to HAPLOSPORIDIA, but now generally recognized as a fungus.

#### 304. Rhinosporidium seeberi (Wernicke). (Fig. 182.)

Coccidium seeberi, Wernicke. 1900.

?, O'Kinealy, 1903 a, pp. 109-12; 1903 b, pp. 43-4, 1 pl. +Rhinosporidium kinealyi, Minchin & Fantham, 1905, pp. 521-32, pls. xxx, xxxi.

Rhinosporidium seeberi, Minchin, 1912, pp. 424-5.

*Rhinosporidium kinealyi*, Tirumurti, 1914, pp. 703-19.

†Rhinosporidium seeberi, Castellani & Chalmers, 1919, pp. 533-4, fig. 190; Ashworth, 1923, pp. 301-42, 5 pls.

Rhinosporidium seeberi, Hegner & Taliaferro, 1924, pp. 375-7, fig. 141; Wenyon, 1926, pp. 777-8, figs. 335, 336; Reichenow, 1929, pp. 1136-7, fig. 1141.

The youngest forms are spherical bodies, about  $6 \mu$  in diameter, embedded in the cytoplasm of connective tissue cells, with chitinoid envelope, vacuolated cytoplasm, and a vesicular nucleus containing a karyosome. Growth is accompanied by nuclear multiplication and the cytoplasm becomes laden with numerous food-granules. When the parasite is about  $100\,\mu$  in diameter and has about 128 nuclei the chitinous envelope becomes much thickened by the deposition of cellulose on the inner surface, except at one point where the future pore will be formed. Nuclear divisions continue, and when there are about 4,000 nuclei the cytoplasm divides into rounded cells, which divide twice to form about 16,000 young spores within the sporangium. In the mature sporangium there are usually fully developed spores in the centre, and small, immature ones arranged peripherally. The fully-formed spore has a chitinous envelope, a vesicular nucleus with a karyosome, and cytoplasm containing one to sixteen refringent spherules of reserve food material. The spores are spherical or oval. They are discharged in hundreds through the pore of the sporangium, and are scattered throughout the connective tissue by the lymph exudate. The reserve food material is gradually used up and the spores enter into fresh connective tissue cells to repeat the cycle. The ripe sporangia are seen as white dots covering the surface of the nasal polypus.

Dimensions.—Ripe sporangia  $250-300\,\mu$  in diameter; spores  $7-10\,\mu$  in diameter.

*Remarks.*—The organism has been found in polypoid growths in nose, naso-pharynx, uvula, conjunctiva, lacrymal sac, ear, and penis of man.

Tirumurti (1914) recorded fifteen cases from man, affecting various parts of the body, in the Madras Presidency.



Fig. 182.—Rhinosporidium seeberi (Wernicke). A, section of stage with about 500 nuclei, showing the outer chitinous and inner cellulose layer of the envelope and the position of the future pore; B, section of the stage in which the contents of the sporangium have divided into about 4,000 nucleated cells; C, discharge of mature spores; D, section of spore showing nucleus with karyosome, and vacuolated cytoplasm. (After Ashworth.) 2 B 2 Researches of Ashworth (1923) apparently prove that the organism belongs to the fungi, and ought to be classed among the *Phycomycetes*.

Habitat.—Polypi in the nose of man : BENGAL, Calcutta; MADRAS, Madras; CEYLON.

#### 305. Rhinosporidium sp.

† Not identified, Vasudevan, 1932, pp. 299-302, pl. viii.

Young trophozoite a spherical encapsuled body, with a distinct nucleus and nucleolus. The capsule is hyaline, doubly-contoured, and  $1.6\mu$  in thickness. The cytoplasm is vacuolated and in some trophozoites contains thin protoplasmic granules. Nucleus central, occasionally peripheral and marginal,  $4.8\mu$  in diameter, with nucleolus  $1.6\mu$  in diameter. In later stages the organism is larger and protoplasmic strands and granules are pronounced. One of them was somewhat pearshaped, probably due to an oblique section.

Dimensions.—The organism measured  $40\mu$  by  $23\mu$ .

*Remarks.*—The organism somewhat resembles the earlier stages of *Rhinosporidium* in shape, hyaline capsule, protoplasmic strands, and granules, but differs from *Rhinosporidium seeberi* in that the earliest stage is much larger. The later stages, with spore-bearing sporangia, were not found. The infection was in the skin and not in a mucous membrane.

Habitat.—Inside walls of abscess cavities, and sometimes in the chronic inflammatory fibrous tissue in man : CEYLON.

#### 306. Rhinosporidium equi Zschokke.

Rhinosporidium equi, Zschokke, 1913, pp. 641-50; Hartmann, 1921, p. 1387; Wenyon, 1926, p. 778.
†Rhinosporidium sp., Rao, 1938, pp. 263-4.
†Rhinosporidium equi, Sahai, 1938, p. 264.

Probably identical with R. seeberi from man.

Remarks.—According to Sahai (1938) the first case in an equine was recorded by Krishnamurti Ayyar in Madras in 1932. Sahai has recorded another case in a country-bred mare in Orissa. The animal had noisy breathing and a bloodstained mucous discharge oozing from one of the nostrils. There was a small cauliflower-like growth about one inch long by half an inch thick situated in the anterior part of the nasal chamber, slightly obstructing the passage. In the connective tissue were found numerous cysts or sporangia in various stages of development, the fully mature ones bursting to discharge the spores. Wenyon (1926) doubted if the equine form is distinct from the human form, and Sahai (1938) supports his view.

Habitat.—Nasal cavities of Equus caballus Linn. : MADRAS, Madras; ORISSA, Bargarh (Sambalpur).

#### 307. Rhinosporidium sp.

†Rhinosporidium sp., Rao, 1938, p. 263.

Probably identical with R. seeberi from man.

Remarks.—According to Rao (1938), Rhinosporidiosis was recorded a few years ago affecting two bullocks, one cow, and one pony in Madras, and has since been diagnosed in eighteen bullocks and one pony in the same province. Rhinosporidiosis in bovines appears so far to have been reported from the Madras Presidency only. The lesions are found in the nose, and the presence of a trauma in the nose appears to be necessary for the development of the lesions.

Rao thinks that the causal organism may be the same as in man, and suggests that the infection to man and animals may be through the dust raised while ploughing fields manured with bovine dung, as a large number of cases are met with in man and cattle engaged in agriculture.

Habitat.—Nasal cavities of Bos indicus Linn.: MADRAS PRESIDENCY.

# APPENDIX.

(i) Supplementary List of Parasites and their Hosts.

Parasite.	Host.	Seat.
GREGARINIDA.		
Fam. STOMATOPHORIDÆ (D.	. 18).	
Stomatophora primitiva	Eunice siciliensis	Intestine.
Fam. LECUDINIDÆ (p. 18).		
Lecudina eunicæ	Eunice siciliensis	Intestine.
Lecudina lysidicæ	Lysidice collaris	Intestine.
Fam. GREGARINIDÆ (p. 19)	•	
Ulivina eunicæ	Eunice siciliensis	Intestine.
Deuteromera cleava	Eunice siciliensis	Intestine.
Contortiocorpa prashadi	Eunice siciliensis	Intestine.
Fam. SELENIDIDÆ (p. 19).		
Selenidium amphinomi	Amphinome rostrata	Cœlomic cavity.
Coccidia.		
Fam. EIMERIIDÆ (p. 21).		
Eimeria arloingi	Capra hircus	Alimentary canal.
Eimeria culindrica	Bos indicus	Alimentary canal
Eimeria aunti	Natrix piscator	Rectum.
Eimeria koormæ	Lissemus punctata	Rectum.
Eimeria labbeana	Columba livia	Intestine.
Eimeria emithi	Bos bubalus	Alimentary canal
	Bos indicus	Alimentary canal.
Eimeria wassilewskyi	Axis axis	Alimentary canal.
Eimeria zürnii	Bos bubalus	Alimentary canal.
	Bos indicus	Alimentary canal.
INCERTÆ SEDIS (p. 22).		
Taxoplasma butasturis	Butastur teesa	Loucocytes.
HÆMOSPORIDIA.		
Fam. HAMOPROTIEDA (pp.	. 23–4).	
Hæmoproteus (?) halcyonis fuscæ.	Halcyon smyrnensis fusca.	Blood.
Hæmoproteus (?) lanii	Lanius schach erythro- notus.	Blood.
Hæmoproteus (?) therei- cerwis var. zeulonica	Thereiceryx viridis	Blood.
Leucocytozoon ardeolæ	Ardeola gravii	Blood.
Leucocytozoon enriquesi	Chloropsis jerdoni	Blood.
Leucocytozoon molpastis	Molpastes cafer cafer	Blood.
Leucocytozoon (?) sp	Oriolus oriolus kundoo.	Blood.
Leucocytozoon (?) sp	Oriolus xanthornus xan- thornus.	Blood.

Parasite.	Host.	Seat.		
Fam. PLASMODIIDÆ (pp. 24-5).				
Proteosoma centropi	Centropus sinensis par- roti.	Blood.		
Proteosoma heroni	Pond heron	Blood.		
Proteosoma passeritæ	Passerita mycterizans .	Blood.		
Laverania malariæ	Anopheles varuna	Body.		
Plasmodium malariæ	Anopheles varuna	Body.		
Plasmodium vivax	Anopheles varuna	Body.		
Fam. THELLERIIDÆ (p. 25).				
Theileria hirci	Ovis sp	Blood.		
Theileria sp	Bos indicus	Blood.		
Fam. BABESIIDÆ (p. 25).				
Babesia bovis	Bos indicus	Blood.		
Babesia taylori	Capra hircus	Blood.		
INCERTÆ SEDIS (p. 26).				
Anaplasma marginale	Bos indicus	Blood.		
Bertarellia calotis	Calotes versicolor major.	Blood.		
Bertarellia sp	Lissemys punctata	Blood.		
CHIDOGROPHIA	granosa.			
CNIDOSPORIDIA.				
MYXOSPORIDIA.				
Fam. CERATOMYXIDÆ (p. 2	6).			
Ceratomyxa hilsæ	Hilsa ilisha	Gall-bladder.		
Fam. MYXIDIIDÆ (p. 26).				
Myxidium glossogobii	Glossogobius giuris	Gall-bladder.		
Fam. MYXOBOLID & (D. 26).				
Murobolus calbasui	Labeo calhasu	Gall-bladder.		
	Labeo rohita	Gall-bladder.		
Myxobolus mrigalæ	Cirrhina mrigala	Scales.		
HAPLOSPORIDIA.				
INCERTÆ SEDIS (D. 27).				
Rhinoeporidium equi	Emus caballus	Nasal cavities.		
Rhinosporidium sp.	Bos indicus	Nasal cavities.		
(ii) Supplementary	List of Hosts and the	ir Parasites.		
Host.	Parasite.	Seat.		
MAMMALIA (DD. 27-9).				
Axis axis	Eimeria wassilewskui	Alimentary canal.		
Bos bubalus	Eimeria smithi	Alimentary canal.		
Bos indicus	Eimeria culindrica	Alimentary canal.		
200 ///////////////////////////////////	Eimeria smithi	Alimentary canal.		
	Eimeria zürnii	Alimentary canal.		
	Theleria sp	Blood.		
	Babesia bovis	Blood.		
	Anaplasma marginale .	Blood.		
Deserbalas increases here	Kninosporidium sp.	Ricod		
Doseg navus vragocameius .	Rimenia antoinai	Alimentary canel		
Cupia hurun	Rabesia taulori	Blood.		
Equus cabalus	Rhinosporidium equi .	Nasal cavities.		
Ovis sp	Theileria hirci	Blood.		
-				

Bosephalus tragocamelus .	Eimeri
Capra hircus	Eimeri
Equus cabalus	Rhinos
Ovis sp	Theiles
#### Parasite. Seat. Host. Aves (pp. 29-31). Ardeola grayii Leucocytozoon ardeolæ. Blood. . . . . . . . . Butastur teesa ..... Toxoxlasma butasturis. Loucoevtes. Blood. Centropus sinensis parroti Proteosoma centropi ... Chloropsis jerdoni ..... Leucocytozoon enriquesi Blood. Columba livia ..... Eimeria labbeana .... Intestine. Blood. Halcyon smyrnensis fusca. Hæmoproteus (?) halcyonis fuscæ. Hæmoproteus (?) lanii . Blood. Lanius schach erythronotus Molpastes cafer cafer . . . . Leucocytozoon molgas-Blood. tis. Oriolus oriolus kundoo .... Leucocytozoon (?) sp. ... Blood. Oriolus xanthornus xan-Leucocytozoon (?) sp. .. Blood. thornus. Pond heron ..... Proteosoma heroni .... Blood. Thereiceryx viridis ..... Hæmoproteus (?) therei-Blood. cerucis. REPTILIA (pp. 32-3). Calotes versicolor major ... Bertarellia calotis .... Blood. Lissemys punctata granosa Blood. Bertarellia sp..... Natrix piscator ...... Eimeria gupti ..... Rectum. Passerita mycterizans .... Protesoma passeritæ ... Blood. PISCES (p. 34). Cirrhina mrigala ..... Myxobolus mrigalæ ... Scalos. Glossogobius giuris ..... Myxidium glossogobii . Gall-bladder. Hilsa ilisha ..... Ceratomyxa hilsæ .... Gall-bladder. Gall-bladder. Labeo calbasu..... Myxobolus calbasui ... Labeo rohita ..... Myxobolus calbasui ... Gall-bladder. Otolithus maculatus ..... Henneguya otolithus... Bulbus arteriosus. INSECTA (p. 35). Laverania malariæ ... Anorheles varuna ..... Body. Plasmodium malariæ . Body. Plasmodium vivax ... Body. CHÆTOPODA. POLYCHÆTA (p. 36). Amphinome rostrata Selenidium amphinomi. Cœlomic cavity. . . . Eunice siciliensis ..... Stomatophora primitiva Intestine. Lecudina eunicæ ..... Intestine. Ulivina eunicæ..... Intestine. Deuteromera cleava ... Intestine. Contortiocarpa pra-Intestine. shadi.

Lecudina lysidicæ ....

Intestine.

Lysidice collaris

. . . . . . .

ADDENDA.

### ADDENDA.

Genus **STOMATOPHORA** Drzewecki, 1907, emend. Hesse, 1909, and Bhatia, 1924.

(Pages 68-72.)

308. Stomatophora primitiva Bhatia & Setna. (Fig. 183.)

†Stomatophora sp., Bhatia & Setna, 1935, p. 312. †Stomatophora primitiva, Bhatia & Setna, 1938 (in press).

Trophozoite almost spherical, with a cup-like sucker near the anterior end. The sucker is not provided with a central



Fig. 183.—Stomatophora primitiva Bh. & Set. (After Bhatia and Setna.)

mucron. Numerous epicytal striations radiate outwards over the body for some distance round the sucker. Nucleus rounded, with a large spherical karyosome. Gametocysts slightly ellipsoidal.

Dimensions.—Length of trophozoite  $222 \mu$ , width  $203.5 \mu$ ; nucleus  $44.4 \mu$  by  $37 \mu$ : gametocyst measures  $222 \mu$  by  $203.5 \mu$ .

Remarks.—All the previously known species of the genus Stomatophora and of other genera included in the family Stomatophoridæ are parasites of earthworms belonging to the genus *Pheretima*. The present species is the first record from a polychæte. It recalls in its structure S. simplex Bhatia, 1924, from which it differs in the absence of a central mucron in the sucker. Seen under the oil-immersion lens the sucker is seen to be surrounded by a clear area, and the surrounding epicytal striations appear to run into each other and interlace.

The sucker was seen to be retained in the case of one of the gametocytes in a gametocyst.

Habitat.—Intestine of Eunice siciliensis Grube : ANDAMAN ISLANDS, Port Blair.

#### Genus LECUDINA Mingazzini, 1891.

#### (Pages 83-5.)

Key to Indian Species.

1 (3).	Cytoplasm of the anterior portion of the body differentiated	2.
2.	Trophozoite cylindrical; epimerite variable in form, with an anchor plate at the end.	[Aiyar, p. 83. L. hrasili Ganan, &
3 (1).	Cytoplasm of the anterior portion of the body not differentiated	4.
4 (5).	Trophozoite elongate oval ; epimerite knob-like	[p. 378. L. eunicæ Bh. & Set.,
5 (4).	Trophozoite oval; epimerite a long conical trunk	[p. 379. L. lysidicæ Bh. & Set.,

### 309. Lecudina eunicæ Bhatia & Setna. (Fig. 184.)

†Lecudina eunicæ, Bhatia & Sotna, 1938 (in press).

Trophozoite elongate oval, widest in the middle, narrowing at both ends, bluntly pointed anteriorly, and more sharply



Fig. 184.—Lecudina eunicæ Bh. & Set. (After Bhatia and Setna.)

pointed posteriorly. Epimerite forming a knob-like structure, with its cytoplasm differentiated from that of the rest of the body. Nucleus large, subspherical, with a single karyosome.

Dimensions.—Length of the trophozoite  $475 \mu$ , maximum width  $94.5 \mu$ ; epimerite  $14 \mu$  in length; nucleus  $38.5 \mu$  by  $28 \mu$ .

*Remarks.*—The parasite somewhat resembles *L. elongata* (Mingazzini) in the differentiation of its anterior end, which is described by Mingazzini as a small spherical button, but according to Reichenow (1932) an epimerite is absent, and the

ADDENDA.

parasite attaches itself to the epithelial cells of the host by its differentiated anterior portion in a sucker-like manner. L. eunicæ differs from L. elongata in the form of the trophozoite and the structure of its nucleus.

Habitat.—Intestine of Eunice siciliensis Grube : ANDAMAN ISLANDS, Port Blair.

### 310. Lecudina lysidicæ Bhatia & Setna. (Fig. 185.)

†Lecudina lysidicæ, Bhatia & Setna, 1938 (in press).

Trophozoites oval, broadly rounded anteriorly and narrower and pounded off posteriorly, broadest in the anterior third of its body. Cytoplasm does not show any differentiation in the anterior portion of the body. Epimerite in the form of a long conical trunk. Nucleus spherical, oval or irregularly quadrilateral, with a large central karyosome. Sporonts much larger in size, without an epimerite, but showing a triangular area slightly raised from the anterior end of the body.



Fig. 185.—Lecudina lysidicæ Bh. & Set. (After Bhatia and Setna.)

Dimensions.—Cephalonts with a total length of 71.9– 185 $\mu$ , maximum width 14.8–62.9 $\mu$ , length of epimerite 11.1– 37 $\mu$ , and nucleus 14.8 $\mu$  by 10 $\mu$ . Sporonts may reach a length of 370 $\mu$ , with a maximum width of 155.4 $\mu$ .

Remarks.—The parasite invites comparison with L. aphroditæ (Lank.), but differs in its much smaller size, in the nuclear structure, and also in the epimerite not being marked with successive constrictions. The nucleus varies considerably in form and position. It may be spherical, oval or irregularly quadrilateral in form, and may be situated in the anterior middle or posterior part of the body, but it always presents the same structure. The central karyosome is surrounded by a clear area, and chromatin particles are arranged in a peripheral zone within the nuclear membrane. The sporonts frequently and the cephalonts occasionally are seen to be full of elongated spindle-shaped bodies, each containing a definite central nucleus; these are probably algæ, and measure up to  $74\mu$  by  $3\cdot7\mu$ .

Habitat.—Intestine of Lysidice collaris Grube : ANDAMAN ISLANDS, Port Blair.

### Family GREGARINIDÆ Labbé, 1899.

(Pages 96-107.)

#### Genus ULIVINA Mingazzini, 1891.

Ulivina, Mingazzini, 1891, p. 235; Labbé, 1899, p. 34; Minchin, 1903, pp. 203, 325.

Doliocystis (part), Crawley, 1903, p. 56.

Ulivina. Saint-Joseph. 1907, pp. 164, 174. Doliocystis (part), Ellis, 1913, p. 287.

Ulivina, Kamm, 1922, pp. 28-9; Reichenow, 1929, p. 893; 1932, pp. 35-6; 1935, p. 369.

Young trophozoites with simple papillate epimerite. Sporonts free, without epimerite. Not forming syzygies. Sporocysts not known.

Remarks.-The genus was based on U. elliptica Mingazzini, 1891, and among the characters mentioned were "external membrane forms a continuous sac round the animal" and " protomerite the more dense." Neither of these characters is now insisted upon. Porter (1899) described an unnamed septate Gregarine from Rhynchobolus, but Crawley (1903), thinking that the part of the animal which Porter took to be protomerite plus epimerite was only the epimerite, and that the Gregarine was a dicystid form, named it as Doliocystis rhyncoboli. Kamm (1922) referred it to Ulivina as a second species in that genus. Reichenow (1932, 1935) has given an amended definition of the genus and considers Sycia inopinata Léger, 1892, as idential with Ulivina elliptica Mingazzini. The generic character that the epimerite is surrounded at its base by a ring-like thickening is based on Léger's description of S. inopinata. This feature is not possessed by U. elliptica, U. rhynchoboli or by the species described below. I therefore regard Ulivina as distinct from Sycia.

311. Ulivina eunicæ Bhatia & Setna. (Fig. 186).

†Ulivina sp., Bhatia & Setna, 1935, p. 312. <sup>†</sup>Ulivina eunicæ, Bhatia & Setna, 1938 (in press).

Trophozoites elliptical, with the protomerite drawn out like a neek and curved backwards. Epimerite slender, wider at its base and tapering into a fine needle. The deutomerite full of inclusions. Length of protomerite to total length as 1: 4.3; width of protomerite to width of deutomerite as 1: 6.3. Nucleus oval and situated in the anterior narrower part of the deutomerite. Sporoeysts not known.

Dimensions.—Cophalont  $340.4 \mu$  in length; epimerite  $14.8 \mu$ in length; protomerite  $77.7 \mu$  in length and  $11.1 \mu$  in maximum width : deutomerite  $247.9\mu$  in length and  $70.3\mu$  in maximum width.

*Remarks.*—The parasite resembles closely *U. rhynchoboli* (Crawley), but the cytoplasm in the protomerite is not more dense than in the deutomerite, and the nucleus is oval and not spherical as in that species. Also there is no external membrane continuous around the animal.

Habitat.—Intestine of Eunice siciliensis Grube : ANDAMAN ISLANDS, Port Blair.



Fig. 186.—Ulivina eunicæ Bh. & Set. (After Bhatia and Setna.)

#### Genus DEUTEROMERA Bhatia & Setna, 1938.

Deuteromera, Bhatia & Setna, 1938 (in press).

Sporonts solitary. Epimerite subconical, apex cup-shaped. Protomerite and deutomerite showing incomplete secondary segmentation.

### 312. Deuteromera cleava Bhatia & Setna. (Fig. 187).

†Septate Gregarine, Bhatia & Setna, 1935, p. 312.

†Deuteromera cleava, Bhatia & Setna, 1938 (in press).

Body elongate oval, distinctly septate. Epimerite simple,

elongated and subconical, with its apex somewhat cup-shaped, marked with distinct epicytal striations. Protomerite not more densely granular than the deutomerite, broadest at its base, width usually greater than its length, showing a somewhat obliquely running furrow. Protomerite, together with the epimerite, bent at an angle on the deutomerite. Deutomerite widest anteriorly, and narrower and rounded posteriorly, its maximum width may be somewhat greater than the length ; marked by two incomplete septa running inwards from one side. Length of the protomerite to total length as 1:3.8, width of the protomerite to width of the deutomerite as 1:1.3. Nucleus large, oval, situated near the posterior end, containing a single large, spherical, eccentrically placed karyosome, surrounded by a narrow clear area and densely packed



Fig. 187.—Deuteromera cleava Bh. & Set. (After Bhatia and Setna.)

chromatin granules. Sporont solitary, proportionately narrower and much more elongated than the cephalont; may be bent upon itself, and the deutomerite may show incomplete septa. Nucleus in the sporont may be oval or spherical, but is similar in structure to that in the cephalont. Cyst- and spore-formation not known.

Dimensions.—Cephalont up to  $402 \cdot 5\mu$  in total length; epimerite  $105\mu$  in length and  $77\mu$  in maximum width; protomerite  $105\mu$  in length and  $147\mu$  in maximum width; deutomerite  $192 \cdot 5\mu$  in length and  $203\mu$  in maximum width, nucleus  $52 \cdot 5\mu$  by  $45 \cdot 5\mu$ . Sporont up to  $420\mu$  in total length and  $105\mu$  in maximum width, nucleus  $42\mu$  in diameter.

Remarks.--The furrow on the protomerite and the incomplete septa and a furrow in the deutomerite strongly recall the more complete segmentation of the posterior part of the deutomerite in *Metamera schubergi* Duke, 1910, known from the intestine of certain leeches, and the segmentation of both protomerite and deutomerite in *Tæniocystis légeri* Cognetti, 1911, from the cœlom of an oligochæte, but there are various important differences.

Habitat.—Intestine of Eunice siciliensis Grube : ANDAMAN ISLANDS, Port Blair.

#### Genus CONTORTIOCORPA Bhatia & Setna, 1935.

Contortiocorpa, Bhatia & Setna, 1935. p. 312; 1938 (in press). Sporont solitary. Body spirally twisted upon itself.

### 313. Contortiocorpa prashadi Bhatia & Setna. (Fig. 188.)

†Contortiocorpa prashadi, 1935, p. 312; 1938 (in press).

Sporont solitary. Body elongate oval, broadest at about one-third the length of the body from the anterior end, rapidly



Fig. 188.—Contortiocorpa prashadi Bh. & Set. (After Bhatia and Setna.)

narrowing behind the middle, and posteriorly drawn out into a narrow rounded tip. It is generally twisted upon itself, presenting a number of turns of a spiral and a number of marginal projections where the spiral is turning round to run over the other surface. Nucleus oval or spherical, with a single karyosome, generally situated in the anterior half of the body. Some individuals are also found in an untwisted or only partially twisted condition.

Dimensions.—Length of the body in a twisted individual  $318\cdot5\mu$ , maximum width  $87\cdot5\mu$ , diameter of the nucleus  $37\cdot5\mu$ . A partially twisted individual measured  $398\cdot4\mu$ , with a maximum width of  $170\cdot2\mu$ .

*Remarks.*—In addition to the typical twisted individual there are in the preparations a number of partially twisted or untwisted individuals which are believed to be of the same species. One such individual shows an indication of a narrow protomerite and a retracted bluntly conical epimerite. Typical

cephalonts have not been met with, nor have we come across any cysts or spores which may be definitely assigned to this species.

Habitat.-Intestine of Eunice siciliensis Grube : ANDAMAN ISLANDS, Port Blair.

## Family SELENIDIIDÆ Brasil, 1907.

The family includes a single genus.

### Genus SELENIDIUM Giard, 1884, emend. Brasil, 1907, and Ray, 1930.

Selenidium, Giard, 1884, p. 192. Polyrhabdina, Labbé, 1899, p. 48.

 Selenidium, Caullery & Mesnil, 1899, pp. 80–99; Minchin, 1903,
 p. 204; Brasil, 1907, pp. 370–97; Brasil & Fantham, 1907,
 p. 518; Wenyon, 1926, pp. 1127, 1136–8; Reichenow, 1929,
 pp. 876–7; Ray, 1930, pp. 370–98; Reichenow, 1932, pp. 22–5; 1935, p. 366.

Trophozoites elongate, vermiform, very narrow and cylindrical or wider and more or less flattened, with longitudinal myonemes along the entire length of the body. The anterior end of the body is provided with a small knob-like organ of and usually contains characteristic chromatic fixation. bodies. Schizogony, where known, takes place during the intracellular condition of the parasite. Gametocyst, where known, contains many occysts, each containing either four or eight sporozoites.

Remarks.--Ray (1930) has re-studied several imperfectly known species of this genus and described several new ones. He has shown that intracellular schizogony does not normally occur in the majority of species studied by him, and is, in fact, known to occur in two species only. He lays stress on the occurrence, in all the species examined and at all stages of their development, of characteristic chromatic bodies at the anterior end of the animal. These are usually thread-like, sometimes club-shaped, but always of a definite type and length in any particular species, and usually fairly constant in number. Very little is known about sporogony in the species of this genus. In two species gametocytes were seen by Caullery and Mesnil in association in the gut, and according to them the attachment was by their anterior ends. Ray found that in the species examined by him the associates become attached by their posterior ends. Spores had been previously seen in one species only, and were known to contain four sporozoites. Ray found gametocysts and spores in two species, and the spore contained four sporozoites in one species and eight

in the other. In view of the above-mentioned considerations, he came to the conclusion that the genus requires drastic revision, and will probably have to be dismembered. He also supports the view, previously held by Mesnil (1899) and Keilin (1923), that the SCHIZOGREGARINARIA are a heterogeneous and artificial group, and that certain genera now placed therein will some day be transferred to the EUGREGARINARIA.

314. Selenidium amphinomi Bhatia & Setna. (Fig. 189.)

†Nematocystis sp., Bhatia & Sotna, 1935, p. 312.
†Selenidium amphinomi, Bhatia & Setna, 1938 (in press).

Trophozoites elongate, vermiform, wider anteriorly, and narrower and tapering posteriorly. Anterior end provided



Fig. 189.—Selenidium amphinomi Bh. & Set. (After Bhatia and Setna.)

with a conical knob-like projection which usually does not show the chromatic bodies. The body is more or less circular in transverse section and the surface is marked by about sixteen longitudinal striations. Nucleus spherical or sub spherical, situated in the broader anterior part of the body, with a large central karyosome. Larger individuals, apparently gametocytes, show a somewhat flattened body, with the anterior end drawn in but still differing in its appearance from the rest of the body, and the posterior end is wider than in the younger trophozoites and narrows gradually to a spon. 2 o point. The myoneme striations are more numerous and may be about twenty in number. The nucleus in these specimens is subspherical or oval, and is placed with its long axis along the length of the body. Association of the individuals is by their anterior ends. Gametocysts are subspherical or oval. Spore-formation was not observed.

Dimensions.—Trophozoites  $126\cdot8-253\cdot6\mu$  in length, with a maximum width ranging from 10 to  $25\cdot3\mu$ ; epimerite  $4\cdot7 9\cdot5\mu$  in length and about the same in its width. Gametocysts are  $47\cdot1-81\cdot6\mu$  in length by  $45\cdot5-62\cdot8\mu$  in width.

Remarks.—Bhatia and Setna (1938) have not found any evidence of schizogony in this species, but their observations are based on the examination of smears only. The chromatic threads or bodies, on the occurrence of which during all stages Ray (1930) lays so much stress, were not found in many specimens. One specimen, however, showed a single deeplystained thread and two other specimens showed a varying number of chromatic dot-like bodies. The association of the individuals was found to be by their anterior ends, as was described by Caullery and Mesnil (1899) in Selenidium echinatum Caullery & Mesnil, and contrary to what Ray (1930) found in other species. A few gametocysts were encountered, but none contained ripe oöcysts.

Habitat.—Cœlomic cavity of Amphinome rostrata (Pallas): taken off Port Blair, ANDAMAN ISLANDS.

### Family EMERIIDÆ Léger.

#### Genus EIMERIA Aime Schneider, 1875.

(Pages 173–97.)

#### [106. Eimeria gupti, nom. nov.

*Eimeria cylindrica*, Ray & Das-Gupta, 1936 a, p. 345.

A new species was recorded by Ray and Das-Gupta from Natrix piscator and named E. cylindrica (see p. 179). As the name is pre-occupied for E. cylindrica Wilson from cattle, the species found by Ray and Das-Gupta is re-named E. gupti.]

#### 315. Eimeria arloingi (Marotel).

Coccidium arloingi, Marotel, 1905, p. 52.

*Eimeria arloingi*, Martin, 1907, p. 6; Stevenson, 1911, pp. 365–9, pls. xix, xx; Speigl, 1919, p. 451; Velu, 1919, pp. 298–301; Lerche, 1921, pp. 380–99, pl. xviii; Schein, 1921, pp. 380–2.

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*Eimeria faurei*, Nöller, Schürjohann, & Vorbrodt, 1922, 15 pp., 7 figs.; Wenyon, 1926, pp. 843-4; Knowles, 1928, p. 364: Reichenow, 1929, pp. 945-6. Eimeria arloingi, Reichenow, 1935, p. 374. †Eimeria arloingi, Taylor, 1938, p. 42.

Obcysts measure  $21-33 \mu$  by  $16\cdot 5-22\cdot 5 \mu$ .

Remarks .--- Nöller, Schürjohann, and Vorbrodt (1922), as a result of cross-infection experiments with the Coccidia of goats and sheep, established the identity of E. arloingi of goats with E. faurei of sheep, and since then the two have been regarded as identical. Taylor (1938) has recently reported that two strains of Eimeria, viz., E. arloingi and E. faurei, have been isolated from local goats at Muktesar, and experiments are being conducted to test the pathogenesis of these two species in goats and sheep. A strain of these parasites is claimed to have been established by feeding sporulated oöcysts to goats and sheep.

E. arloingi is considered as highly pathogenic and was recently found in an outbreak among goats and sheep at Etah breeding farm.

Habitat.—Fæces of goats and sheep: UNITED PROVINCES. Muktesar, Etah.

### 316. Elmeria cylindrica Wilson.

Eimeria cylindrica, Wilson, 1931, pp. 1-44, 7 pls. +Eimeria cylindrica, Taylor, 1938, p. 42.

Occysts approximately cylindrical, with uniformly thick walls,  $19.4-26.8\mu$  in length by  $11.9-14.9\mu$  in width, average  $23 \cdot 3 \mu$  by  $13 \cdot 3 \mu$ . Ratio of length to width  $1 \cdot 65$ .

Habitat.--Fæces of hill bulls : UNITED PROVINCES, Muktesar.

### 317. Eimeria labbeana Pinto.

Eimeria pfeifferi, Labbé, 1896, p. 539; 1899, p. 59; Nieschulz, 1921, pp. 71-82, pl. iv; 1925, pp. 479-94, pls. xvii-xx; Wenyon, 1926, pp. 857-8; Nöller, 1928, p. 195. Eimeria labbeana, Pinto, 1928, pp. 1564-5. Eimeria pfeifferi, Reichenow, 1929, p. 948. †Eimeria labbeana, Taylor, 1938, p. 42.

Oöcysts spherical, without a micropyle, and without a residual body. Sporocysts with one end pointed and the other end rounded, with a refractile knob at the pointed end. The sporocyst contains a large residuum.

Dimensions.—Occysts measure  $15-26\mu$  in length by  $14-24\mu$ in breadth. Sporocysts measure on an average  $12.5\mu$  by 6.5µ.

. Remarks.—Nieschulz (1921) at first failed to infect chicks with the Coccidium of the pigeon, but later (1925) succeeded in doing so. Wenyon (1926) doubts if E. pfeifferi (Labbé) of the pigeon is distinct from E. avium (Rivolta & Silvestrini) of the chick. Pinto (1928) has proposed a new name, E. labbeana, for Coccidium pfeifferi Labbé and Eimeria pfeifferi Labbé.

### Genus PROTEOSOMA Labbé, 1894.

(Pages 247–59.)

# 318. Proteosoma passeritæ (de Mello & da Fonseca). (Fig. 190.)

†Plasmodium passeritæ, de Mello & da Fonseca, 1938, pp. 47-8, pl. vi.

Ring-forms with the cytoplasm very compact or more or less vacuolated, with small dots of pigment scattered all over the body. Schizonts minute, pyriform or amœboid, devoid



Fig. 190.—Proteosoma passeritæ (de Mello & da Fonseca). A, B, ring forms; C, D, schizonts; E, cruciform rosette. (After de Mello and da Fonseca.)

of pigment, which may even be absent in larger roundish forms. Chromatin divides into two, three or four granules, thus determining the number of resulting merozoites, which may be arranged in a cross-like manner. Infected red corpuscles do not show any alteration.

Dimensions.—Ring-forms  $1-3\mu$ , rosettes  $3-4\mu$ .

Remarks.—Apart from the fact that the rings are pigmented, the resemblance to Babesia quadrigemina (Nicolle) is very striking, as is also the case with *P. minasense* (Carini & Rudolph) from certain lizards.

Habitat.—Blood of the green snake, Passerita mycterizans Daud.: PORTUGUESE INDIA, Nova Goa.

#### Genus ANAPLASMA Theiler, 1910.

(Pages 325–6.)

#### 319. Anaplasma marginale Theiler.

"Marginal points," Smith & Kilborne, 1893.

Anaplasma marginale. Theiler, 1910, pp. 135-7; Wenyon, 1926, pp. 1053-6.

†Anaplasma sp., Ware, 1932, pp. 31-2.

†Anaplasma marginale, Ware, 1938, p. 212.

Spherical granule, varying in size from 0.1 to  $0.5\mu$ ; staining bright red with Romanowsky's stain, and situated near the margin of the red blood-corpusele.

*Remarks.*—Anaplasmata have been encountered in the blood of hill bulls at Muktesar and the animals were regarded as "carriers." Dias and Aragão (1914) claimed to have brought about the production of these bodies in cattle by the simple injection of certain poisonous substances such as trypan blue. Du Toit (1928) expressed the view that the production of true Anaplasmata in cattle by repeated injection of trypan blue can only be brought about when the cattle are "carriers," the injections causing a breakdown in their immunity, with the appearance of parasites in their blood. Ware (1932) reported the appearance of these parasites in hill bulls after a series of trypan blue injections.

Habitat.—Blood of cattle : UNITED PROVINCES, Muktesar ; CENTRAL PROVINCES.

#### Genus BERTARELLIA Carini, 1930.

(Pages 326-7.)

#### 320 Bertarellia carinli de Meyrelles.

Bertarellia carinii, de Meyrelles, 1938, pp. 49-53, figs. 1 & 2.

Usually roundish or elliptical, rarely oval, pyriform or ringlike bodies stained violet with Leishman's or May-Grunwald Giemsa's stain. Each such chromatic point or anaplasmoid body is surrounded by a clear halo, often difficult to distinguish, and roundish or elliptical in form. The parasite may be central, polar or peripheral in position in the red blood-cells, or may even be found free in the plasma. The red cell may contain more than one parasite, and the halolike portion may contain two or even three chromatic granules, suggesting binary division.

*Remarks.*—The organism differs from *B. calolis*, as the cytoplasm is never stained blue as in that species. It is, however, identical with the parasite found in blood-films from two Brazilian tortoises.

Habitat.—Blood of the Indian tortoise, Lissemys punctata granosa Smith : PORTUGUESE INDIA, Nova Goa.

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The Bibliography is divided into several sections, viz., Text-books and Sporozoa in General; GREGARINIDA; COCCIDIA; HÆMOSPORIDIA; CNIDOSPORIDIA; SARCOSPORIDIA; and HAPLOSPORIDIA. If a reference is not found under "Text-books and Sporozoa in General," it should be looked for under the particular Order to which the work relates.

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## Hæmosporidia.

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### PLATE I.

- Fig. 122. Laverania malarize Grassi & Feletti.  $(\times 2000.)$ A, young "ring" form; B, accolé form; C, multiple infection with "rings"; D, "tenue" form; E, fully developed schizont; F, "rosette" stage; G, male gametocyte; H, female gametocyte. (Stained with Leishman's stain.) (After Wenyon.)
  - 123. Plasmodium vivax (Grassi & Feletti). ( $\times$  2000.) A, young ring-form; B, two rings in a corpusele; C, trophozoite with pigment in its cytoplasm, red cell enlarged and with Schüffner's dots; D, schizont showing multiplication of nuclei; E, schizont with many nuclei; F, "rosette" stage; G, male gametocyte; H, female gametocyte. (Stained with Leishman's stain.) (After Wenyon.)
  - 124. Plasmodium malariæ (Laveran). ( $\times$  2000.) A, young ring-form; B, C, well-developed trophozoites with pigment in the cytoplasm; D, schizont showing multiplication of nuclei; E, schizont with several nuclei; F, "rosette" stage; G, male gametocyte; H, female gametocyte. (Stained with Leishman's stain.) (After Wenyon.)

A E

E





G





Fig. 123



Fig.124



G

н

F

## PLATE II.

- Fig. 125. Plasmodium cynomolgi Mayer. A, very early non-vacuolated trophozoite and young ring-form;
  B, larger ring-form; C, D, amœboid trophozoite showing pigment and enlargement and stippling of the infected red cell; E, large, solid, presegmenting form; F, early segmenting form; G, H, advanced segmenting forms; I, female gametocyte: J, male gametocyte. (After Mulligan.)
  - 126. Plasmodium inui Halberstädter & Prowazek. A, young ring-form; B, two rings in a corpuscle with double chromatin dots; C, large irregular ringform with a few pigment granules; D, large, solid trophozoite with enlarged nuclear vesicle; E, presegmentation stage; F, early schizont; G, H, mature schizonts; I, J, female and male gametocytes. (After Sinton.)
  - 127. Plasmodium knowlesi Sinton & Mulligan. A, B, young ring-forms with one or two chromatin masses and an accessory chromatin dot; C, larger ring with double chromatin and accessory chromatin dot; D, large trophozoites with dark and coarse pigment; E, F, large, solid, non-vacuolated, presegmenting forms: stippling seen in blood-films stained by the "panoptic" method; G, H, mature schizonts; I, J, female and male gametocytes. (After Mulligan.)



Fig. 126









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