PLASMODIOPHORALES

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THE PLASMODIOPHORALES

Including a
Complete Host Index, Bibliography,
and a Description of Diseases
Caused by Species of this Order

BY

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Dedicated to

R. A. HARPER

On the Occasion of the 80th

Anniversary of His Birthday

PREFACE

This treatise is part of a series of lectures presented to graduate and research students of mycology at Columbia University on the development, origin, and phylogeny of the lower fungi. It had been originally planned to incorporate this material in a general treatment of the simple, biflagellate Oomycete-like fungi, Lagenidiales, and Chytridiales, but inasmuch as the Plasmodiophorales at present appear to be a fairly coherent phylogenetic group, it seems advisable to treat them separately. The Plasmodiophorales are an important and significant group of organisms from the standpoints of plant pathology and phylogeny of the lower fungi. As destructive parasites of crucifers and potatoes, some species cause serious economic losses of basic food crops. Phylogenetically, they possess certain developmental phases which are strikingly similar to those of the Proteomyxa, Myxomycetes, and simple fungi —similarities which suggest either a common origin or parallelism in development.

Although the Plasmodiophorales have been studied for more than a half century, no serious effort to summarize the accumulated data was made until 1933 when Cook monographed the group. Cook gave a detailed description of the known genera and species and also discussed their cytology and development in relation to phylogeny. Unfortunately, this otherwise worthy and excellent treatise is marred by certain inconsistencies, based on the author's observations, which are confusing and misleading to beginners in this field. Since that time, several new genera and species have been added to the group. Particularly significant is the discovery of Ledingham, Couch, et al., and Barrett that the zoospores are biflagellate and heterocont and that thin-walled evanescent zoosporangia are a characteristic developmental phase of the Plasmodiophorales. These discoveries have greatly modified our concepts of the group and make the present revision opportune, worthwhile, and essential.

This book is intended primarily for graduate and research students of mycology and the lower organisms. Nevertheless, botanists and biologists in general as well as protozoologists and phytopathologists will doubtless find the summarized data, life cycle diagrams, and descriptions of diseases of considerable value. As a treatise for research students, it necessarily includes much that is questionable and controversial in nature and which ordinarily might be omitted or discussed more briefly. Some of the data presented are of doubtful value and significance, in the author's opinion, but they are nonetheless included with as little bias as possible in order that students may draw their own conclusions and interpretations. Although the author agrees with Cook and others that *Rhizomyxa, Sorolpidium,* and *Anisomyxa* are probably synonyms of *Ligniera,* these genera are discussed separately as doubtful members. Likewise, full treatment is given to the excluded genera and species, thereby making these data available to research workers. The author's critical attitude and seeming skepticism toward existing data on "akaryosis," extrusion of chromatin, sexuality, meiosis, and other critical developmental phases of this group is not intended as a direct criticism of the veracity and accuracy of certain workers, but rather to indicate how inconclusive present-day knowledge and interpretations are and thereby to stimulate more intensive study of these phases. The Plasmodiophorales are unfavorable for cytological study because of the minuteness of the nuclei. Likewise, the intramatrical habitat of all species makes direct observation of gametic fusion, schizogony, etc., extremely difficult in living material. It is therefore to be expected that many data are conflicting and inconclusive.

Separate bibliographies are provided at the close of each chapter to expedite reference to literature on particular subjects, genera, and species. Since many of the cited papers are general in nature and relate to several genera, they have been listed several times, which makes the bibliography somewhat redundant. A host index is also provided with each species. Due to war conditions abroad, it has been impossible to secure many of the publications relating to club root and powdery scab, so that the host index and bibliography of *Plasmodiophora Brassicae* and *Spongospora subterranea* are unfortunately incomplete. In a bibliography of this magnitude errors are likely to occur, and the author will appreciate having mistakes and omissions called to his attention. The glossary is purposely brief and relates almost entirely to terms used in the text.

The writer has drawn freely from the illustrations of authors in this country and abroad, to whom he is very grateful. The list of contributors is too long for individual mention, but full credit is given in the descriptions of the drawings. The life cycle diagrams presented in Chapters III and V have not been copied directly from other authors' illustrations but are based on their descriptions of the successive developmental phases. The author feels particularly grateful to Miss Amy L. Hepburn, Natural Science Librarian of Columbia University, for her unstinted help with the literature, without which this work would have been impossible.

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CONTENTS

Preface i

CHAPTER I

Introduction 1
Glossary 2
Bibliography 3

CHAPTER II

Cytology 4
"Promitosis" 4
"Akaryote" stage 10
Meiosis 12
Schizogony and Cleavage 14

CHAPTER III

Sexuality and Alternation of Generations 15
Plasmodiophora 15
Tetramyxa 16
Sorosphaera 16
Sorodiscus 18
Spongospora 18
Ligniera 18
Polymyxa and Octomyxa 18
Bibliography 20

CHAPTER IV

Classification and Description of Species 20
Key to genera 22
Plasmodiophora 22
Biological races 26
Relation to bacteria 26
Relation to cancer 28
Bibliography 33
Excluded species 34
Bibliography 36

7533

Tetramyxa 37
Octomyxa 40
Sorosphaera 41
Sorodiscus 46
Membranosorus 52
Spongospora 54
Ligniera 58
Polymyxa 63
Doubtful genera 64
Rhizomyxa 64
Sorolpidium 66
Anisomyxa 68
Trematophlyctis 70
Pyrrhosorus 71
Excluded genera 72
Sporomyxa 74
Peltomyces 76
Cystospora 76

CHAPTER V

Phylogeny and relationships of the Plasmodiophorales 78
Historical 78
Relationships with the Myxomycetes 79
Relationships with the Chytridiales 85
Relationships with *Woronina* 85
Relationships with the Proteomyxa and Protozoa 88
Bibliography 91

CHAPTER VI

Diseases caused by species of the Plasmodiophoraceae 93
Club root of crucifers 93
Losses due to disease 93
Discovery of disease 93
Symptoms 93
Cellular relations between host and pathogen 95
Entrance and spread of parasite 96
Dissemination of parasite in nature 96
Environmental factors 97
Hosts and degree of infection 99
Control of club root 104
Sanitary practices 104
Seed, seed bed, and seedling disinfection 105
Disinfection of fields 105

Liming 110
Basic fertilizers 115
Soil drainage 117
Crop rotation 117
Eradication of wild hosts 118
Resistant varieties of crucifers 118
Nature of susceptibility and resistance 119
Geographical distribution of club root and bibliography 120
Powdery scab of potatoes 129
Significance of disease 129
Predisposing factors 129
Symptoms 130
Cellular relations between host and pathogen 131
Control
Sanitary practices 132
Seed tuber disinfection 132
Soil disinfection 132
Effects of fertilizers 133
Resistant varieties 133
Distribution and bibliography 133

Species Index 137

Subject Index 137

Author Index 139

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Chapter 1

Introduction

The Plasmodiophorales include one family of organisms which are often referred to as parasitic slime molds because they are characterized by a multinucleate plasmodial stage as in the true slime molds and parasitize filamentous fungi, algae, cryptogams, and higher plants. While this common name may be descriptive, its use is unfortunate, since it suggests a relationship with the Myxomycetes which has not been definitely established. Most genera of this order have rather complex life cycles which include zoospores, amoebae, sporangiosori, zoosporangia, secondary zoospores, plasmodia, cystosori, resting spores, and probably isomorphic gametes. Sporangiosori and thin-walled evanescent zoosporangia were first observed by Borzi in *Rhizomyxa hypogea* as early as 1884, and later by Nemec ('11, '13) in *Sorolpidium* and *Anisomyxa*, but at that time the relationship of these genera to the Plasmodiophoraceae was not clearly understood. Zoosporangia were subsequently rediscovered by Cook ('26), Cook and Schwartz ('30), Ledingham ('33, '34, '35, '39), Fedorintschik ('35), Couch, *et al.* ('39) in *Ligniera*, *Plasmodiophora*, *Polymyxa*, *Spongospora*, and *Octomyxa* and are now generally believed to be a characteristic developmental phase of the order as a whole. The zoosporangia are regarded by some workers as gametangia in which meiosis precedes gametogenesis, but this has not been conclusively proven.

The sporangial phase is followed by the development of a comparatively large multinucleate sporogenous plasmodium in which meiosis is reported to occur before or during cleavage into resting spores. The latter may remain loose and free of each other or unite in more or less compact cystosori. Upon germination, the resting spores produce uninucleate amoebae or motile flagellate zoospores. These cells are regarded by many workers as isomorphic gametes which fuse in pairs and thus initiate the diploid generation, but so little is known about sexuality in this order that nothing conclusive can be said as yet about the sexual nature of these cells. Some mycologists contend that a true plasmodium does not exist in the Plasmodiophorales on the grounds that the naked multinucleate thallus is not formed by the coalescence of numerous mutually attracted amoebae in the manner described by Cienkowski ('63) for the Myxomycetes. In so doing, these mycologists disregard the reports of Woronin ('77), Halsted ('93), Nawaschin ('99), Eyeleshymer ('01), Massee ('08), Osborn ('11), Kunkel ('15), Terby ('24), Jones ('28), Horne ('30), Cook and Schwartz ('30), Milovidov ('31), Ledingham ('39), and others that amoebae as well as small plasmodia coalesce in *Plasmodiophora*, *Spongospora*, *Polymyxa*, etc. Whether or not these reports are accurate may be open to question, because they are not all based on observations of living material. These data nevertheless exist in the literature and must be given serious consideration. Furthermore, the above-mentioned reasons for excluding the term plasmodium from the Plasmodiophoraceae would also preclude its use in relation to the Myxomycetes according to recent data on this group. Jahn ('11, '36), Skupienski ('28), Wilson and Cadman ('28), Cadman ('31), and others have shown that the plasmodium is initiated by fusion in pairs of isomorphic gametes and that the zygotes may subsequently ingest unfused haploid amoebae as food material. Thus, the conception of a plasmodium as Cienkowski interpreted it has undergone considerable modification and is now used principally as a descriptive term for the naked, multinucleate, assimilative phase of the slime molds. In this sense it may be equally well employed for the naked multinucleate thallus of the Plasmodiophorales. Cook's use of the term myxamoeba for this stage is unfortunate, misleading, and obviously unwarranted. According to standard dictionaries and glossaries, the term myxamoeba relates to the naked, amoeboid, and usually uninucleate protoplasts formed by the germinating resting spores of the Myxomycetes, and its introduction as a descriptive name for the naked multinucleate plasmodial stage of the Plasmodiophorales will lead to nothing but confusion. Likewise, his use of the term "swarm cells" for the products of spore germination as a distinctive contrast to the name "zoospores" for the flagellate cells formed in zoosporangia is not warranted at present and should be avoided. Ledingham and Barrett have clearly shown that the zoospores are biflagellate and heterocont regardless of whether they are formed in zoosporangia or from resting spores and that there are no structural distinctions between the so-called swarm cells and zoospores. If in the future it is found that the resting spores form gametes and the sporangia zoospores, or vice versa, the two products may then be distinguished and designated as gametes and zoospores, respectively.

Although most species of this order, except *P. Brassicae* and *S. subterranea*, appear to be comparatively rare in occurrence, they are nevertheless world wide in distribution and have been reported from North and South America, Africa, Europe, Asia, Australia and several Atlantic and Pacific islands. Three species occur in fungi, algae, and cryptogams, while the remainder parasitize higher plants. All species, except members of the genus *Ligniera*, cause distortion of the host and marked changes in its cells. These changes involve enlargement and divison of infected as well as of adjacent

healthy cells, with the result that conspicuous excrescences and galls are usually formed. However, only two species are economically important as parasites. *Plasmodiophora Brassicae* and *Spongospora subterranea* are destructive pathogens of crucifers and potatoes, respectively, and cause the diseases commonly known as club root and powdery scab.

While these diseases had been recognized since early times, their causative agents were not identified until the latter part of the 19th century. The discovery of *P. Brassicae* in hypertrophied roots of crucifers by Woronin in 1877 may be said to have initiated the study of the Plasmodiophorales as a distinct group of organisms. A second genus, *Tetramyxa*, was found by Goebel in 1884, and in the same year Zopf created a new family, Plasmodiophoraceae, in the zoosporic Monadineae to include these genera. Two additional genera, *Spongospora* and *Sorosphaera* were reported by Brunchorst and Schroeter in 1886, but the relationship of the former genus was not generally recognized until much later. Schroeter ignored Zopf's classification and created a new order, *Phytomyxini*, with one family, Phytomyxaceae, to include these genera as well as the legume tubercle organism which he redescribed as *Phytomyxa leguminosarum*. Inasmuch as Schroeter's Phytomyxinae was later ('97) incorporated in Engler and Prantl's Die Natürlichen Pflanzenfamilien, it was widely recognized and accepted. *Phytomyxa* as well as *Plasmodiophora Alni* and *P. Elaeagni* were excluded by Tubeuf and Smith ('97) and other pathologists in their discussions of the parasitic slime molds, but Schroeter's order and family names nonetheless continued to be used. In 1909 Maire and Tison made an extensive review and study of these doubtful species and showed again that *P. leguminosarum*, *P. Alni*, *P. Elaeagni*, *Tylogonus Agavae*, and *Pseudocommis Vitis* have little or nothing in common with the true plasmodiophoraceous species. Since *Phytomyxa* had already been excluded, they pointed out that the name Phytomyxaceae was no longer appropriate. They accordingly adopted Zopf's Plasmodiophoraceae to include *Plasmodiophora*, *Tetramyxa*, and *Sorosphaera* and listed Schroeter's Phytomyxinae pro parte and Delage's Protomyxideae zoosporideae as synonyms. Apparently unaware of Maire and Tison's studies, some protozoologists nevertheless still continue the use of Schroeter's Phytomyxinae or some modification of this name.

In the meantime, *Sporomyxa* and *Peltomyces* had been added to the group, and following Maire and Tison's first paper, *Ligniera*, *Molliardia*, *Sorodiscus*, *Ostenfeldiella*, *Cystospora*, *Trematophlyctis*, *Clathrosorus*, *Membranosorus*, *Polymyxa* and *Octomyxa* were successively discovered and included in the Plasmodiophoraceae. However, many of these genera have either been merged or excluded entirely, so that the order includes at present comparatively few valid genera. The group as a whole was finally raised to ordinal rank by Cook ('28, '33), following a suggestion made by Schwartz in 1914.

Taxonomically, the Plasmodiophorales have been bandied back and forth by protozoologists and mycologists for more than half a century, and few workers are in agreement about the taxonomic position and relationships of this order. Its members have been included at various times in the Myxetozoa, Monadineae, Proteomyxa, Rhizopoda, and Chytridiales. Some myxologists, particularly Gwynne-Vaughan, Barnes, and Cook ('33), have maintained that the Plasmodiophoraceae are not fungi and have arisen along independent lines from more primitive forms. However, the rediscovery within the last two decades of zoosporangia in this order and the observations that biflagellate heterokont zoospores are produced in such sporangia and also from resting spores indicate a closer affinity with the simple fungi than was formerly believed to exist.

Glossary

Akaryote stage, a nuclear stage in which little or no chromatin is visible in the nucleus.

Binuclearity hypothesis, the theory that the micro- and macronuclei of infusoria contain the idio- and trophochromatin, respectively, and that the ordinary nucleus of higher forms is accordingly a dual "amphinucleus."

Blepharoplast, the basal granule at the point of insertion of each flagellum.

Capillitium, sterile filamentous, simple, branched, or net-like tubes or fibers formed among spores in a sporogenous body.

Chromidia, trophochromatin granules which are extruded from the nucleus into the cytoplasm.

Chromidia hypothesis, the theory that the nuclei of rhizopods and other similar organisms contain idio- and trophochromatin, the latter of which is extruded into the cytoplasm as chromidia and degenerates or plays a dominant role in the differentiation of specialized structures.

Chromidial stage, a nuclear stage during which the trophochromatin is extruded into the cytoplasm.

Crueiform stage, equatorial ring stage of promitosis in the Plasmodiophorales during which the nucleole is elongate and forms a cross with the chromatin ring.

Cystosorus, a more or less compact aggregate of cysts or resting spores.

Eucarpic, only a portion of the thallus transformed into a reproductive organ; remainder of thallus vegetative.

Extramatrical, outside of host, matrix, or substratum.

Double-anchor stage, anaphase stage of promitosis in the Plasmodiophorales during which the arched daughter chromatin bands and nucleoli are connected by a chromatic strand and form a figure resembling a double anchor.

Dumb-bell stage, more or less synonymous with double-anchor stage of promitosis.

Flagellum, a whip-like protoplasmic organ of locomotion of zoospores, swarmspores, and motile gametes.
Gametangium, a differentiated sac or vesicle which produces gametes.
Garland stage, a prophase stage of meiosis in which the chromatin is aggregated as garlands at the nuclear poles.
Heterocont, (flagella) of unequal length.
Holocarpic, entire thallus transformed at maturity into a reproductive organ.
Hyperplasy, abnormal growth of tissue resulting from undue cell division.
Hypertrophy, abnormal enlargement of an organ.
Hypoplasy, defective development due to insufficient nourishment and consequent cessation of growth.
Homothallic, gametophytic or haploid thalli bisexual.
Heterothallic, gametophytic or haploid thalli unisexual.
Homophytic, sporophytic or diploid thalli bisexual.
Heterophytic, sporophytic or diploid thalli unisexual.
Haplomonoecious, haploid generation bisexual = homothallic.
Haplodioecious, haploid generation unisexual = heterothallic.
Diplomonoecious, diploid generation bisexual = homophytic.
Diplodioecious, diploid generation unisexual = heterophytic.
Haplosynoecious, haploid generation bisexual = homothallic = haplomonoecious.
Haploheteroecious, haploid generation unisexual = heterothallic = diplodioecious.
Diplosynoecious, diploid generation bisexual = homophytic = diplomonoecious.
Diploheteroecious, diploid generation unisexual = heterophytic = diplodioecious.
Indiochromatin, generative chromatin which is concerned with reproduction.
Intramatrical, within the host, matrix, or substratum.
Isogamy, fusion of structurally similar gametes.
Isokont, (flagella) of equal length.
Isomorphic, similar in shape and form but not in essential structure.
Karyogamy, fusion of gametic nuclei.
Meront, a uni- or multinucleate product of schizogony.
Planocyte, a motile cell.
Plasmodiocarp, an irregular, sinuous, asymmetrical fruiting body or sporangium of the Myxogastres.
Plasmodium, a naked multinucleate protoplast capable of amoeboid movement.
Plasmogamy, fusion of gametes, followed sooner or later by karyogamy.
Promitosis, a primitive (?) type of intranuclear mitosis in lower organisms which is characterized by ill-defined chromosomes and a large constricting, dividing nucleole.
Protomitosis, a variety of promitosis described by Alexieff in which no clearly defined equatorial plate is formed. The peripheral chromatin instead is distributed in a diffuse fashion between the polar halves of the divided karyosome.
Pseudoplasmodium, a false plasmodium or aggregate of amoebae which retain their individuality; characteristic of the Acrasieae and Labyrinthulae.
Pseudopodium, a temporary protoplasmic extrusion in amoebae and plasmodia which may be retracted or into which the whole mass may move.
Saturn stage, equatorial ring stage of promitosis in the Plasmodiophorales during which the nucleole lies in the center of a ring of chromatin.
Schizogony, a process of simple or multiple division of a schizont.
Schizont, a naked multinucleate vegetative thallus which undergoes simple or multiple division.
Sorocarp, the fruiting structure of the Acrasieae.
Sorus, a group of sporangia or resting spores.
Sporangiosorus, a more or less compact sorus or aggregate of sporangia.
Sporangium, a sac or vesicle which produces spores endogenously.
Sporocyst, a cyst which produces asexual spores.
Sporogonic, relating to spore formation.
Sporont, a thallus destined to form spores.
Synkaryon, the zygotic nucleus following karyogamy.
Thallus, the vegetative body of algae and fungi, without differentiation into root, stem, and leaf.
Transitional stage, a term used by Winge to describe the transition in nuclear structure between promitosis and meiosis in the Plasmodiophoraceae; synonymous to some degree with the so-called akaryote stage.
Trophochromatin, somatic, vegetative chromatin which is active in nutrition.
Zoocyst, a cyst in Monadineae which produces amoeboid or flagellate cells.
Zoosporangium, a sporangium which produces zoospores.
Zygote, the product of gametic fusion.

BIBLIOGRAPHY: INTRODUCTION

Brunchorst, J. 1887. Bergens Mus. Aarberet. 1866: 219.
Borzi, A. 1884. Rhizomyxa, nuovo ficomicete. Messina.
Cadman, E. J. 1931. Trans. Roy. Soc. Edinburgh 57: 93.
Cienkowski, L. 1863. Jahrb. Wiss. Bot. 3: 100.
Cook, W. R. I. 1926. Trans. Brit. Mycol. Soc. 11: 196.
———. 1928. New Phytol. 27: 240.
———. 1933. Arch. Protistk. 80: 179.
———, and E. J. Schwartz. 1930. Philos. Trans. Roy. Soc. London 218B: 283.
Couch, J. N., J. Leilner, and A. Whiffen. 1939. Jour. Elisha Mitchell Sci. Soc. 55: 399.
Esmarch, F. 1924. Die Kranke Pflanze 1: 169.
Eycleshymer, A. C. 1901. Jour. Mycol. 7: 79.
Fedorintschik, N. S. 1935. Summ. Sci. Wk. Inst. Pl. Protect. Leningrad 1935: 69.
Fitzpatrick, H. M. 1930. The lower fungi—Phycomycetes. New York.

Gwynne-Vaughan, H. C. I., and B. Barnes. 1937. The structure and development of the fungi. 2nd ed. Cambridge.
Halsted, B. D. 1893. New Jersey Agr. Exp. Sta. Rept. 1893: 332.
Horne, A. S. 1930. Ann. Bot. 44: 199.
Jahn, E. 1911. Ber. Deut. Bot. Gesell. 29: 231. 1936, Ibid. 54: 517.
Jones, P. M. 1928. Arch. Protistk. 62: 313.
Kunkel, L. O. 1915. Jour. Agric. Res. 4: 265.
Ledingham, G. A. 1933. Phytopath. 23: 20.
———. 1934. Nature 133: 534. 1935, Ibid. 135: 399†.
———. 1939. Canad. Jour. Res. C, 17: 50.
Maire, R., and A. Tison. 1909. Ann. Mycol. 7: 226.
Massee, G. 1908. Jour. Bd. Agric. London 15: 592.
Milovidov, P. F. 1931. Arch. Protistk. 73: 1.
Nawaschin, S. 1899. Flora 86: 104.
Nemec, B. 1911a. Ber. Deut. Bot. Gesell. 29: 48.
———. 1911b. Bull. Int. Empr. Fran. Joseph Acad. Sci. 16: 69. 1913, Ibid. 18: 18.
Osborn, T. G. B. 1911. Ann. Bot. 25: 271, 327.
Schroeter, J. 1886. Cohn's Krypt. Fl. Schlesiens 3: 133.
———. 1897. Engler und Prantl, Die Nat. Pflanzenf. I, 1: 7.
Schwartz, E. J. 1914. Ann. Bot. 28: 227.
Skupienski, F. X. 1928. Acta Soc. Bot. Poloniae 5: 255.
Terby, J. 1924. Bull. Roy. Acad. Belg. 11: 1.
Tubeuf, K. F., and W. G. Smith. 1897. Diseases of Plants. London.
Wilson, M., and E. J. Cadman. 1928. Trans. Roy. Soc. Edinburgh 55: 555.
Woronin, M. 1877. Arb. St. Petersburg Nat. Gesell. 8: 169.
Zopf, W. 1884. Die Pilzthiere oder Schleimpilze. Encyklop. der Naturwiss. 3: 129.

Chapter II

Cytology

"Promitosis"

Cytological studies of the Plasmodiophorales during the past four decades have centered primarily on the type of nuclear division in the plasmodium, the so-called "akaryote" stage, meiosis, karyogamy, schizogony, and cleavage. Nuclear division in the plasmodium has been described by most workers as promitotic and fundamentally similar to that which occurs in the *limax* group of amoebae and other lower organisms. So consistently has this type of division been reported that many students have regarded promitosis as one of the most diagnostic characters of the whole order, and one which distinguishes the Plasmodiophorales from all other fungi and higher plants. Cook ('28) in particular has stressed this character as follows: "The diagnostic feature which characterizes the Plasmodiophorales is their two methods of nuclear division, and failing to show evidence that both promitosis and mitosis occur in the life cycle, and that these two types are separated by a stage in which at any rate part of the chromatin is extruded into the cytoplasm, no new fungus should be included in this group." At the same time, other workers have maintained that these divisions are typically mitotic with well-defined chromosomes, centrosomes, and astral rays. There is thus sharp disagreement concerning karyokinesis in the plasmodium, and inasmuch as the presence of promitosis has been regarded as an index of relationship to the amoeba, a full discussion of the so-called vegetative divisions in the Plasmodiophorales is essential to an understanding of this order.

Nawaschin ('99) was the first to observe the characteristic appearance of these divisions in *Plasmodiophora* and to point out that they are different from those which occur immediately before or during spore formation. He nevertheless described the former mitoses as karyokinetic and regarded ('01) the presence of the two types of division as an indication of nuclear dimorphism—a view much in vogue among the protozoologists of that time. Nawaschin's observation was confirmed by Prowazek ('02, '05), Maire and Tison ('09), Blomfield and Schwartz ('10), Schwartz ('10), Winge ('13) and Lutman ('13) for other species and genera. Prowazek, particularly, and later Blomfield and Schwartz, also stressed the resemblance of the vegetative divisions to those which had been described by protozoologists in certain coccidia and amoebae.

In the meantime, Nägler ('09) had proposed the term promitosis for the type of nuclear division found in *Amoeba froschi, A. lacustris,* etc., which he interpreted to be a transition between amitosis and mitosis. In these divisions neither chromosomes nor well-defined spindles are formed, according to Nägler. Division is intranuclear, and the large endosome or karyosome functions as a division center. The latter elongates, and as it constricts the chromatin aggregates and forms a band across the equator of the nucleus. The karyosome then divides into two bodies, and as these migrate toward the poles the band of chromatin splits lengthwise. Each half accompanies a karyosome to the poles, and both are there incorporated in the daughter nuclei. Subsequent workers, particularly Chatton ('10) and Alexieff ('13) confirmed in broad outlines Nägler's observations, but distinguished and defined other similar and more advanced types of "primitive" mitosis in amoebae. Since Nägler's time the term promitosis as a distinctive term has lost much of its original significance and has been employed rather generally for mitosis in lower organisms which are characterized by an intranuclear spindle and chromatin derived wholly or in part from a large karyosome. In the process of division the latter is said to elongate and divide and function as a nucleo-centrosome. However, with the use of more refined and specific fixatives and stains, many of the cases reported for-

merly as promitosis in protozoa, fungi, and algae have proven to be typical mitosis.

Nevertheless, students of the Plasmodiophorales immediately recognized the similarity of Nägler's promitotic divisions in *Amoeba* to those in this vegetative plasmodium, and in 1911 Maire and Tison adopted Nägler's term as descriptive of these latter divisions. Subsequent workers, including Cook ('26, '28, '33), Cook and Schwartz ('29, '30), Ledingham ('39), and Couch *et al.* ('39) have used the term protomitosis, a variety of promitosis described by Alexieff, Pavillard ('10), Wernham ('35), and others have employed the term "cruciform" division. Although they figured the same type of division, Nemec ('11, '13), Ferdinandsen and Winge ('20), and Milovidov ('31, '32, '33) avoided extensive use of these terms, while Osborn ('11) described the vegetative division in *Spongospora* as amitotic. His figures and description of the process are nonetheless similar to those of previous and subsequent workers. Favorski objected to the contention that promitosis is specifically characteristic of primitive animals and the Plasmodiophoraceae and pointed out that the karyosome and chromatin may behave in a similar manner during mitosis in higher plants. Terby ('32) likewise condemned the use of promitosis for these divisions in *Plasmodiophora* on the grounds that chromosomes are present and the daughter nucleoli are formed anew from granules in the telophase nuclei and not by division of a mother nucleole. Horne ('30) and Webb ('35) also contended that the vegetative divisions are typically mitotic in *Spongospora* and *Sorosphaera* and thus contradicted all previous workers who maintained that distinct chromosomes are not present.

Two main viewpoints have thus been presented by these cytologists: one that the vegetative divisions are promitotic and fundamentally similar to those in certain amoebae; the other that they are typically mitotic with well-defined chromosomes. Prowazek, Maire and Tison, Schwartz, and Cook in particular have emphasized the former view, and their accounts of the vegetative divisions may be taken as representative of those who held that these mitoses are quite unlike anything present in other fungi and higher plants. Terby, Horne, and Webb may be looked upon as representing the other viewpoint. For the sake of comparison, drawings representative of both views have been brought together in Plate I and contrasted in turn with those illustrating promitosis in certain amoebae.

The resting nucleus of amoebae and plasmodia of the Plasmodiophoraceae is quite small, so that its structure is difficult to see and determine with certainty. Nawaschin described the chromatin in *Plasmodiophora* as a spongy, faintly-stainable reticulum throughout the nucleus, while Prowazek figured the nuclei as having an alveolar achromatic structure with several interspersed granules and a large central nucleole lying in a clear zone. In other nuclei the achromatic material was found to be radially oriented on the nucleole (fig. 1), giving the nucleus a wheel-like appearance. In *Sorosphaera* and *Tetramyxa*, Maire and Tison figured the resting nucleus as devoid of a chromatin reticulum (fig. 2) with the nucleole lying in a vacuole-like clear space filled with hyaloplasm, and numerous granules distributed on the inner periphery of nuclear membrane. They ('09) did not, however, regard these granules as true chromatin but instead as secretory chromidia derived from the karyosome and destined to pass out into the cytoplasm. In *Spongospora*, on the other hand, Osborn figured a wheel-like nucleus with numerous chromatin granules distributed on radially oriented linin threads (fig. 4), but he likewise believed that these granules had been derived from the karyosome. Of the more recent workers, Cook, and Cook and Schwartz have maintained that in *Ligniera* and *Plasmodiophora* the chromatin is aggregated solely in a layer around the inner periphery of the nucleus (fig. 5) with the result that the nucleole appears to lie in a clear vacuolate space, but their observations have not been confirmed. Cook's ('28) studies on *Ligniera*, however, were made from unsectioned material stained in toto, which is obviously unfavorable for study of nuclear details.

Although there is thus considerable difference of opinion among these cytologists as to the structure of the nucleus and the presence of a chromatin reticulum, the "wheel" type of resting nucleus nevertheless has been figured most often and shown to occur in *Plasmodiophora, Spongospora, Sorosphaera, Ligniera, Sorodiscus,* and *Polymyxa*. Milovidov's ('32, '33) observations on resting nuclei of *P. Brassicae* stained by Feulgen's method are particularly pertinent in this relation. In such preparations the karyosome, linin, and granules are colorless, and the only visible structure is the faintly-stained nuclear membrane. Milovidov, nonetheless, believed that small chromatin bodies are present around the inner periphery of the nucleus.

According to Nawaschin, the early prophases of the vegetative divisions in *Plasmodiophora* may be recognized by the emergence of distinct granules in the nucleus (fig. 6) which have a markedly different staining reaction from the karyosome and are not in genetic connection with the latter. Their origin is quite distinct from that of the karyosome, in Nawaschin's opinion. These granules later unite and form an equatorial plate or band. Newaschin's observations were confirmed by Milovidov's ('32, '33) studies which involved Feulgen's nuclear reaction method. As the nuclei enter the prophases, chromatin granules and threads become visible in the nuclear cavity, and these eventually form an equatorial ring (fig. 50). Prowazek ('05), on the other hand, described the karyosome or "Innenkorper" as enlarging and differentiating into a faint-staining achromatic substance and a denser chromatic material (fig. 7). The latter substance then separates into a globular nucleole and a half moon-shaped row of granules (fig. 8), out of which the equatorial ring is formed (fig. 9). Maire and Tison ('09), like Nawaschin, noted the emergence of granules on the linin threads

in *Sorosphaera* during the prophases of promitosis (fig. 10, 11), but they contended that the granules are derived from the karyosome and subsequently aggregate around the latter as an equatorial ring. Blomfield and Schwartz ('10) and Osborn ('11) have figured much the same type of prophases in *S. Veronicae, L. Junci,* and *S. subterranea.* Lutman likewise reported the presence of chromatin granules in the prophases in *Plasmodiophora.* "These granules had been previously concentrated as a hollow sphere enclosing the tropochromatin of the central body" (karyosome), and as the prophases progress the granules of idiochromatin separate from the karyosome and form a spireme, according to Lutman. In *Sorodiscus* Winge also reported a separation of idiochromatin and tropochromatin (fig. 13) in the karyosome in preparation for division, the former giving rise to a thin equatorial plate and the latter forming the nucleole. He believed that in the resting nucleus the idiochromatin may "be partly resolved in the tropochromatin, which later forms the chromophilous filaments radiating from the caryosome." Cook ('26, '28) and Cook and Schwartz ('30) failed to observe any marked prophase stages in *Ligniera* and *Plasmodiophora* but asserted that the peripheral layer of chromatin which is present in the resting nucleus condenses and becomes aggregated in a ring around the karyosome (fig. 14). Shortly thereafter the spindle fibers appear in the nuclear cavity and form a fusiform intranuclear spindle (fig. 15) at right angles to the chromatin ring, which in the meantime has expanded and drawn away from the central nucleole. Many of these cytologists have figured the chromatin ring as a solid continuous band, but Maire and Tison ('11) and Winge reported it to be composed of numerous granules and chromosome-like

PLATE I

Fig. 1. Resting nucleus, *P. Brassicae,* showing wheel-like structure. Prowazek, '05.

Fig. 2. Resting nucleus, *T. parasitica,* with karyosomic granules at periphery. Maire and Tison, '11.

Fig. 3. Uninucleate amoeba, *S. Veronicae,* with centrosome and astral rays. Maire and Tison, '09.

Fig. 4. Resting nucleus, *S. subterranea,* with wheel-like structure. Osborn, '11.

Fig. 5. Resting nucleus, *L. Junci,* with chromatin around inner periphery of nucleus. Cook, '28.

Fig. 6. Early prophase, *P. Brassicae,* showing numerous chromatin granules. Nawaschin, '99.

Fig. 7. Early prophase, *P. Brassicae,* showing separation of idiochromatin and trophochromatin in the karyosome. Prowazek, l.c.

Figs. 8, 9. Differentiation of nucleole and chromatin ring. *P. Brassicae.* Prowazek, l.c.

Figs. 10, 11. Prophases, *S. Veronicae,* showing separation of idiochromatin and its accumulatin on the linin. Maire and Tison, '09.

Fig. 12. Early prophase nucleus, *L. Junci,* with wheellike structure. Schwartz, '10.

Fig. 13. Separation of idio- and trophochromatin in karyosome during early prophase, *S. Callitrichis.* Winge, '13.

Fig. 14. Early prophase, *L. Junci,* showing formation of chromatin ring around nucleole. Cook, '28.

Figs. 15, 16. "Saturn" stages of promitosis, *L. Junci.* Cook, l.c.

Figs. 17, 18. "Cruciform" stages with elongating nucleoli, *L. Junci* and *T. Triglochinis.* Cook, l.c.; Maire and Tison, '11.

Fig. 19. Splitting and separation of chromatin ring, and constriction of nucleole, *L. Junci.* Cook, l.c.

Fig. 20. Later stage, *L. Junci,* showing division of nucleole, Cook, l.c.

Fig. 21. "Double anchor" stage of promitosis, *L. Junci.* Cook, l.c.

Figs. 22, 23. Formation of daughter nuclei, *L. Junci.* Cook, l.c.

Fig. 24. Wheel type of resting nucleus, *A. musicola.* Chatton, '10.

Fig. 25. Early prophase, *A. froschi.* Nägler, '09.

Figs. 26, 27. Equatorial plate stages, *A. lacustris.* Nägler, l.c.

Fig. 28. Same stage, *A. musicola.* Chatton, l.c.

Fig. 29. Early anaphase, *Vahlkampfia limax.* Calkins, '33.

Fig. 30. Later anaphase, *A. froschi.* Nägler, l.c.

Fig. 31. Similar stage, *A. musicola.* Chatton, l.c.

Fig. 32. Telophase, *A. musicola.* Chatton, l.c.

Fig. 33. Reconstructed daughter nuclei, *A. froschi.* Nägler, l.c.

Fig. 34. Wheel type of resting nucleus, *Spongospora subterranea,* with nucleole, radiating linin threads, and chromatin granules. Horne, '30.

Fig. 35. Resting nucleus, *Sorosphaera Veronicae,* with centrosomes and astral rays. Horne, l.c.

Figs. 36, 37. Early prophase, *S. Veronicae.* Webb, '35.

Fig. 38. Same stage, *P. Brassicae.* Terby, '23.

Fig. 39. Spireme stage, *S. subterranea.* Horne, l.c.

Figs. 40, 41. Late prophases, *S. Veronicae,* with four elongate chromosomes. Webb, l.c.

Figs. 42, 43. Later stages, *S. Veronicae.* Chromosomes showing prophase split. Webb, l.c.

Fig. 44. Polar view of equatorial plate, *S. Veronicae,* with four split, twisted chromosomes. Webb, l.c.

Fig. 45. Early equatorial plate, *S. Veronicae,* with four U-shaped chromosomes. Webb, l.c.

Fig. 46. Equatorial plate or "Saturn-stage," *S. Veronicae,* with four chromosomes end to end in a ring around the constricted nucleole. Webb, l.c.

Fig. 47. Similar stage, *P. Brassicae,* with nucleole breaking up into globules. Terby, '32.

Fig. 48. "Saturn-stage," in *S. subterranea* with three of the four chromosomes arranged in a ring. Horne, l.c.

Fig. 49. Oblique view, *S. Veronicae,* of same stage. Webb, l.c.

Fig. 50. Equatorial plate, *P. Brassicae,* stained with Feulgen's nuclear stain; nucleole colorless. Milovidov, '33.

Fig. 51. Metaphase, *S. Veronicae,* showing start of chromosome separation. Webb, l.c.

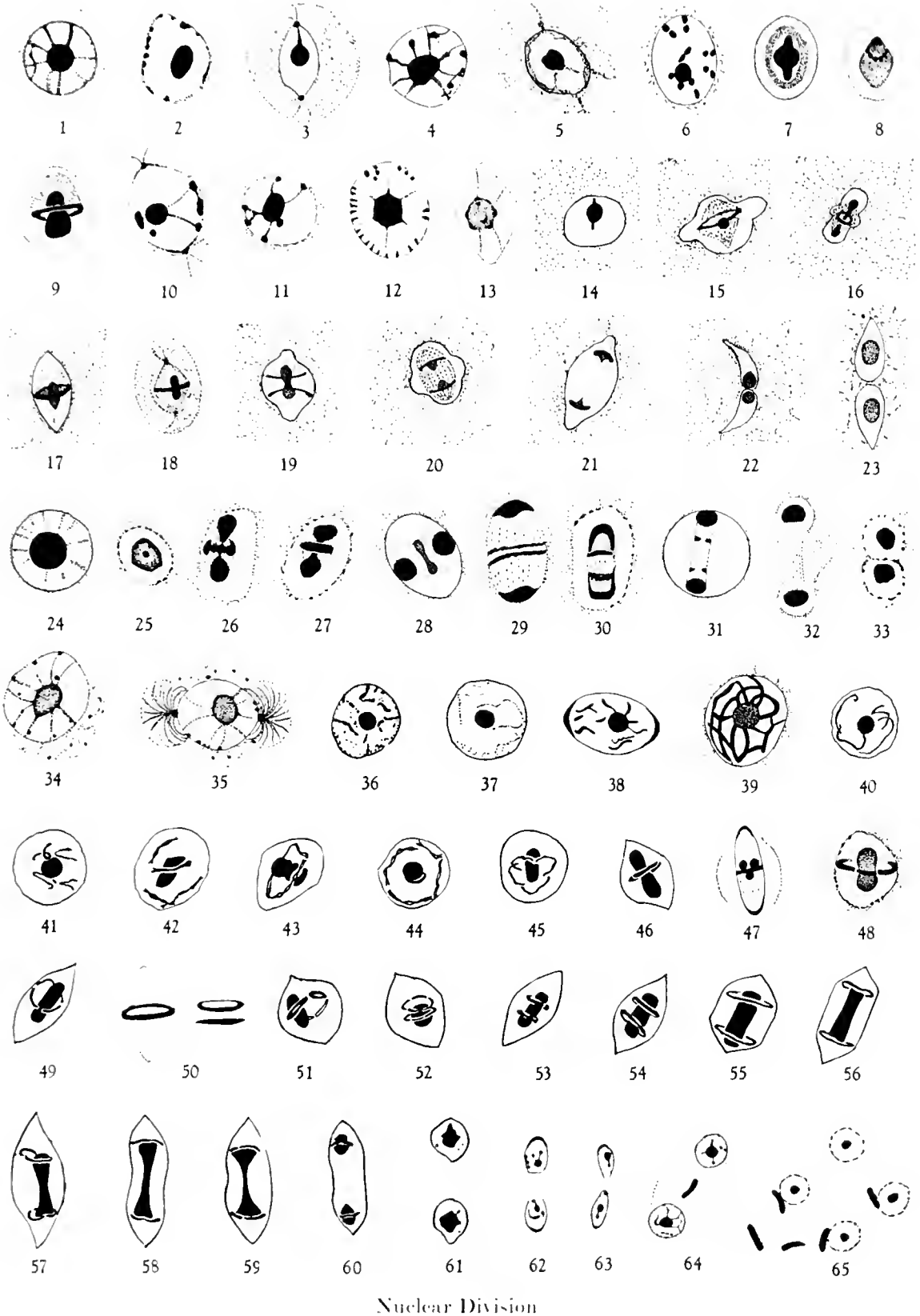
Figs. 52-61. Successive anaphase and telophase stages, *S. Veronicae.* Webb, l.c.

Figs. 62, 63. Formation of daughter nucleoli from granules in telophase nuclei, *P. Brassicae.* Terby, l.c.

Fig. 64. Daughter nuclei, *P. Brassicae,* with remanent of old nucleole between. Terby, l.c.

Fig. 65. New formed nuclei, *P. Brassicae,* with remanents of old nucleoli in the cytoplasm. Terby, l.c.

PLATE I



Nuclear Division

bodies in *Tetramyxa* (Pl. 5, fig. 5) and *Sorodiscus* (Pl. 7, fig. 12), which lends support to the later views of Terby, Horne, and Webb that definite chromosomes are present in the vegetative divisions.

The origin of the spindle has not been clearly demonstrated in promitosis. Whether it originates from achromatic linin material, trophochromatin, or in relation to centrosomes and asters is not sufficiently known. Nawaschin, Favorski, Osborn, Cook and Schwartz, and Webb found no centrosomes and asters during the vegetative divisions in *Plasmodiophora, Ligniera,* and *Spongospora,* but Prowazek, Maire and Tison, Winge, Lutman, Nemec ('13), Horne, and Milovidov ('31) observed them in *Plasmodiophora, Ligniera, Tetramyxa, Anisomyxa* (*Ligniera?*), *Sorodiscus,* and *Sorosphaera* (fig. 2, 10, 18, 35). Nothing is known concerning their presence or absence in *Membranosorus, Polymyxa,* and *Octomyxa.* Maire and Tison figured them in uninucleate amoebae of *S. Veronicae* (fig. 3) and contended that the centrosomes are derived from the karyosome and may retain contact with this body by a slender chromatic strand. In *P. Brassicae,* however, instead of a single body Miss Terby ('23) found a circle of five to six granules around the poles of the nucleus from which the aster-like filaments radiate.

In the early equatorial ring stage, the globular nucleole may often be found in the center of the spindle (fig. 15) surrounded by the peripheral ring of chromatin, according to Cook and others, and because of its characteristic appearance this phase has been described as the "saturn" stage of promitosis. The nucleole or karyosome then begins to elongate and constrict in the center (fig. 16–18). In longitudinal view the ring of chromatin and elongate nucleole present the appearance of a cross, and this phase is accordingly referred to as the "cruciform" stage. The chromatin rings then split lengthwise, according to most workers, and the two daughter rings move apart with the ends of the elongating nucleole (fig. 19). The latter may divide completely in the early anaphase (fig. 20), or the two ends may remain attached for some time by a chromatic strand (fig. 21). The latter condition is usually described as the "double anchor" or "dumb-bell" stage of promitosis. The nucleole finally divides into two daughter nucleoli, and the curved, half-moon-shaped bands of chromatin curve around them (fig. 21, 22) until they are enclosed in a more or less complete sphere, according to Cook. In this manner the karyosome of the daughter nuclei is built up of a peripheral layer of chromatin and a central core of strictly nucleolar material. In the meantime, the spindle fibers disappear, while the nuclear membrane becomes drawn out, curved and somewhat crescentric. It then constricts sharply in the equator and pinches in two, according to Cook (fig. 22), forming the daughter nuclei (fig. 23) which soon move apart and become spherical.

Although variations in the process of promitosis described above have been noted by some workers, most of their views are in agreement about its fundamental outlines. However, P. M. Jones' ('28) description of division in what he believed to be *P. Brassicae* is quite different, contradictory, and as Milovidov characterized it, wholly fantastic. "After the nucleus has become very large, the karyosome moves to one side, and then escapes from the nucleus. The karyosome, during this movement, assumes a dumb-bell shape and starts dividing by promitosis. When the karyosome has completely left the nucleus, it undergoes rapid division, by mitosis, until the plasmodium becomes filled with little nuclei. These nuclei increase in size to form a multinuclear plasmodium. The plasmodium stops feeding and assumes a frothy appearance. The nuclei becomes vacuolated, chromidia are distributed around the vacuoles, and collect into new vacuoles, to form new nuclei . . . ."

It is to be particularly noted that the majority of the early cytologists interpreted the karyosome in terms of the duality concept of the chromatin. They believed that the chromatin which forms the equatorial ring and the division nucleole are derived from the karyosome. This body is accordingly dual in structure and consists of idio- and trophochromatin which separates in the prophases, the latter forming the dividing nucleole and the former the chromatin ring. Maire and Tison described the division of the tropochromatin as amitotic and that of the idiochromatin as indirect or mitotic. According to them, the karyosome at rest is comparable with the nuclei of *Trypanosoma noctieae* or *Amoeba limax;* during division it corresponds to the karyosome of *Caryotropha mesnilii,* to the macronucleus of the Infusoria, to the true chromidia of Goldschmidt, and to some extent to the nucleocentrosome of *Euglena.*

The type of division illustrated in figures 1 to 31 is obviously very similar to promitosis, in the strict sense, which has been described in certain species of amoebae. In order to compare the processes more concretely, drawings by Nägler, Chatton, and Calkins of successive promitotic stages in such species have been brought together in figures 24 to 33. Both the wheel-like (fig. 24) and "vacuolate" resting nucleus (fig. 25) with large, conspicuous karyosomes have been reported in *Amoeba* and these are strikingly similar to the nuclei shown in figures 1, 2, 4, and 34 of the Plasmodiophorales. Division is likewise intranuclear. No sharply defined spindle and chromosomes are formed, but instead the chromatin aggregates into a more or less continuous band across the equator (fig. 26–28). As the karyosome elongates, constricts, and divides, the chromatin band splits lengthwise (fig. 29), and the daughter halves migrate toward the opposite poles of the nucleus (fig. 30, 31) where they are incorporated with the daughter karyosome (fig. 32). As the nuclear membrane disappears in the equator (fig. 32), new membranes are developed around the karyosome, and the daughter nuclei (fig. 33) are thus formed.

To this extent the similarities are very striking—so much so, in fact, that as one reads the accounts of some students of the Plasmodiophorales it becomes obvious that their observations have been in-

thenced by the earlier descriptions of Nägler, Chatton, Alexieff, etc., of promitosis in *Amoeba*. However, one marked difference is apparent. In the Plasmodiophorales the karyosome does not appear to function as a nucleo-centrosome during division. In several genera of this order clearly-defined centrosomes and astral rays have been reported, structures which are lacking in the mitoses shown in figures 24 to 33. Whether or not the division spindle originates in relation to the centrosomes in the Plasmodiophorales is still uncertain. The most striking difference, however, between the two types of division in these two groups, according to Terby, Horne, and Webb, is the presence of sharply-defined chromosomes in the prophases and the equatorial ring stages. This difference will become more apparent in the discussion which follows.

Turning now to the observations of Terby, Horne, and Webb, these workers contended that the failure of previous investigators to find chromosomes in the vegetative divisions was due to insufficient study of the prophases where the chromosomes originate. In *Spongospora* and *Sorosphaera*, Horne found wheel-like resting nuclei (fig. 34) with large karyosomes, radially oriented achromatic strands, and numerous chromatin granules, but he did not regard this as a constant and static structure of resting nuclei. According to him, the structure may change during the various developmental phases. In *Sorosphaera* he found conspicuous centrosomes and astral rays during the prophases (fig. 35), but such structures were never observed by Webb, possibly because the latter employed a strictly nuclear stain. By using a modification of Newton's gentian violet-iodine method, Webb was able to detect small chromatin granules connected by fine threads throughout the resting nucleus. The first visible evidence of division is an increase in the staining capacity of the chromatin granules (fig. 36) which soon becomes alined on the threads (fig. 37), according to Webb. These threads contract as they move away from the periphery and form slender chromosomes (fig. 40) directly, without previous development of a coiled spireme stage. Horne likewise observed a thickening of chromatic rods projecting from the nucleole as the first indication of prophase. Later an irregular chromatic network emerges which goes into a typical coiled spireme (fig. 39) from which the chromosomes eventually emerge. Horne found numerous post-spireme configurations with only two or three V-shaped chromosome, but he nevertheless believed that the haploid number in *Spongospora* is four. Miss Terby also found numerous rods and threads in the prophase nuclei of *P. Brassicae* (fig. 38) from which the chromosomes are subsequently formed. Milovidov, on the other hand, was unable to recognize chromosomes in material stained by Feulgen method.

Returning to Webb's account of *Sorosphaera*, the four chromosomes contract further in the prophases and become V- and U-shaped (fig. 41, 45), and soon thereafter split ends become visible (fig. 42), indicating a splitting of the chromosomes in preparation for division. Up to this time the nucleole remains more or less globular, but it soon begins to elongate in the direction of the poles. The chromosomes then become arranged end to end in an irregular, broken ring in the equator of the nucleus (fig. 43). A polar view of such a stage is shown in figure 44 with the split and twisted chromosomes lying near the periphery of the nuclear membrane. Following this stage, they contract and thicken, so that the longitudinal split is no longer visible (fig. 46). The chromosomes, nonetheless, retain their individuality, according to Horne's and Webb's drawings, as is shown by the breaks in the equatorial ring (fig. 46, 48, 49). This ring stage persists for a comparatively long time and is the one most frequently observed in the vegetative divisions.

According to Horne's and Webb's figures, the elongate nucleole may become slightly constricted at this stage in preparation for division (fig. 46, 48). Miss Terby ('23), however, found that the nucleolar changes vary considerably in *P. Brassicae*. Instead of constricting and dividing more or less equally, it may fragment into two or more unequal parts (fig. 47) or move intact as a single body to one of the poles. Oftentimes, parts of it remain stranded between the daughter nuclei (fig. 64, 65) as in higher plants.

The metaphase split reappears first in the median region of the chromosomes (fig. 51) at the conclusion of the equatorial ring stage and travels outward to the ends, which suggested to Webb that the spindle fiber attachment is median. As the chromosome halves separate, two daughter rings are formed (fig. 52) which migrate toward the opposite poles (fig. 53–57) until they reach the ends of the elongate nucleole (fig. 56, 58, 59). According to Webb, the nucleole in *Sorosphaera* does not constrict as a rule until telophase (fig. 57–59). The two parts finally separate and become surrounded by daughter chromosomes (fig. 60) at the poles of the nucleus. The nuclear membrane then constricts and divides in much the same manner as the nucleole and thus forms the daughter nuclei (fig. 61). The chromosomes adhere to the nucleole at first, but later separate from it. Miss Terby, however, maintained that the nucleoli are formed anew at each telophase in *P. Brassicae*. After the daughter nuclei have been formed, the chromatin mass gives off material which unites to form the daughter nucleoli (fig. 62, 63). As to the origin of the daughter nuclear areas, Miss Terby ('23) reported that they begin in the prophases as two hyaline vesicles on the polar sides of the nucleole. As the latter elongates, divides, and the two segments separate, the vesicles precede them to the poles of the nucleus. The vesicles then pass through the nuclear membrane at the poles and expand, and shortly thereafter the daughter nucleolar segments and chromatin enter and are thus incorporated in the vesicles. The boundaries of the vesicles become the nuclear membranes and thus constitute the limits of the daughter nuclei. In a later paper, however, Miss Terby ('32) modified this account and reported

that the polar vesicles contract to small globular areas surrounded by granules from which astral rays radiate, as noted elsewhere. Thus the vesicles themselves do not become the nuclei, but the daughter nuclei are formed in the areas occupied by the vesicle before contracting.

The type of division described by these three workers is distinctly mitotic and, except for the behavior of the nucleole, according to Horne's and Webb's figures, is fundamentally similar to nuclear division in the higher plants. Miss Terby, as noted before, held that the nucleole also undergoes the same changes as in the higher plants, so that there is no difference in this respect either. On the other hand, the divisions figured by Horne and Webb are also similar to the promitoses illustrated in figures 1 to 23. The chief difference is the presence of chromosomes. It is not improbable, as Webb contended, that the earlier workers overlooked the early prophases and the origin of the chromosomes and that their fixation and staining technique did not differentiate chromosomes in the equatorial ring. As noted elsewhere, the nuclei of the Plasmodiophorales are quite small, and their structure is difficult to interpret. The use of more specific and refined technique in intensive study of the early prophases and equatorial ring stages may thus possibly eliminate the present controversy on the nature of the vegetative divisions.

In this relation it is to be noted that typical mitosis without large nucleoli has been reported in the vegetative zoosporangial stage of *Ligniera, Plasmodiophora, Polymyxa,* and *Octomyxa* by Cook ('26, '28), Cook and Schwartz ('30), Ledingham ('39), and Miss Whiffen ('39). In these as well as other genera the zoospores from germinating resting spores develop into plasmodia which eventually cleave into uninucleate segments—the rudiments of zoosporangia. These segments develop walls, and their nuclei divide twice to several times in a strictly mitotic manner in preparation for zoospore formation. Cook and Schwartz reported that up to the time of cleavage into zoosporangial segments the nuclei in the plasmodia of *Ligniera* and *Plasmodiophora* divide promitotically, but in *Polymyxa* Ledingham reported that division in the thalli which form zoosporangia is mitotic from the start. Miss Whiffen also found that the divisions in the zoosporangia of *Octomyxa* are mitotic. These authors thus reported a regular alternation of mitosis and promitosis. The zoosporangial stage is characterized by mitosis, then follows a phase of promitotic division in the early development of the sporogenous plasmodium which is terminated by the so-called transitional stage, and finally two meiotic divisions. Inasmuch as the divisions in the zoosporangia are mitotic and very similar to the two divisions at sporogenesis, Cook ('26, '28, '33) and Fedorintschik ('35) concluded that they are meiotic in *Ligniera* and *Plasmodiophora,* respectively. In *P. Brassicae,* however, Cook and Schwartz described them as merely mitotic. In an attempt to explain the alternation of meiosis and promitosis in this species, they proposed the theory that promitosis is characteristic only of diploid nuclei, a theory which is contradicted by their own observation that the first meiotic division of the diploid nucleus in spore formation is indirect and not promitotic. Furthermore, if Cook's ('28, '33) previous report is correct that the primary nucleus of the incipient zoosporangia in *Ligniera* is diploid (and undergoes meiosis), it should accordingly divide promitotically. However, he described and figured such nuclei as dividing mitotically.

The report of typical mitotic divisions during zoospore formation, promitosis in the developmental stages of the sporogenous plasmodium, and the reoccurrence of mitosis during the reduction divisions nevertheless raises numerous questions on the significance of this alternation (if it actually does occur), and it is thus obvious that future studies of karyokinesis in the Plasmodiophorales must be closely correlated with the various developmental phases.

"Akaryote Stage"

The period of vegetative divisions in the development of the sporogenous plasmodium is reported to be followed shortly by the so-called "enucleate," "akaryote," "chromidial" or "transitional" stage. According to most workers, this phase is characterized by a reduction in size and disappearance of the karyosome, comparatively empty, vacuole-like nuclei, and the presence of numerous deeply-stainable bodies or chromidia in the cytoplasm around the nuclei. Nawaschin first observed this stage in *Plasmodiophora* in 1899, and since that time it has been reported by most subsequent students for the other genera of this order. In the opinion of many cytologists it is thus as constant and diagnostic a character of the Plasmodiophorales as promitosis.

Stages in the development of the akaryote stage are shown in Plates 2 to 13, which illustrate the life cycles of all the plasmodiophoraceous genera, and will not be illustrated separately at this point. After the vegetative divisions have been completed, the karyosome decreases in size as the somatic or trophochromatin is extruded into the cytoplasm in the form of secretory chromidia, according to Prowazek ('05), Maire and Tison ('09), and others. Maire and Tison regarded this extrusion as a cleansing proecss by which the generative chromatin is separated from the nutritive chromatin in preparation for the sporogonic divisions which follow. As a result of this extrusion, the nuclei, when stained with haematoxylin, appear comparatively empty and devoid of stainable material and frequently have the appearance of vacuoles in a cytoplasm filled with deeply-stained chromidia.

According to Blomfield and Schwartz, Schwartz, and Osborn, extrusion of chromatin in *Sorosphaera, Ligniera,* and *Spongospora* takes place along the linin threads until the chromatin reticulum and karyosome have disappeared. These workers believed that the nuclear membranes also disappear during this stage. In *L. Junci,* Schwartz described the process as follows: "the nuclear membrane dis-

appears, and the karyosome diminishes in size and finally disappears also, so that we have a number of vacuoles more or less circular in outline situated in the spherical mass of plasma." Osborn likewise described the disappearance of the nuclear membrane in *Spongospora* and the formation *de novo* of new nuclei. P. M. Jones ('28) also maintained that the nuclei disappear completely in *P. Brassicae* and that the new nuclei are formed by the aggregation and fusion of chromidia within small vacuoles. Cook ('26, '28) described a complete extrusion of chromatin from the nuclei of *L. Junci*, but later he and Schwartz reported that in *P. Brassicae* a small amount of chromatin may remain within the nuclei. They, nevertheless, refuted the reports of previous workers that the nuclear membrane disappears. However, in *Sorodiscus radicicolus*, Cook later ('31) reported that all of the chromatin is extruded during the akaryote stage and later re-enters (!) the nucleus in preparation for meiosis. Winge, on the other hand, found no marked chromatin extrusion and akaryote condition in *S. Callitrichis* and referred to the changes which the nuclei undergo in preparation for meiosis as the transitional stage, a term later adopted by Horne and Webb. In *Spongospora*, Horne also noted that the nuclear membrane remains clear and distinct throughout this stage and the nuclei have a well-defined chromatin reticulum, chromidia, and a faintly-stainable nucleole. Similar stages were found by Miss Terby ('24) who denied the existence of an akaryote stage in *P. Brassicae*. By using Newton's gentian violet iodine stain on *Sorosphaera*, Webb also found the normal interphase chromatin reticulum and a large faintly-stainable nucleole present in the nuclei during the transitional stage. His observations were later confirmed in part by Ledingham's study of *Polymyxa*. The latter worker observed a well-defined reticulum in nuclei stained by Newton's method, whereas in preparations stained with iron-alum haematoxylin the nuclei appeared to be devoid of chromatin. The latter four workers accordingly refuted previous cytologists on the presence of marked akaryote stage at the conclusion of the vegetative divisions.

With the exception of Terby, Horne, Milovidov, Webb, and Ledingham, most workers have described a definite reorganization of nuclei following the so-called akaryote stage. As noted before, Schwartz, Osborn, and Jones contended that the generative nuclei arise *de novo* on new sites in the cytoplasm from extruded chromatin, while Blomfield and Schwartz were uncertain about their origin in *Sorosphaera*. All other workers, however, held that the nuclear membranes persist and that the nuclei undergo certain characteristic changes. During this process centrosomes and astral rays become quite conspicuous in the cytoplasm, but it is not certain whether they arise *de novo* and divide or originate from the karyosome, as Maire and Tison contended. Whereas several workers denied the presence of these structures during the vegetative divisions, most of them agreed that centrosomes and astral rays are conspicuous features of the reconstructed nuclei and sporogenous divisions. However, Blomfield, Schwartz, and Cook apparently never found these structures in any of the developmental stages of *Ligniera*, *Sorosphaera*, and *Plasmodiophora*, since none of their figures show centrosomes and asters. Concomitant with the development of these cytoplasmic structures, chromatic strands, granules, and other configurations appear in the nuclei, which are generally regarded as prophases of meiosis and will be discussed in greater detail below.

It is apparent from this discussion that the observations of the early cytologists of the Plasmodiophorales were greatly influenced by the chromidia hypothesis of Goldschmidt, Schaudin, Popoff, and other protozoologists of that period. Its influence is also evident in the more recent contributions of P. M. Jones and to a large extent in the papers by Cook and Schwartz. Lack of space does not allow a detailed account of the chromidia hypothesis here. Suffice it to note that in *Actinosphaerium*, *Arcella*, *Arachnula*, *Entamoeba*, and numerous other rhizopoda R. Hertwig, Schaudin, Popoff, Dobell, and others reported a gradual disappearance of the nucleus as chromidia are extruded into the cytoplasm and the subsequent formation of new nuclei in reproductive cells from chromidial granules. These observations among others were the foundation of Goldschmidt's theory and eventually led to the "binuclearity hypothesis" of Schaudin, Prowazek, Maire and Tison, Blomfield and Schwartz, and others interpreted the akaryote and reconstruction stages of the Plasmodiophorales in terms of this chromidia hypothesis, while Schwartz, Osborn, and Jones appear to have adopted this theory completely as an explanation of the changes undergone by the nuclei during these phases.

The chromidia hypothesis has been largely discredited in the last three decades by researches involving the use of mitochondrial fixatives, Feulgen's nuclear stain, and other more specific fixatives and staining techniques. In *Arcella* and *Clamydophrys*, for instance, the nuclei do not disintegrate as was previously claimed, according to Jollos, but instead are masked during certain stages by a chromidial network which can be dissolved away in trypsin and pepsin, leaving the nuclei sharp and clear. That this network is not composed of chromatin derived possibly from the nucleus is evident by its negative reaction to Feulgen's stain. Likewise, in most of the earlier reported cases of chromidia extrusion and growth, the so-called chromidia have been found to relate to chondriosomes, ergastic, reserve, and degenerative products of metabolism, etc. In *Actinosphaerium*, classic example of chromidia extrusion, Rumjantzew reported that the chromidia appear to be composed of a carbohydrate held in a mechanical or perhaps adsorbtive union with a protein. In *Difflugia* they appear to be composed of glycogen, according to Zuelzer, while in *Eimeria* they are made up of volutin or metachromatin which have a strong affinity for basic dyes. Additional cases of this na-

ture may be cited to show that what had previously been regarded as chromatin extruded from the nucleus is now known to be chondriosomes, reserve, and degenerative products of metabolism. Belar ('26) thus characterized the present status of the chromidia theory as follows: "Die Lehre vom Chromatindualismus steht und fällt mit einer unkritischen Fassung des Chromatinbegriffs, sie is das posthume Produkt einer naiven Interpretation der histologischen Färbung." In light of these more recent data from the field of protozoology, Prowazek's, Maire and Tison's, Blomfield and Schwartz's, Schwartz's, Osborn's, Cook's, and Jones' interpretations of chromatin extrusion, chromidia, and the origin of the generative nuclei in the Plasmodiophorales need revision.

Milovidov attempted to do so in a restudy of the akaryote and nuclear reconstruction stages in *Plasmodiophora* with the aid of mitochondrial fixatives and Feulgen's nuclear stain. From these studies he concluded that the so-called chromidia in the cytoplasm are nothing more than chondriosomal residue, excretions, or secretions. He found that shortly before spore formation the plasmodium becomes quite vacuolate and that chondriosomes and other bodies may frequently lie within such vacuoles. This appearance, according to him, is the basis for Schwartz, Osborn, and Jones' claim that new nuclei arise *de novo* in vacuole-like areas from extruded chromatin granules. With Feulgen's stain such granules show no positive chromatin reaction. As to the presence of a marked akaryote stage with nuclei partly or completely devoid of chromatin, Milovidov discredited previous workers and maintained that it does not exist as a distinct developmental phase of the Plasmodiophorales. He contended that the plasmodium does not fix and stain uniformly throughout all of its developmental phases, so that fixatives and stains which give good preparations of one phase are unsuitable for another stage. In none of the properly fixed and stained plasmodia did he find empty, vacuole-like nuclei. Instead, when so-called enucleate- and akaryote-like stages described by previous workers were stained by Feulgen's method, the nuclei were found to have numerous chromatic granules, strands, and spireme-like threads, all characteristic of meiotic prophases. Milovidov thus concluded that the akaryote and nuclear reconstruction stages of earlier cytologists relate in part to artifact, as Miss Terby had earlier pointed out, misinterpreted meiotic prophases and telophases, poorly fixed and stained resting nuclei of vegetative plasmodia, and abnormal nuclei of degenerating schizonts and plasmodia. His contentions are supported to a great extent by the failure of Maire and Tison, Winge, Terby, Horne, Webb, and Ledingham to find marked akaryote stages in *Tetramyxa, Sorodiscus, Plasmodiophora, Spongospora, Sorosphaera,* and *Polymyxa,* respectively.

Meiosis

It is now rather generally believed that meiosis occurs during the last two divisions before or during cleavage into resting spores, and these divisions are respectively referred to as hetero- and homeotypic. Nawaschin first noted these divisions in *Plasmodiophora* but reported only one mitosis before spore formation. Prowazek ('05) found two mitoses, and since that time two divisions have been universally reported. Maire and Tison ('09) were the first to count the chromosomes during these divisions in *Sorosphaera,* and because the number appeared to be halved in the first divisions they accordingly concluded that these divisions are reductional. Their interpretation has been accepted by most subsequent workers. Exceptions to this view, nevertheless, may be found in the literature. Prowazek ('05) reported that reduction in *Plasmodiophora* occurs during maturation of the resting spores following autogamy or a previous fusion of cleavage segments or incipient spores. Winge contended that a numeral reduction of chromosomes takes place in the *second* instead of the first sporogonic division in *Sorodiscus.* More recently Cook ('26, '28, '33) and Fedorintschik ('35) reported a second reduction division in the zoosporangia or gametangia of *Ligniera* and *Plasmodiophora* in addition to the one which occurs at sporogenesis. According to Cook ('33, p. 221), the two reductions in *Ligniera* are necessitated by a double fusion, one between "swarm cells" and the other between zoospores. Fedorintschik reported only one fusion in *P. Brassicae.* However, neither of these workers counted the chromosomes during the first two divisions in the zoosporangia, and their contention that these divisions are meiotic is based solely on the similarity in appearance of the latter to the reduction divisions at sporogenesis. In *Tetramyxa Elaeagni,* Yendo and Takase ('32) reported that the sporonts are haploid, which presupposes a reduction before the plasmodium cleaves into spore mother cells or sporonts.

As noted before, most cytologists reported that the vegetative meiotic divisions are separated by a marked akaryote stage, but Terby, Horne, Webb, Ledingham, and particularly Milovidov failed to confirm these reports. Thus, the latest data from carefully fixed and stained material suggest that the akaryote stage of the early workers relates in part to an achromatic phase of the nucleus and partly to the meiotic prophases. Prowazek's ('05) figures 17 to 22, for example, show chromatin reticula, loops, garlands, spireme threads, and eight chromosome-like bodies which are very characteristic of the meiotic prophases of later workers.

Following the more or less achromatic transitional phase, the nucleole and chromatin filaments of *Sorosphaera, Plasmodiophora,* and *Spongospora,* according to Maire and Tison ('09), Terby ('24) and Horne, become more basophyllic and clearly visible in the nucleus. At the same time sharply defined centrosomes and asters appear at the poles. The chromatin then aggregates at the poles into two more or less dense masses, which may remain connected by fine chromatic filaments. This is the so-called "garland stage" of meiosis. Horne found that each polar

mass is composed of four distinct chromosomes in *Spongospora*. Somewhat similar early changes were reported by Maire and Tison ('11) and Winge for *Tetramyxa*, Winge for *Sorodiscus* (Pl. 7, fig. 19, 23, 24), Winge and Webb for *Sorosphaera* (Pl. 6, fig. 29, 30), Terby ('24) for *P. Brassicae*, and Cook ('31) for *S. radicicolus*. Winge found that the two polar masses may be arranged in the form of garlands with connected filaments, an arrangement previously reported by Prowazek for *P. Brassicae*, and subsequently by Terby (l.c.) and Cook (l.c.). The nucleole may gradually disappear during this stage or become flattened and aggregated with the chromatin masses and filaments at one side of the nucleus. This nuclear configuration is strikingly similar to the collapsed synizetic (zygotene) stage in higher plants. In the Plasmodiophorales, however, it is generally referred to as synapsis and has been so far reported as such in *Plasmodiophora* (Terby, Cook and Schwartz, Milovidov), *Spongospora* (Osborn, Horne), *Sorosphaera* (Maire and Tison, Webb), and *Sorodiscus* (?) (Winge). In *Spongospora*, Horne found two contraction stages and designated the second one as synapsis. Each loop in the second contraction stage is converted directly into a heterotypic chromosome. Cook ('28) found no meiotic prophases in *L. Junci*, and in *P. Brassicae* he and Schwartz reported and figured only one stage which might be interpreted as such. The chromatin was arranged in a thick thread with several globular nucleole-like bodies distributed along its length. Cook and Schwartz regarded this stage as comparable to synapsis, but it bears little or no resemblance to the synaptic stage figured by other workers.

Before or during the contracted stage, the nucleole disappears, while the chromatin threads loosen up and take on the appearance of elongate chromosomes. According to Webb, in *Sorosphaera* the chromatin at this stage consists of beaded threads spread over the periphery and has the appearance of a normal pachytene. The threads occasionally appear double, and after further contraction four chromosomes become visible (Pl. 6, fig. 34). This stage corresponds to diplotene in higher plants, according to Webb. Then follows diakinesis (Pl. 6, fig. 35), during which four well-defined bivalents are visible. In *P. Brassicae*, Cook and Schwartz failed to find comparable stages and merely reported that the chromatin thread segments into chromosomes as the nuclear membrane disappears. Miss Terby ('24), on the other hand, found well-defined strepsitene, early and late diakinetic stages (Pl. 3, fig. 68–71) with four bivalent chromosomes in *P. Brassicae*, which indicates that Cook and Schwartz overlooked these phases. A diakinetic stage with thick broadly V-shaped and ring chromosomes was also observed by Horne in *Spongospora*.

Shortly after diakinesis the nuclear membrane disappears, and the chromosomes become oriented in the equator of a well-defined division spindle with centrosomes and asters. All other workers reported that the nuclear membrane disappears during metaphase, but in *Plasmodiophora* and *Spongospora*, Prowazek and Horne figured it as persisting until the telophases. The origin of the meiotic spindle has not been solved, but Webb believed that it grows inward from the poles to the equator. According to most cytologists the heterotypic chromosomes are closely associated on the equatorial plate and in metaphase and often appear as an irregular band or row of connected globules, so that the profile and polar views are not very characteristic of heterotypic divisions. In *Spongospora*, however, Horne figured the chromosomes as short and thick with conspicuous intervening gaps in the equatorial plate, which makes it possible to recognize and count the individual members. At this stage they may often show four blunt ends, which indicates their tetrad nature, according to Horne.

With the exception of Winge, most cytologists held that the homologues separate at metaphase of the first division and move to the poles where they are incorporated in the daughter nuclei. In *Sorosphaera*, Webb found the late anaphase and telophase chromosomes to be double, which suggests that the equatorial split for the homeotypic division occurs quite early. Cook ('28) failed to see nuclear membranes in the late telophases of *L. Junci* and thus concluded that they are not formed between the first and second divisions. All previous and subsequent workers, however, have shown that a well-defined membrane develops around the telophase groups of chromosomes and that daughter nucleoli are subsequently formed. Interkinesis is usually short in duration. In *P. Brassicae* Miss Terby ('24) reported that the telophase nuclei go directly into the prophases of the next division, but in *Spongospora* wheel-like resting nuclei and distinct prophases may intervene between the two divisions.

The second division is likewise mitotic or indirect but considerably smaller in size than the first one. Failing to count the chromosomes, Cook and Schwartz regarded this size difference as proof that these two divisions are respectively hetero- and homeotypic, a criterion which is obviously of no critical value in this respect. Osborn, Milovidov, Wernham, Whiffen, and others also made the same assumption without counting the chromosomes. On the other hand, Maire and Tison, Winge, Terby, Horne, Yendo and Takase, and Webb based their contention on a numerical reduction in chromosome number during these divisions. Whether or not their chromosome counts are accurate remains, however, to be shown from more intensive study of these divisions.

The chromosomes of the Plasmodiophorales are quite small and are not always clearly defined on the equatorial plate, so that it is difficult to make accurate counts. Nevertheless, numerous attempts have been made, as is shown in table 1.

The numbers are low multiples of 2, with 8 predominating as the diploid number. In *S. Veronicae* Maire and Tison reported 16 and 8 chromosomes, but Webb later found only 8 and 4. Winge, as noted before, described the first division as vegetative or so-

Table 1. Chromosome numbers in the Plasmodiophorales.

Species	Author	Number *N*	Number *2N*
Sorosphaera Veronicae	Maire and Tison, '09	8	16
S. Veronicae	Webb, '35	4	8
Tetramyxa parasitica	Maire and Tison, '11	?	8
T. Elaeagni	Yendo and Takase, '32	6	12
Sorodiscus Callitrichis	Winge, '13	4	?
S. radicicolus	Cook, '31	2	4
S. Heterantherae	Wernham, '35	?	4–6
Plasmodiophora Brassicae	Maire and Tison, '09	?	8
P. Brassicae	Lutman, '13	?	8
P. Brassicae	Terby, '24	4	8
P. Brassicae	Nawaschin, '24	?	6–8
P. Brassicae	Jones, '28	?	8
Spongospora subterranea	Osborn, '11	?	8 (?)
S. subterranea	Horne, '30	4	8

matic and recorded 16 individual chromosomes associated in eight pairs on the equatorial plate, which separated and were distributed to the daughter nuclei. In the second division this number was halved to four. Winge's confusion as to the nature of the respective divisions has led to the belief that the chromosome numbers in *S. Callitrichis* are 16 and 8, but it is evident from Winge's report that 4 should be recorded as the reduced number. In *P. Brassicae* four workers have recorded 8 as the diploid number, and curiously enough this is the number of bodies figured earlier by Prowazek ('05) in the transitional phase. In *Spongospora subterranea,* Osborn figured polar views of the second division with 7 chromosomes, which corresponds closely to the number later recorded by Horne for the first division. It is not improbable, however, that Osborn's figure relates to the first meiotic division.

Schizogony and Cleavage

Vegetative multiplication of young plasmodia by division, segmentation or fragmentation has been reported for all genera of the Plasmodiophorales except *Membranosorus* and *Octomyxa.* In *Ligniera* it is said to be lacking entirely or reduced to the formation of a few daughter segments, while in *T. Triglochinis* and *P. graminis* true "multiple division" has been reported. Nawaschin did not observe segmentation in *Plasmodiophora,* but he believed that its occurrence is the only plausible explanation of the frequent presence of numerous uni- and multinucleate amoebae and plasmodia in a single host cell. Since that time most cytologists have reported its occurrence, although none of them, with the possible exception of Ledingham, actually observed it in living material. Like Nawaschin, they found several amoebae and plasmodia in the same cell and assumed that the former were the products of fragmentation. Maire and Tison ('10) found similar stages in *Sorosphaera* but interpreted them as fusion stages of amoebae and young plasmodia in the formation of the sporogenous plasmodium. Brasil, however, suggested that these stages relate instead to fragmentation or schizogony—a suggestion which Maire and Tison adopted. These workers thus introduced the protozoologists' term "schizogony" as descriptive of the vegetative fragmentation of the plasmodium, and since that time it has been rather widely adopted. Pavillard ('10), however, contended that schizogony in *Sorosphaera,* as described by Maire and Tison, resembles plasmotomy instead of true schizogony as in *Trichosphaerium sieboldii* (Doflein, '09, '27) and *Hepatazoon anis* (Wenyon, '26), for example. He thus restricted the term schizogony to the "multiple division" of Doflein, while Maire and Tison ('11) interpreted it in the broad sense of most protozoologists to include the plasmotomy of Doflein as well as all other methods of simple and multiple divisions.

Schizogony in the Plasmodiophorales is reported to occur most frequently during the 8- and 16-nucleate stages of the plasmodia or schizonts. A few uni- and multinucleate meronts may be formed as in *Ligniera,* or the whole plasmodium may undergo multiple division as in *T. Triglochinis* and *P. graminis.* The latter type of complete fragmentation appears to be limited as far as present-day knowledge goes. Most cases so far reported involve primarily the constriction and cutting off of peripheral uni- and multinucleate segments. No cases have yet been described in which all or most of the nuclei migrate to the periphery of the schizont and become enveloped in cytoplasmic buds, which are subsequently pinched off, leaving a central mass of degenerating cytoplasm and nuclei, as in *Hepatazoon anis,* for example. The mechanics of schizogony are unknown, because the process has not been extensively observed in living material. In *Polymyxa graminis,* Ledingham merely reported that the pseudopodia are retracted and the protoplasm becomes denser before the thallus splits up into meronts.

In cases in which only a few meronts are formed the remaining portion of the schizont may mature directly into a sporogenous plasmodium or sporont. The delimited meronts grow in size and become multinucleate and may in turn function as schizonts. Otherwise, they develop into sporonts. The destiny of the various portions depends to some extent on the length and activity of the vegetative period. Inas-

much as schizogony will be discussed further in the description of individual species in Chapter IV, further discussion of the process need not be presented here.

Cytokinesis or division of the plasmodium or sporont into resting spores takes place by cleavage, and as far as is now known may be closely associated in point of time with the two meiotic divisions. In *Sorosphaera Veronicae*, according to Maire and Tison ('09), cleavage begins in the late prophases of the first division (Pl. 6, fig. 33), and by the time of the equatorial plate stage, spore mother cells have been completely delimited (fig. 37). These cells divide into two uninucleate segments (fig. 38) in which the second division then takes place (fig. 41). At the completion of this mitosis these segments in turn divide into the definite spore rudiments (fig. 42). In this species at least cytokinesis may follow each mitosis. A similar sequence has been reported by Winge for *Sorodiscus* (Pl. 7, fig. 26–30), although the stages do not appear as sharply defined. Figure 27, however, suggests that the sequence varies and that the first division may be complete before cleavage begins. Similar variations have been reported for *Plasmodiophora* also. Lutman ('13) and Milovidov ('31) found that the first meiotic division is usually complete by the time the initial segments are delimited. The latter may be uni-, bi-, or multinucleate, and after the second meiotic division has been completed (Pl. 4, fig. 80, 81) they cleave into uninucleate spore segments (fig. 82, 83). Cook ('28), Cook and Schwartz ('30), however, reported that cleavage in *Ligniera* and *Plasmodiophora* does not begin until both divisions have been completed. Ledingham's photomicrographs suggest the same sequence of events in *Polymyxa*. In *Tetramyxa* the peripheral plasmodium first cleaves (Pl. 5, fig. 8) into uninucleate segments or sporonts (fig. 9–12). Two meiotic divisions occur in these segments, and these mitoses are usually over (fig. 13–17) by the time cleavage into definite spores is complete. In *Octomyxa* large uninucleate segments are delimited in which the two meiotic divisions occur, and following the completion of the second divisions, which are quadripolar, the segments cleave into spores, according to Whiffen. Very little is known about cytokinesis in the other genera. The time relations of cleavage to the successive meiotic divisions doubtless varies in different species and probably in the same species, so that under varying conditions it may occur during as well as after meiosis.

Marked changes take place in the cytoplasm prior to cleavage. In *P. Brassicae*, according to Nawaschin ('99), Lutman, and Milovidov, the cytoplasm becomes highly vacuolate (Pl. 4, fig. 78) and thus fills the host cell more or less completely. In *Polymyxa*, however, Ledingham reported that the cytoplasm becomes less vacuolate, smaller in volume, and denser, while numerous oil globules emerge and increase the refringency of the plasmodium. According to Lutman, the denser cytoplasm collects around the nuclei in *P. Brassicae*, while the vacuoles fuse and cut the plasmodium up into uninucleate segments. He contended that the process of spore formation in the Plasmodiophorales is quite different from that described by Harper ('00) for certain myxomycetes, but his figure 34 (Pl. 4, fig. 79) nevertheless shows a well-defined cleavage furrow progressing between the nuclei. Milovidov's ('31) text-figure 3 likewise shows that large segments are first delimited by furrows, and these in turn cleave into uninucleate spores. Progressive cleavage by furrows is also suggested by figure 99, Plate 4 of *P. Diplantherae*, figure 33, Plate 6 of *S. Veronicae*, figures 3 and 10, Plate 7 of *Sorodiscus karlingii*, etc. Contrary to Lutman's belief, cleavage in the Plasmodiophorales appears to take place by progressive furrowing as in the myxomycetes. Furthermore it does not appear to be simultaneous as Woronin, Nawaschin, and Maire and Tison reported.

Other cytological details such as cellular relations between host and pathogen are discussed in Chapters IV and VI in connection with the descriptions of individual species and the diseases which they cause.

Chapter III

Sexuality and Alternation of Generations

Very little is known about sexual reproduction in the Plasmodiophorales, yet most workers have assumed that it occurs. So far, actual fusion of gametes in living material has been observed only in *Spongospora subterranea*. The evidence of sexuality in the group as a whole is therefore largely indirect. It is based on isolated observations of paired amoebae and zoospores, binucleate amoebae, the appearance of paired and fusing nuclei in the plasmodium, and primarily on the reported occurrence of meiosis at sporogenesis, which presupposes a nuclear fusion at some stage of development. Inasmuch as there is considerable difference of opinion about the time, place, and nature of plasmogamy and karyogamy in different genera as well as in the same species, the data on sexuality for each genus will be considered separately.

In *Plasmodiophora*, Nawaschin ('99) reported the union of several amoebae in the formation of the plasmodium, but he did not regard these fusions as having any sexual significance. Later, Miss Terby ('24), Milovidov ('31), and several other workers expressed the same view concerning the development of the sporogenous plasmodium. Prowazek ('05), however, contended that the incipient spore segments or "sporogametes" fuse in pairs following cleavage (Pl. 4, fig. 89), after which the zygote or binucleate spore begins to encyst (fig. 90, 91). One

of the so-called gametic nuclei then divides (fig. 92), during which division it undergoes a chromatin reduction and forms a variable number of reduction bodies (!). Meiosis is followed almost at once by karyogamy. Apparently all but two nuclei degenerate (fig. 93), and the two remaining ones fuse to form a synkaryon (fig. 94). Prowazek's account of reduction is not very clear, and his drawings of the process do not clarify the accompanying description. It is accordingly difficult to determine from his confusing account the duration of the respective diploid and haploid generations in *Plasmodiophora*. According to him, the diploid phase is apparently quite short (text-fig. 1).

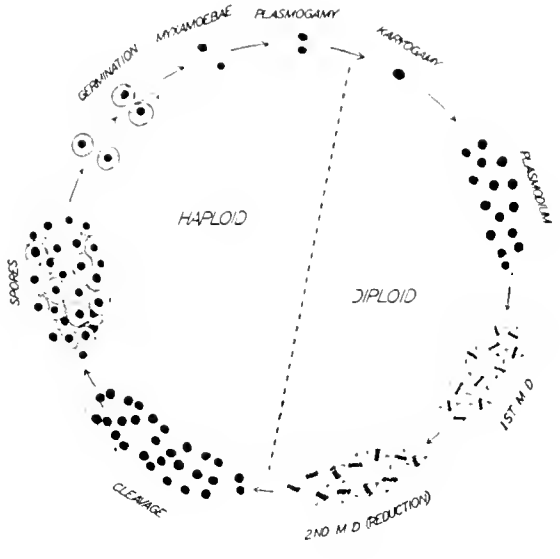
Prowazek's account was refuted by Maire and Tison ('09) who failed to find any evidence of plasmogamy and karyogamy following cleavage. They nevertheless believed that sexual fusions occur in *Plasmodiophora* and postulated that it might take place between two amoebae from germinating spores. Pavillard ('10) rejected this view and considered it more plausible that karyogamy occurs in the plasmodium shortly before meiosis, presumably following a coalescence of amoebae. His theory, however, relates to the Plasmodiophoraceae as a whole rather than to *Plasmodiophora* specifically. Winge concurred with Maire and Tison's view and assumed that the motile cells from resting spores copulate in pairs to form small myxoplasma which penetrate the host and develop into plasmodia. The diploid phase persists until the *second* sporogonic division where reduction occurs, according to Winge (text-fig. 2). This text-figure is also representative of his view concerning alternation of generations in all genera of the Plasmodiophorales. Lutman, Chupp, and Milovidov ('31) were uncertain about the time and place of plasmogamy and karyogamy, but believed that they must occur at some stage on the grounds that a reduction in chromosome number takes place during the first sporogonic division (Pl. 3, fig. 63–73). Miss Terby ('24) postulated that fusion occurs outside of the host cell between pairs of zoospores, a view which Nawaschin had accepted by 1924.

P. M. Jones ('28) described and figured the formation of two types of gametes from germinating spores in culture. In some cases a large pyriform uniflagellate gamete is formed in germination, while in others the content of the spore emerges, grows, and then divides into as many as 20 minute gametes. Both types of gametes may fuse in pairs and form zygotes, but sometimes a large number of microgametes which have not completely separated reunite to form a plasmodium. Jones furthermore reported that during the chromidial stage a whole plasmodium may break up into gametes which subsequently fuse in pairs, as is shown in text-figure 3. However, his account of the life cycle of *P. Brassicae* is so unorthodox and confused that most later workers have seriously questioned the accuracy of his observations. As noted elsewhere, Cook and Schwartz ('30) maintained that the small flagellate cells produced in zoosporangia are gametes which fuse in pairs either in the root hairs of the host or after migrating into the cortex. They regarded these small zoospores as comparable to the minute gametes reported by Jones. Cook and Schwartz, however, never observed actual fusion, and their hypothesis is based entirely on the observation of zoospores lying side by side in pairs and the subsequent occurrence of binucleate amoebae. Obviously, neither of these phenomena are conclusive proof of fusion. According to these workers, the zygote thus formed develops into the sporogenous plasmodium, and reduction occurs during the first sporogonic division. As is shown in text-figure 4, the diploid generation thus embraces only the zygote and sporogenous plasmodium. Cook and Schwartz were uncertain whether the gametes come from the same or from different gametangia. If sex is genotypically segregated at meiosis, the resting spores, zoospores, haploid plasmodia, gametangia, and gametes are of two types, as is indicated in text-figure 5. Fedorintschik confirmed Cook and Schwartz's report of fusion of gametes from zoosporangia or gametangia but believed that it occurs later, following a period of vegetative budding within the host. As noted before, he also believed that two reductions occur in *P. Brassicae*—one during the first division of the sporangium nucleus and another at sporogenesis. Fedorintschik may have been influenced by a previous report by Cook of two similar reductions in *Ligniera*. If two reductions occur, obviously there must be two nuclear fusions, but Fedorintschik reported only one. No additional or more convincing evidence of sexuality in *Plasmodiophora* has since been presented as far as the author is aware, and the question thus remains in this uncertain state. It will doubtless remain thus until intensive monospore studies have been made.

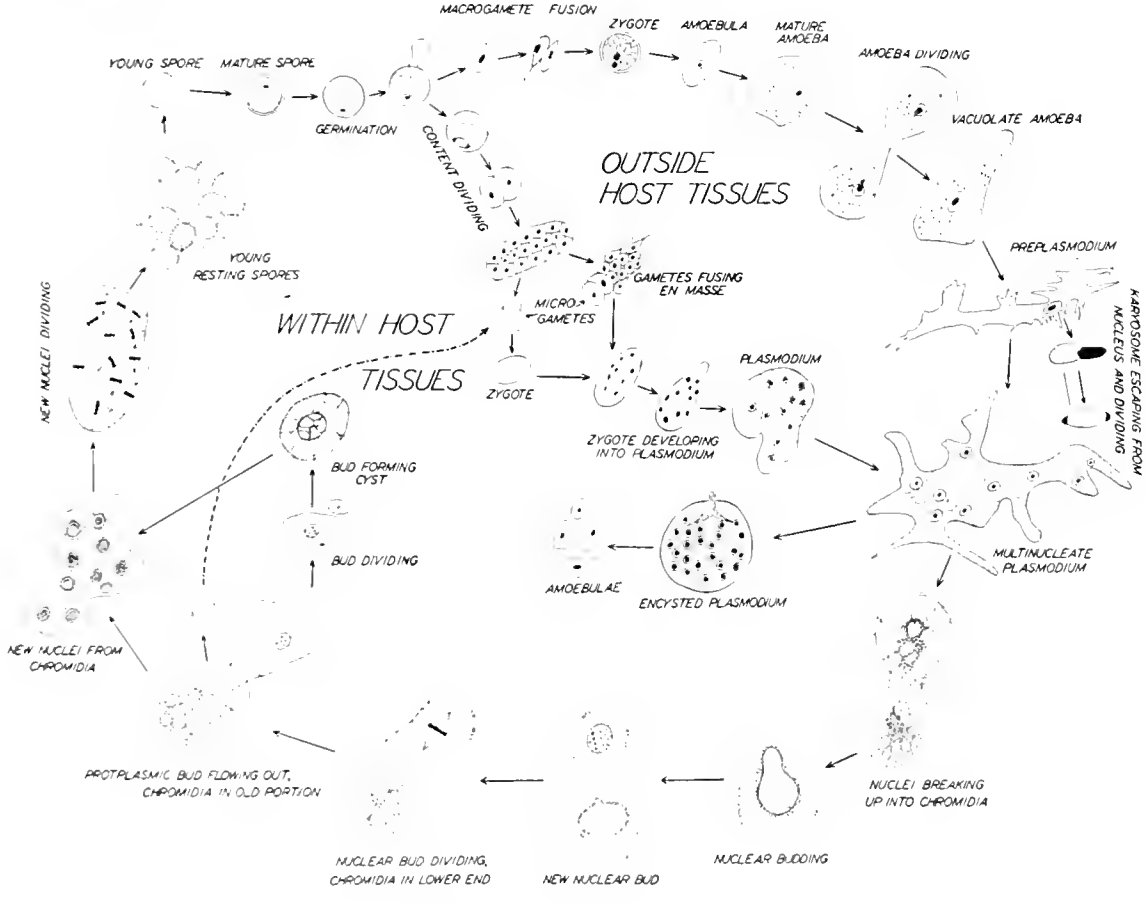
In *Tetramyxa*, Cook ('33) reported that "swarm cells" fuse in pairs at their anterior ends as they migrate from cell to cell and thus form amoeboid zygotes in which karyogamy soon occurs. These observations were apparently made from slides of fixed material furnished by Prof. O. V. Darbishire and do not relate to fresh material. No other data on sexuality in this genus exist so far as the author is aware. According to Cook, *T. parasitica* has a distinct alternation of haploid and diploid generations, as is illustrated in text-figure 6.

Nothing definite is known about sexuality in *Sorosphaera*. Cook ('33, p. 198) stated that amoebae from resting spores fuse in pairs and form amoeboid zygotes, but this statement is not based on observation. No one has yet reported actual observation of gametic fusion. However, inasmuch as reduction is said to occur at sporogenesis, most workers have nevertheless assumed that plasmogamy and karyogamy take place at some stage of development. Webb found no evidence of plasmogamy but reported that the chromosome number is doubled during the transitional phase. He thus concluded that karyogamy occurs at this stage, as Horne had previously described for *Spongospora*. According to Webb, the diplophase of *Sorosphaera* is very short and includes only the

TEXT-FIG 1. LIFE CYCLE OF P. BRASSICAE, ACCORDING TO PROWAZEK, 1905.



TEXT-FIG 2. LIFE CYCLE OF P. BRASSICAE, ACCORDING TO WINGE, 1913.



TEXT-FIG 3. LIFE CYCLE OF P. BRASSICAE, ACCORDING TO JONES, 1928.

maturation stages of the plasmodium and sporogenesis, while in Cook's opinion it extends from the time of gametic fusion through schizogony and "akaryosis" to sporogenesis, as is shown in text-figure 7.

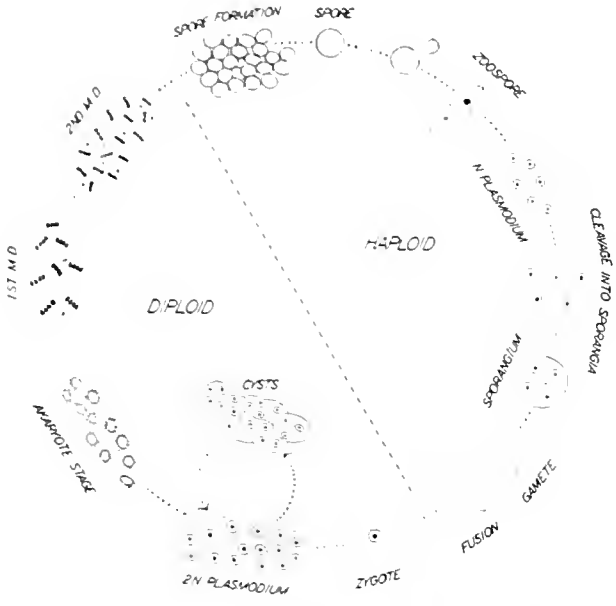
The data relative to sexual reproduction are even more scanty in *Sorodiscus*. Winge assumed, as he had for all members of the Plasmodiophoraceae, that gametes from germinating spores copulate in pairs and thus initiate the diploid phase of *S. Callitrichis*, but he never actually observed fusion. Likewise, plasmogamy and karyogamy have not been seen in *S. karlingii*. In *S. radicicolus*, however, Cook ('31) figured and described fusion of amoebae in pairs within the host cell (Pl. 7, fig. 6). The two gametes here figured are unequal in size, but Cook did not say whether or not this species is heterogamous. His study was made on fixed material sent from South Africa, and figure 6 shows the only case of pairing observed in such material. This may possibly represent only a chance association of amoeba without sexual significance. Obviously, additional data are needed before definite conclusions can be drawn about sexuality in *Sorodiscus*. Cook ('33), nevertheless, believed that fusion of gametes occurs in *S. radicicolus* and that this species has a well-defined alternation of diploid and haploid generations as is shown in text-figure 8.

In *Spongospora subterranea*, Massee, Kunkel, and Osborn reported that the sporogenous plasmodium is formed by coalescence of numerous amoebae, but they were uncertain about the origin and sex of the latter. Horne was of the opinion that the amoebae are of opposite sex and that in this respect the plasmodium is similar to that of *Dictyostelium mucoroides* reported by Skupienski ('18). According to Osborn, coalescence is followed by the akaryote stage during which the nuclei disappear. New nuclei are reconstructed *de novo*, and these subsequently associate in pairs (Pl. 10, fig. 28). Karyogamy soon follows as the nuclear membranes break down at the points of contact (fig. 29). Nuclei which do not pair degenerate the manner described by Jahn for *Ceratiomyxa*. Horne confirmed Osborn's report of karyogamy before meiosis but maintained that it occurs during instead of after the transitional or akaryote stage. He did not observe paired and fusing nuclei but based his conclusion on the discovery that the chromosome number following the transitional stage is twice that in amoebae and young plasmodia. According to Osborn and Horne, the diploid phase of *S. subterranea* is quite short in duration and includes only the sporogenous plasmodium, as is shown in text-figure 9. Osborn's, and to some extent Horne's, observations and reports of karyogamy shortly before sporogenesis in *Spongospora* are strikingly similar to the earlier accounts of the nuclear fusion in the myxomycetes. In *Ceratiomyxa, Arcyria,* and *Trichia,* Olive ('07), Kränzlin ('07), and Jahn ('07, '08) described nuclear pairing and fusion in the plasmodium shortly before resting spores are delimited, but these accounts have subsequently been refuted.

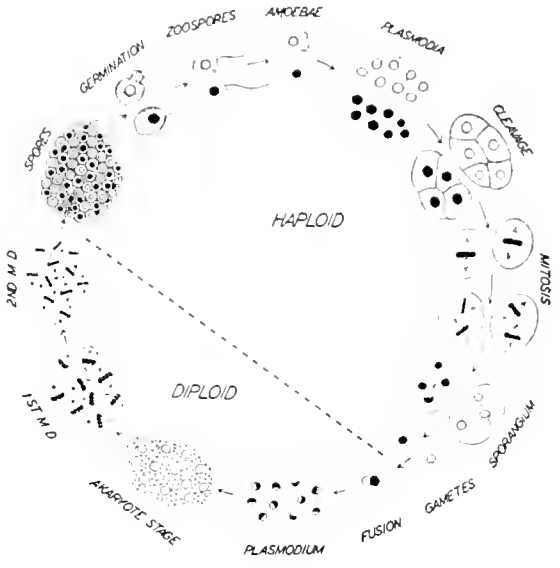
Cook ('33), on the other hand, reported that the zoospores from germinating resting spores pair at the anterior end, retract their flagella, and fuse (Pl. 10, fig. 20–22). Plasmogamy is followed shortly by nuclear pairing and fusion (fig. 22). The zygote may become flagellate again, and later, apparently, infects the host. Its nuclei divide promitotically, according to Cook, and at the 6- or 8-nucleate stage the zygote undergoes schizogony. Whether or not the meronts later coalesce and thus form the sporogenous plasmodium is not apparent from this account. Ledingham ('35) also observed germination of resting spores and formation of biflagellate zoospores, but he found no evidence of gametic fusion. A few binucleate zoospores with four flagella were present in Ledingham's cultures (Pl. 10, fig. 9), but he was not certain whether these were the product of fusion or incomplete cleavage. Thus, Cook's report of isogamy has not been substantiated. He nevertheless believed that the diploid generation of this species embraces the zygote, schizonts, meronts, and plasmodia, while the haploid phase is limited to the cystosori, resting spores, and gametes, as is shown in text-figure 10. The zoosporangia and zoospores found by Ledingham are apparently a means of rapid vegetative multiplication and doubtless relate to the haploid phase, as is indicated in this diagram. Barrett found fusion stages between zoospores or gametes derived from zoosporangia in *S. Cotulae,* but these relate only to fixed and stained preparations.

In *Ligniera,* Maire and Tison, and other workers, assumed that plasmogamy and karyogamy take place at some stage of development, because the nuclei appear to undergo reduction at sporogenesis. Cook ('26, '33), however, reported a double fusion and reduction in *L. Junci*. The zoospores from resting spores fuse in pairs at the anterior end and give rise to diploid plasmodia. As noted before, these cleave into uninucleate segments, which develop walls and become incipient zoosporangia. The first nuclear division in these sporangia is meiotic, and the zoospores or gametes subsequently produced are haploid. These fuse in pairs and form the diploid sporogenous plasmodium in which meiosis later occurs at sporogenesis. *Ligniera Junci* thus has two diploid phases each of which is separated by a haploid phase, according to Cook, as is illustrated in text-figure 11. Cook neither observed plasmogamy and karyogamy nor counted the chromosomes at meiosis, so that he had no direct evidence for his assumption. It is not improbable that the zoosporangia and zoospores are merely means of vegetative multiplication without sexual significance and relate to the haploid generation, provided an alternation does occur, in much the same manner as is indicated in text-figure 10 of *S. subterranea*.

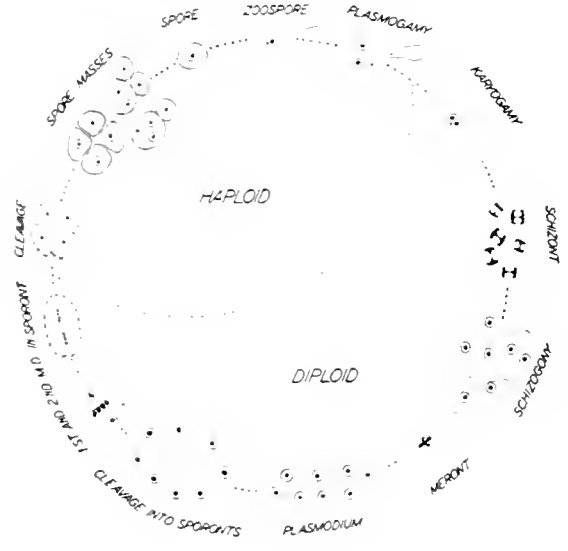
No direct evidence of gametes, gametic fusion and karyogamy have been observed in *Membranosorus, Polymyxa, Octomyxa,* and the doubtful genera, *Rhizomyxa, Anisomyxa,* and *Sorolpidium*. Ledingham found a few tetraflagellate binucleate zoospores in *Polymyxa,* but he was not certain whether these were



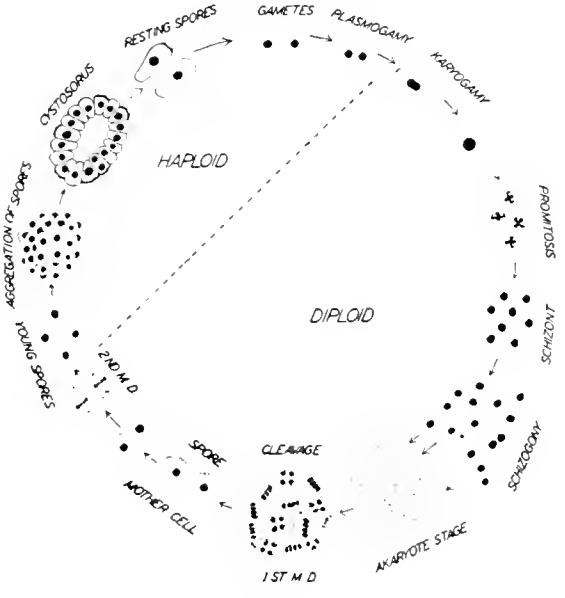
TEXT-FIG 4 LIFE CYCLE OF P BRASSICAE, ACCORDING TO COOK AND SCHWARTZ, 1930



TEXT-FIG 5 LIFE CYCLE OF P. BRASSICAE, SUGGESTED BY COOK AND SCHWARTZ'S STUDIES, 1930.



TEXT-FIG 6 LIFE CYCLE OF TETRAMYXA, ACCORDING TO COOK, 1933



TEXT-FIG 7 LIFE CYCLE OF SOROSPHAERA, ACCORDING TO COOK, 1933.

the products of gametic union or incomplete cleavage. Tetraflagellate zoospores were likewise found by Couch *et al.* in *Octomyxa*, but no fusions were observed. However, in this genus as well as in *Membranosorus* Whiffen and Wernham each reported reduction at sporogenesis, which presupposes karyogamy at some state of development. Miss Whiffen believed that karyogamy occurs during the akaryote stage of *O. Achlyae*.

It is obvious from this review that the data on sexuality in the Plasmodiophorales are quite limited. In *S. subterranea*, the only species in which gametic fusion has actually been observed, the respective gametes are reported to be alike and show no structural, mobile, and physiological differences. In this species at least sexual reproduction appears to be isogamous. Whether it is homo- or heterothallic is not known, since no studies involving monospore cultures have yet been made. Therefore, any discussion at present of sex determination, haplosynoecism, haploheteroecism, antithetic alternation of gameto- and sporophytic generations, etc., in the Plasmodiophorales must be speculative and, in light of the meager present-day knowledge, largely futile.

BIBLIOGRAPHY: CYTOLOGY AND SEXUALITY

Alexieff, A. 1913. Arch. Protistk. 29: 344.

Bělař, K. 1926. Ergeb. Fortschr. Zool. 6: 235.

Blomfield, J. E., and E. J. Schwartz. 1910. Ann. Bot. 24: 35.

Calkins, G. N. 1933. Biology of the protozoa. Philadelphia.

Chatton, E. 1910. Arch. zool. Exp. 5 ser. 5: 239, 267.

Cook, W. R. I. 1926. Trans. Brit. Mycol. Soc. 11: 196.

———. 1928a. New Phytol. 27: 230, 298.

———. 1928b. Ann. Bot. 42: 347.

———. 1931. Ann. Mycol. 29: 321.

———. 1933. Arch. Protistk. 80: 179.

———, and E. J. Schwartz. 1929. Ann. Bot. 43: 81.

———, and ———. 1930. Philos. Trans. Roy. Soc. London 218 B: 283.

Couch, J. N., J. Leitner, and A. Whiffen. 1939. Jour. Elisha Mitchell Sci. Soc. 55: 399.

Doflein, E. 1916. Lehrbuch der Protozoenkunde 4th ed. Jena.

Favorsky, W. 1910. Mem. Soc. Nat. Kieff 20: 149.

Ferdinandsen, E., and O. Winge. 1920. Ann. Bot. 34: 467.

Fedorintschik, N. S. 1935. Summ. Sci. wk. Inst. pl. protect. Leningrad 1935: 69.

Harper, R. A. 1900. Bot. Gaz. 30: 217.

———. 1914. Amer. Jour. Bot. 1: 127.

Horne, A. S. 1911. Rept. Brit. Assoc. Adv. Sci., Portsmouth, p. 572.

———. 1930. Ann. Bot. 44: 199.

Jahn, E. 1907. Ber. Deut. Bot. Gesell. 25: 23. 1908, *Ibid.* 26a: 342.

Jones, P. M. 1928. Arch. Protistk. 62: 313.

Kränzlin, H. 1907. *Ibid.* 9: 170.

Ledingham, G. A. 1939. Canad. Jour. Res. C, 17: 38.

Levine, L., and M. Levine. 1922. Jour. Cancer Res. 7: 163, 171.

Lutman, B. F. 1913. Vermont Agr. Exp. Sta. Bull. 175.

Maire, R., and A. Tison. 1909. Ann. Mycol. 7: 226.

———, and ———. 1910. Bull. Soc. Linn. Normandie 6 ser. 2: 57.

———, and ———. 1911. Ann. Mycol. 9: 226.

Milovidov, P. F. 1931. Arch. Protistk. 73: 1.

———. 1932. C. R. Soc. Biol. 109: 170.

———. 1933. Arch. Protistk. 81: 138.

Nägler, K. 1909. *Ibid.* 15: 1.

Nawaschin, S. 1899. Flora 86: 404.

———. 1901. Kiev Lap. Obsch. Jest. 17: 1: XXXVI.

———. 1924. C. R. Acad. Sci. Russie 1924: 173.

Nemec, B. 1911. Bull. Inter. Empr. Fran. Joseph Acad. Sci. 16: 69. 1913, *Ibid.* 18: 18.

Olive, E. W. 1907a. Trans. Wisc. Acad. Sci., Arts and Letters 15: 753.

———. 1907b. Sci. n. s. 25: 266.

Osborn, T. G. B. 1911a. Rept. Brit. Assn. Adv. Sci. Portsmouth, p. 572.

———. 1911b. Ann. Bot. 25: 271, 327.

Palm, B. T., and M. Burk. 1933. Arch. Protistk. 79: 263.

Pavillard, J. 1910. Prog. Rei. Bot. 3: 475.

Prowazek, S. 1902. Österr. Bot. Zeitschr. 52: 213.

———. 1905. Arb. Kais. Gesundheit 22: 396.

Schwartz, E. J. 1910. Ann. Bot. 24: 511.

Skupienski, F. X. 1918. C. R. Acad. Sci. Paris 167: 960.

Terby, J. 1923. Mem. Roy. Acad. Belg. 7: 1.

———. 1924. Bull. Roy. Acad. Belg. 5 ser. 10: 519.

———. 1932. Mem. Roy. Acad. Belg. 11: 1.

Webb, P. C. R. 1935. Ann. Bot. 49: 41.

Wenyon, C. M. 1926. Protozoology 1: 66.

Wernham, C. C. 1935. Mycologia 27: 262.

Whiffen, Alma. 1939. Jour. Elisha Mitchell Sci. Soc. 55: 243.

Winge, O. 1913. Ark. f. Bot. 12, no. 9: 1.

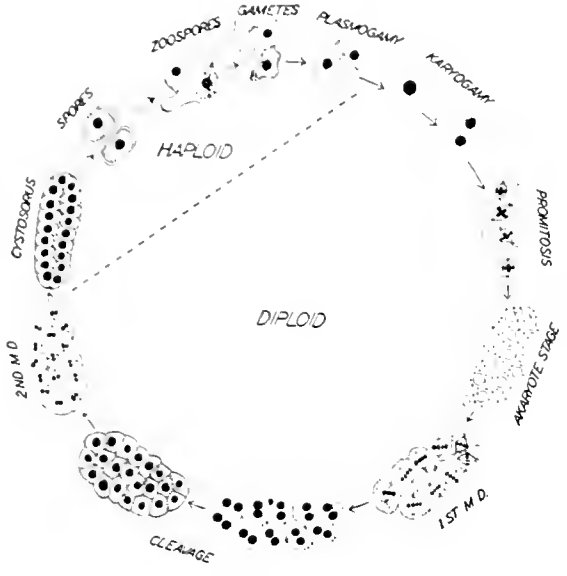
Wissenlingh, C. 1898. Jahrb. Wiss. Bot. 31: 619.

Yendo, Y., and K. Takase. 1932. Bull. Sericult and Silk Inc. Japan 4, no. 3: 4.

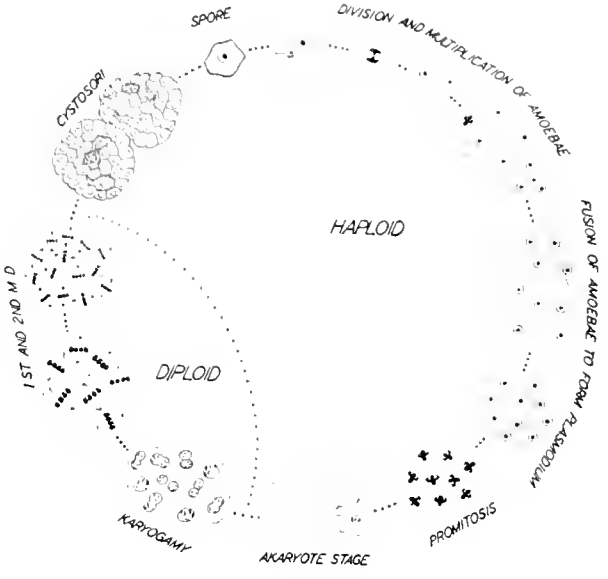
Chapter IV

Classification and Description of Species

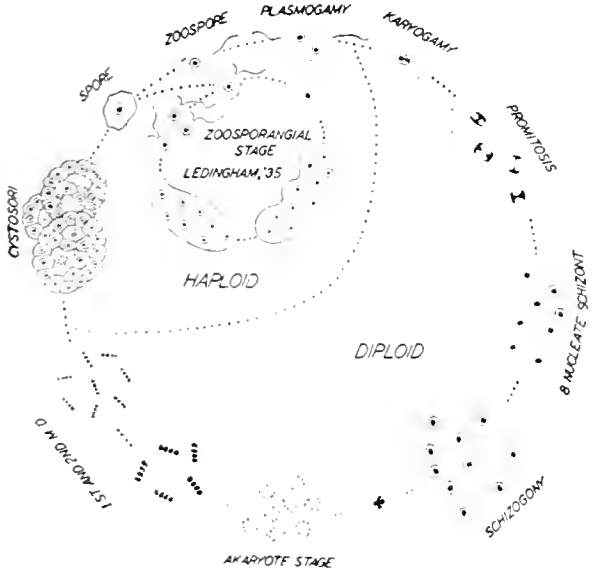
The Plasmodiophorales include one family, Plasmodiophoraceae, and approximately eight genera and twenty-three species. Numerous other genera and species have been added at various times, but these have either been merged with existing genera or excluded entirely as invalid. A natural classification is well nigh impossible at present because so little is known about the critical diagnostic characters of most species. Furthermore, the genera are not sharply defined and, as Palm and Burke ('33) have so well emphasized, tend to merge and overlap, so that in certain members generic distinctions are difficult to recognize. The oldest and most frequently used criterion of classification is the grouping assumed by the resting spores at maturity. This criterion was introduced by Schroeter in 1897, who separated the genera on the basis of whether the spores are free or united in clusters and cystosori. Schroeter also emphasized the presence or absence of a soral membrane as a distinctive character of *Tetramyxa* and



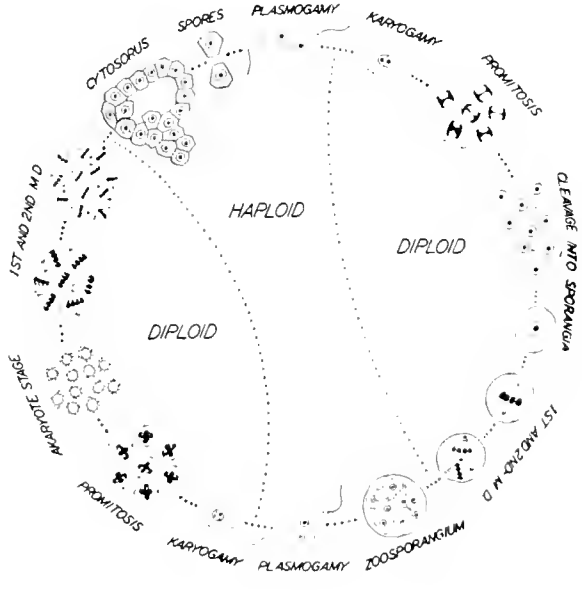
TEXT-FIG 8 LIFE CYCLE OF SORODISCUS RADICICOLUS. ACCORDING TO COOK, 1931.



TEXT-FIG.9 LIFE CYCLE OF SPONGOSPORA SUBTERRANEA, ACCORDING TO OSBORN, 1911.



TEXT-FIG 10 LIFE CYCLE OF S. SUBTERRANEA, ACCORDING TO COOK, 1933



TEXT-FIG 11 LIFE CYCLE OF LIGNIERA JUNCI. ACCORDING TO COOK, 1933.

Sorosphaera. Although mycologists and protozoologists have clearly recognized the inadequacy of these criteria, they have nevertheless continued to use them as the basis of classification. More recently, Cook ('33) has used the presence of zoosporangia and zoospores as another basic distinction. However, since zoosporangia have been subsequently found in numerous genera, the mere presence of such structures is no longer generically distinctive. Likewise, his emphasis on the presence or absence of a membrane around the cystosori as a diagnostic character is open to question, since there is considerable doubt about the occurrence of soral membranes in any of the genera. Palm and Burke, in particular, have severely criticized the present-day system of classification and characterized it as artificial. From their observations on the wide variations exhibited by cystosori of *S. Veronicae*, they concluded that *Spongospora*, *Ligniera*, *Sorodiscus*, *Ostenfeldiella*, *Clathrosorus*, and *Membranosorus* should be regarded as synonyms of *Sorosphaera*. On the basis of similarity of life cycles and general structure, they further advised the merging of all known genera except *Cystospora* into one large genus, presumably *Plasmodiophora*. The author is in complete agreement with these workers on the low taxonomic value and inadequacy of present-day generic distinctions. However, Palm and Burke's suggestion of reducing the number of genera or merging them does not solve the difficulties of classification in this group. As Ledingham ('39) pointed out, it merely shifts the generic indistinctions to the species.

Further taxonomic distinctions appear to be emerging from the discovery of zoosporangia in old and new genera. When these developmental stages have been fully investigated, the relationship of the various genera will doubtless become clearer, and it may then be possible to separate or merge them with greater accuracy. In the meantime, Schroeter's system of classification serves as a working basis, and although an unsatisfactory expedient, it may be used to advantage. In the key which follows, size, number, and shape of zoosporangia are used to some extent in diagnosis, but these characters are of doubtful generic value. Some of the genera—*i.e.*, *Membranosorus* and *Ligniera*—listed here are obviously questionable and should perhaps be merged with *Sorodiscus* and *Sorosphaera*, but until more is known about the family as a whole, it may be worth while to treat them separately.

PLASMODIOPHORACEAE

Zopf, 1884. Die Pilzthiere oder Schleimpilze

Thallus a naked, plasmodial, multinucleate protoplast capable of amoeboid movement and undergoing schizogony into uni- or multinucleate meronts, which in turn may function as schizonts. Sporogenous thallus cleaving into uninucleate spores at maturity. Resting spores loose and free or united in small clusters and cystosori; usually producing one zoospore or amoeba in germination. Zoospores anteriorly biflagellate and heterocont. Zoosporangia formed directly from zoospores or cleavage segments of young plasmodia; free or united in sporangiosori; producing a few to numerous zoospores which are similar to those formed from resting spores.

Key to Genera

I. Resting spores not united, free and loose. Zoosporangia few or numerous, small, and producing few zoospores 1. **PLASMODIOPHORA**, p. 22.
II. Resting spores united in small clusters or more or less compact cystosori
  A. Spores usually in tetrads or dyads. Zoosporangia unknown 2. **TETRAMYXA**, p. 37.
  B. Spores usually in octads. Zoosporangia numerous, small, oval, and spherical with or without exit papillae 3. **OCTOMYXA**, p. 40.
  C. Cystosorus predominantly spherical to ellipsoidal and hollow; often variable in size and shape. Zoosporangia small 4. **SOROSPHAERA**, p. 41.
  D. Cystosorus predominantly disc-shaped, two-layered and flattened; often variable in size and shape. Zoosporangia unknown 5. **SORODISCUS**, p. 46.
  E. Cystosorus oval, spherical, and sponge-like, lacking a central cavity but traversed by prominent canals and fissures. Zoosporangia numerous or few, small, oval and spherical, or large and irregular 6. **SPONGOSPORA**, p. 54.
  F. Cystosorus indefinite in size and shape
    1. Zoosporangia small, oval and spherical; producing few zoospores 7. **LIGNIERA**, p. 58.
    2. Zoosporangia usually large, elongate, lobed and irregular with prominent exit tubes 8. **POLYMYXA**, p. 63.

PLASMODIOPHORA

Woronin, 1877. Arb. St. Petersburg Nat. Gesell. 8: 169.

Ostenfeldiella Ferdinandsen and Winge, 1914. Ann. Bot. 28: 64.

(PLATES 2, 3, 4.)

Resting spores lying free in host cell, not united in cystosori, variable in size and shape, usually producing one zoospore in germination. Zoospores anteriorly biflagellate and heterocont, becoming intermittently amoeboid, infecting the host as an amoeba (?), dividing and budding (?), and eventually forming multinucleate plasmodia, which cleave into uninucleate segments. Cleavage segments developing into small zoosporangia which produce few zoospores. Secondary zoospores reinfecting host and forming additional plasmodia. Sporogenous plasmodium partly or completely filling host cell, moving slowly in amoeboid fashion within the host cell and in migrating from cell to cell; occasionally undergoing schizogony into uni- and multinucleate meronts; rarely encysting; cleaving into resting spores at maturity.

Plasmodiophora includes at present five species, of which only one, *P. Brassicae*, is fairly well known. Most of the other species are so little known that their validity as members of the genus has been seriously questioned. They nevertheless possess the common ability of causing conspicuous galls and malformations of the host tissues. Numerous other organisms with plasmodial stages have been included in the genus from time to time, but careful reinvestigation has shown them to be invalid. *Plasmodiophora* is distinguished from the other genera of the family by the lack of a distinct cystosorus. The resting spores are not united or attached to form a sorus of definite size and shape but lie loose in the host cell, as is shown in figures 88 and 100. When the host cell disintegrates, the spores are liberated into the soil where they may germinate at once, as in *P. Brassicae*, or remain viable up to seven or eight years (Jorstad, '23). So little is known about the other species of *Plasmodiophora* that present day discussions of the genus must necessarily be based principally on *P. Brassicae*. Although this species has been intensively studied for more than 50 years, there is still considerable disagreement and controversy about its critical developmental stages, and doubtless much remains to be discovered.

The resting spores of *P. Brassicae* normally give rise to a single zoospore in germination (fig. 13–17), but it is not improbable that more than one may be produced by the occasional large, bi- and multinucleate resting spores. Some workers, including Pollacci ('12) and Honig ('31), have seriously questioned the production of zoospores in this species. Honig, in particular, maintained that only non-flagellate amoebae are formed in germination (fig. 16–18). Also, most investigators have figured and described the zoospores as anteriorly uniflagellate, but Ledingham ('34) clearly demonstrated that they are biflagellate and heterocont (fig. 22, 23). They have also been described in the literature as varying from oval, pyriform, and fusiform to spherical in shape (fig. 19–23). After emerging from the spore case they may swim rapidly away or become intermittently amoeboid (fig. 19, 20), during which the anterior end may double back and forth and thus jerk the spore body along.

According to most students, the zoospores come to rest on the host and enter as amoebae through the root hairs and epidermal cells, where they soon cause local hypertrophy (fig. 28, 29). A few workers, however, have questioned these observations. Kunkel ('18) found nothing but thalli of *Olpidium Brassicae* in the root hairs of the host species which he studied and like Favorski ('10) concluded that the previous reports on the occurrence of *P. Brassicae* in such host cells were erroneous. However, subsequent investigators, including Cook and Schwartz ('30), Honig ('31), Rochlin ('33), and Fedorintschik ('35) have clearly shown infection of root hairs. Honig appears to have been the first to observe, describe, and figure actual penetration of the parasite into root hairs (fig. 27). He maintained that the protoplasts derived from germinating resting spores are true amoebae without flagella, a contention which has been refuted by subsequent observers. Honig found small amoebae as well as giant ones measuring 1–26 μ by 24–36 μ abundant around root hairs and observed that both types may readily enter the host cell. In so doing they become closely applied to the root hair, and soon thereafter a hole appears in the wall at the region of attachment, through which they then enter. The hole closes up immediately afterwards, so that it is no longer visible after the parasite has entered. Honig also observed that amoebae may live saprophytically for weeks in the soil and increase markedly in size (fig. 35).

It is not improbable, as Rochlin's ('33) study suggests, that under certain environmental conditions and particularly when spores germinate in contact with the host cell, flagella are not formed, and the parasite enters the host almost at once in the amoeboid state. Rochlin found that the resting spores become attached to the root hairs and epidermal cells of the root and cap and cause localized swelling of the cell wall (fig. 24–26). These regions become gelatinized and show no cellulose reactions when tested with chloro-iodide of zinc, indicating that a chemical change has taken place. Small plastic, spherical protoplasts which presumably emerge from the attached spores, pass through these swollen and gelatinized regions (fig. 26) and enter the host cell.

The amoebae and young plasmodia in the host cell may bud and divide repeatedly (fig. 30), according to Gaylord ('04), Chupp ('17), Kunkel, Fedorintschik and others, and thus multiply in number. They may also encyst and develop fairly thick hyaline walls (fig. 44–46) under unfavorable conditions. As the plasmodia grow in size and their nuclei multiply, they penetrate the walls of the adjacent cells and thus migrate from cell to cell (fig. 31–33), according to Woronin ('78), Lutman ('13), Chupp ('17), Kunkel, Honig, Rochlin, Larson ('34), and Fedorintschik ('35). As to the method of cell wall penetration, Rochlin noted that the plasmodium may become closely applied to a region of the wall and through lysic action cause localized swelling and gelatinization (fig. 7) of the latter. Passages in the walls are thus formed through which the plasmodium enters. Kunkel believed that only young and small plasmodia free of oil bodies and other products are capable of migration. Cook and Schwartz ('30) were uncertain on this phase of development and somewhat vague in their description of it. In one part of their paper (p. 287) they stated that the amoebae "have the power of penetrating the walls of the host cells and in this way can travel through the cortical tissues of the host," but they thought it improbable (p. 297) that the plasmodia are able to do so. Finally (p. 301) they expressed the belief that only gametes from sporangia have the ability of passing from cell to cell, and after gametic fusion the zygote is distributed only by division of infected host cells.

Cook and Schwartz, nonetheless, discovered a hitherto unknown stage in the life cycle of *P. Brassicae*. The amoebae derived from flagellate zoospores penetrate root hairs, grow in size, and by regular mitosis become multinucleate plasmodia which soon cleave into uninucleate portions. These segments round up, develop thin hyaline walls, and become incipient zoosporangia (fig. 37). In view of this discovery, it seems probable that the small cleaving plasmodia which Chupp described and figured (p. 436, fig. 104H) as stages in resting spore formation in root hairs relate to the development of zoosporangia. The nuclei of the zoosporangia (fig. 38) divide mitotically two or three times, after which the protoplasm cleaves into uninucleate segments (fig. 39–42), forming thus four to eight pyriform zoospores (fig. 43). These are smaller than those derived from resting spores, according to Cook and Schwartz, who regarded them as gametes. As the walls of the sporangia collapse the zoospores emerge, fuse in pairs, either in root hairs or after migrating into the cortex, and form zygotes which grow into diploid sporogenous plasmodia, as has been described in Chapter III.

Fedorintschik confirmed Cook and Schwartz's discovery of zoosporangia. He reported that individual amoebae in root hairs develop directly into large plasmodia containing up to 100 or more nuclei. These plasmodia cleave into uninucleate segments which develop walls and become rudimentary zoosporangia. The first division of their nucleus is *meiotic*, and then follow a second and sometimes a third mitosis, after which the protoplasm cleaves into four to eight zoospores. A single amoebae in a root hair may, according to Fedorintschik's observations, ultimately result in the formation of 100 to 800 zoospores. These zoospores become amoeboid and migrate into the cortical tissues and multiply rapidly by budding. After the content of the host cell is exhausted, they fuse, presumably in pairs (?), and later develop into plasmodia. Fedorintschik believed this fusion constitutes the sexual phase of *P. Brassicae*, and thus confirmed Cook and Schwartz's earlier report of sexual reproduction in this genus. In light of Ledingham's ('35, '39), Couch, Leitner, and Whiffen's ('39) studies on *Spongospora, Polymyxa*, and *Octomyxa*, however, it seems more probable that these so-called gametes are only secondary zoospores which reinfect the host and give rise to an additional amoebae and plasmodia in much the same manner as is indicated in text-figure 10 of *Spongospora*.

Several workers have reported that the plasmodium of *P. Brassicae* may undergo schizogony and give rise to a few or several meronts, whereby the parasite is rapidly multiplied. Nawaschin ('99) did not actually observe the process, but he believed that the large number of small thalli in a host cell could be explained only on the assumption that they had arisen by division of a preexisting thallus. He thought that the extended pseudopodia of the plasmodium were cut off as buds, a belief which was later supported by Lutman and by Henckel ('23). Subsequently, Maire and Tison ('09), Chupp, Kunkel, Jones ('28), and Cook ('33) also reported schizogony of the plasmodium of *P. Brassicae*. It must be noted, however, that many of the early described cases of schizogony in the superficial host cells may possibly relate to the development of zoosporangia. As is shown in figure 36, the meronts may be uni- or multinucleate, and it is not improbable that after a period of growth they in turn may function as schizonts and form secondary meronts.

With the exception of Cook and Schwartz and Fedorintschik who reported that the sporogenous plasmodium is formed by the fusion of two gametes, many investigators who studied this phase of development were of the opinion that the plasmodium arises by the union of several vegetative amoebae or small plasmodia. Woronin was uncertain whether it originates from a single amoebae or by the fusion of several, although he thought the latter method more plausible. Eyeleshymer ('01) observed that if a slide with zoospores and amoebae is kept in a moist chamber, larger plasmodia appear, which he assumed had arisen by fusion of amoebae. Honig, however, maintained that the amoebae observed by Eyeleshymer do not relate to *P. Brassicae*. Halsted ('93) also believed that amoebae coalesce to form large plasmodia. Nawaschin ('99) thought that the schizonts and meronts remain more or less independent in the host cell until shortly before sporogenesis, when they flow together and form a large plasmodium. He admitted also that single amoebae may grow independently into large plasmodia. Gaylord, Ericksson ('13), Esmarch ('24), Prowazek, and Terby ('24) supported Nawaschin's belief on the union of amoebae, but Maire and Tison ('09) refuted this contention. They pointed out that although meronts and schizonts may appear to be fused, they are nonetheless separate and distinct. They based their view primarily on the lack of synchronism in nuclear division in the closely associated amoebae and plasmodia in the same host cell. Lutman, Chupp, and Kunkel were uncertain about the union of amoebae, but Lutman noted that the nuclei in a plasmodium do not all divide simultaneously, which suggests that they may have been derived from several amoebae of different ages. Later, Jones ('28a, '28b) also reported fusion of amoebae and plasmodia in cultures of *P. Brassicae*, but there is considerable doubt about the validity of the organism he had in culture. In addition to describing the origin of the plasmodium from a zygote, Cook and Schwartz reported that in the early stages of development several amoebae and later small plasmodia may fuse vegetatively to form the incipient sporogenous plasmodium. Since that time Milovidov ('31) also reported vegetative fusion of several amoebae.

The plasmodium of *P. Brassicae* is capable of slow amoeboid movement, and this mobility apparently enables it to move from cell to cell. Rochlin reported that the plasmodium first sends out a hyaloplasmic thread (*Geissel*) in the direction of movement, and shortly thereafter the more granular mass begins to

move. In young plasmodia the pseudopods are relatively long and tenuous, but as the plasmodium matures, they become less extensive and more rounded at the periphery. Figure 34 shows a mature plasmodium with several dense, opaque, pseudopodial lobes at the anterior end. The posterior end in contrast is quite vacuolate, thin, and relatively hyaline. The amoebae and young plasmodia are hyaline, somewhat transparent, viscous and slimy, and comparatively free of oil droplets and other bodies, but as the plasmodium increases in size, the protoplasm becomes denser, more opaque, and very rich in oil globules. Infected hypertrophied host cells are often rich in starch grains, and according to Woronin, Nawaschin ('99), Prowazek ('05), and Lutman, these grains may frequently be found in the folds of the plasmodium. Nawaschin, Favorsky ('10), and Henckel ('23) did not believe that amoebae and plasmodia are capable of engulfing solid particles, and Nawaschin pointed out that starch grains, such as those shown in figure 74, are often caught between fusing meronts and thus come to lie within the plasmodium. Woronin, Eycleshymer, and Lutman inferred that the plasmodium feeds on these grains, because by the time sporogenesis begins they have almost entirely disappeared, although a few may occasionally be found later scattered among the spores. Although Honig did not observe the plasmodium engulfing solid particles, he nevertheless described it as nourishing itself saprophytically outside of the host for several weeks. In addition to oil globules, starch grains, and other bodies, chondriosomes are quite abundant in the plasmodia (fig. 48), according to Vonwiller ('18) and Milovidov ('31). They also occur abundantly in the resting spores (fig. 86) and amoebae.

Under unfavorable environmental conditions plasmodia and segments of the same in *P. Brassicae* may encyst and develop thick walls, according to Prowazek, Cook and Schwartz, and Milovidov (fig. 46, 47). Prowazek ('05) and Milovidov regarded these cysts as pathological and involution forms. Cook and Schwartz described the plasmodium as becoming enveloped by a distinct wall and then segmenting into several portions which in turn developed thick walls (fig. 47). With the return of favorable conditions the walls disappear, and the plasmodium continues to function normally. Encysted plasmodia have also been described in *P. Fici-repentis* by Andreucci ('26). The cysts in this species are globular, 9.15–73 μ in diameter, with sculptured, thick walls, and in germination give rise again to plasmodia. The significance of these cysts as a phase in the life cycle of *Plasmodiophora* is not clearly understood, but they are doubtless comparable with the sclerotia of the myxomycetes.

The majority of resting spores are uninucleate, but occasional globose and irregular ones (fig. 87) have been reported by Prowazek, Milovidov, and others. Milovidov, in particular, has figured numerous tetra-, tri-, and binucleate spores. The binucleate and multinucleate condition may have resulted from the failure of large cleavage segments to divide after the completion of the second nuclear division. On the other hand, it is not altogether improbable that it may have arisen as the result of a third mitosis in the incipient spore segments in the manner described by Maire and Tison and Horne in *Sorosphaera Veronicae* and *Spongospora subterranea*, respectively. Lutman ('13) and Terby ('24) also figured binucleate spores (fig. 95) and believed they had arisen as the result of division in the spore. After the spores have been formed they may remain stuck together for a short time by the slimy intercellular substance left from the plasmodium. However, they soon develop hyaline walls, dehydrate, and separate. At no stage are they enveloped by a common membrane or form a cystosorus of definite structure, size, and shape. According to the reports in the literature the resting spores may vary up to and more than 200 per cent in size. The early investigators found the spores to be quite small, but measurements by subsequent workers have shown them to be considerably larger. Woronin ('78) reported them to be 1.6 μ in diameter; Lowenthal ('05), 4 μ; Molliard ('09), 1.8–2.2 μ; Chupp ('17) and Appel ('28), 1.9–4.3 μ, and 2.5–6.9 μ for the irregular ones; Esmarch ('24), 1–2 μ; Pape ('25) and Hönigmann ('26), 2.8–3.3 μ; Wellman ('30), 1.7 μ; Cook and Schwartz ('30), 2–3 μ, and 4.6×6 μ for the oval ones; and Honig ('31), 3.9 μ. The last-named worker made extensive measurements from numerous hosts grown in different types of soil and under varying climatic conditions and found that the spores did not differ more than 0.5 μ in diameter. According to Wisseulingh ('98) the spore wall consists of chitin and shows no cellulose reaction when tested.

The account given above is generally considered to be the usual developmental cycle of *P. Brassicae*. Henckel and P. M. Jones ('28b), on the other hand, have reported life cycles for this species which vary markedly from the orthodox type. In his study of club root of radishes Henckel described the resting spores as "aplanoamoebae" which by a process of gelatinization or softening give rise to "limax amoebae." These multiply outside of the host by budding, and when this process is completed, the numerous daughter amoebae enter the host and form a plasmodium. At no stage in the life cycle are zoospores or flagellate gametes developed, according to Henckel. In connection with his account it may be noted that Favorsky also figured and described spore germination in rotten tumors as a process of gelatinization and softening of the spore wall, whereby large *limax*-like amoebae are formed. P. M. Jones reported that he had isolated eight pure cultures of *P. Brassicae* from cabbage roots and maintained them in tap water under laboratory conditions for two months. These cultures caused galls on turnips when used as an inoculum and were subsequently recovered in culture from the diseased roots. According to Jones (text-fig. 3), the following successive stages occur within the host: gametes, zygotes, preplasmodia, plasmodia, cysts and spores; while in culture outside

of the host, gametes, zygotes, cysts, amoebae, preplasmodia, and buds are formed. If conditions are favorable, however, *P. Brassicae* does not develop all of these phases. Jones' account has never been confirmed, and most subsequent investigators have doubted the accuracy of his observations. In light of present-day knowledge about *P. Brassicae* it seems likely that he may have been dealing with developmental phases of more than one organism. Milovidov and Honig contended that some of the stages figured by Jones relate to *Olpidium Brassicae* and *Astero-cystis radicis*.

P. BRASSICAE Woronin, l.c., pls. 1-6. 1878. Jahrb. Wiss. Bot. 11: 548. Pls. 29-34.

Resting spores globose, spherical 1.6–4.3 μ, average 3.9 μ, oval, ellipsoidal, 4.6×6 μ, sometimes constricted, elongate and irregular, 2.5–6.9 μ, with smooth, relatively thin, hyaline walls. Zoospores pyriform, spherical, 2.5–3.5 μ, swimming rapidly and becoming intermittently amoeboid. Sporangial plasmodia variable in size. Zoosporangia few or numerous, small, oval, spherical, 6–6.5 μ, angular and elongate with thin hyaline walls; producing 4 to 8 zoospores which are liberated by the collapse of the sporangium wall. Sporogenous plasmodia 100–200 μ in diameter, hyaline to pale-grey in color, amoeboid; encysting occasionally, undergoing schizogony into uni- and multinucleate meronts.

Parasitic in the roots of wild and cultivated crucifers in temperate climates throughout the world, causing spindle-shaped, globose and irregular swellings, or galls and occasionally dark sunken spots and lesions.

A complete list of hosts, degree of infection, geographical distribution, and bibliography of *P. Brassicae* are given in Chapter VI.

Biological Races of Plasmodiophora Brassicae

Marked differences in degree of infection have been found in various species and varieties of wild and cultivated crucifers, and this has led to the belief that *P. Brassicae* may include several biological races or strains which are more or less virulent and specific for certain hosts. Appel and Werth ('10), Ericksson ('13), Höstermann (according to Honig, '31), and Gleisberg ('23) suggested the existence of such races, and numerous attempts have been made to demonstrate their presence. Between 1924 and 1929 Honig made six experiments involving a large number of cruciferous hosts from which he ('31) reported positive results. A strain of *P. Brassicae* from kohlrabi was found to be readily transmissible to kohlrabi, cauliflower, rape, turnips, and *Cammelina sativa,* but could be transmitted only with difficulty to radishes. A cauliflower strain was also discovered which proved to be similar to the one on kohlrabi, but strains from Savoy cabbage and radish were found to be distinct. Motte ('33, '35) and MacLeod ('31) believed that they had obtained evidence of biological specialization, but later after making tests of spores from 50 different sources, the latter worker found no evidence to confirm this belief. Motte ('33) found that the form from charlock grew especially well on turnips. Gibbs ('31) likewise tested various inocula for evidence of specialization, but all of his results were negative. In 1939 J. C. Walker observed a high degree of resistance to club root in swedes in Wisconsin, which was contrary to results obtained elsewhere, and thought that this difference indicated a variation in pathogenicity of the causal organism. He accordingly secured spores from widely separated regions of the United States and tested their virulence on swedes, but found little difference in pathogenicity.[1] The data in the literature on the existence of biological strains are therefore conflicting, and most investigators, with the exception of Honig and Motte, have doubted the presence of such strains in *P. Brassicae*.

[1] However, in a paper presented before the Dallas, Texas, meeting of the American Phytopathological Society, December, 1941 (Phytopath. 32: 18), Walker gave additional data on physiological specialization in *P. Brassicae*. Purple Top Milan turnip remained completely free of club root when grown in heavily infested soil in Wisconsin, but when planted in naturally infected soil in England, about 20 per cent of the plants were diseased. On the other hand, an English variety, White Stone, which showed 87 per cent infection in an English test, failed to develop clubbed roots in inoculation tests with a representative American isolate of *P. Brassicae*. Walker accordingly considered this evidence as proof of the existence of physiological races.

Bacteria in Relation to P. Brassicae

The association of bacteria with *P. Brassicae* in roots of diseased crucifers was noted by Eycleshymer in 1894 and confirmed by Pinoy ('05), E. F. Smith ('11), and other early workers. From his pre-

PLATES 2, 3, 4

Plasmodiophora Brassicae

Fig. 1. Infected cabbage roots with spindle-shaped swellings. Woronin, '78.

Fig. 2. Beginning of club formation on roots of cabbage inoculated experimentally. Woronin, l.c.

Fig. 3. Heavily infected turnip root. Woronin, l.c.

Fig. 4. Cross section of infected cabbage root. Note two compact groups of infected cells on lower right side, the so-called "krankheitsherde." Woronin, l.c.

Fig. 5. Nuclear division in an enlarged infected host cell. Lutman, '13.

Fig. 6. Division of infected cell. Lutman, l.c.

Fig. 7. Enlarged cells of cabbage with mature plasmodia showing the swelling and dissolution of intervening cell walls. Rochlin, '33.

Fig. 8. Normal host nucleus. Lutman, l.c.

Fig. 9–12. Enlargement, distortion, and degeneration of host nuclei in infected cells. Lutman, l.c.

Fig. 13. Germination of resting spore. Chupp, '17.

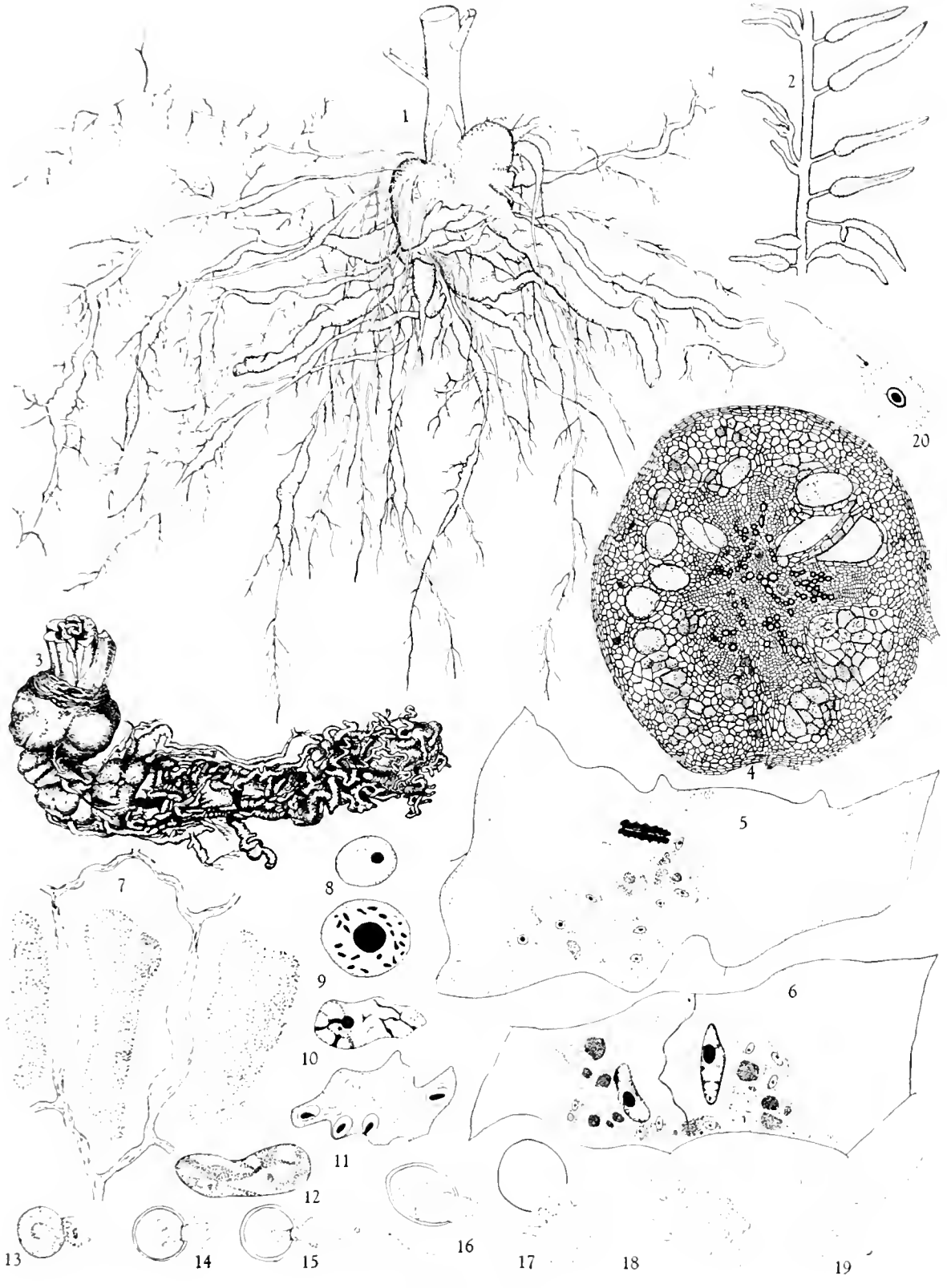
Fig. 14, 15. Same. Woronin, l.c.

Fig. 16–18. Germination of resting spore, and amoebae. Honig, '31.

Fig. 19, 21. Amoeboid, anteriorly uniflagellate zoospores. Woronin, l.c.

Fig. 20. Fixed and stained zoospore with anterior blepharoplast. Cook and Schwartz, '30.

PLATE 2



Plasmodiophora

vious studies ('02, '03) on bacteria in relation to the myxomycetes, Pinoy ('05, '07) concluded that bacteria were essential to the development of *P. Brassicae* and described the relationship between them as true symbiosis, a viewpoint which Vouk ('13) later supported. Pinoy reported that the spores of the fungus will germinate only in the presence of these bacteria, and as the zoospores or amoebae enter the host they are accompanied by cocci which continue to live in constant association with the parasite throughout its entire life cycle. Chupp ('17) repeated Pinoy's experiments to some degree and found that bacteria are absent in small, young swellings and do not appear until the galls have become quite large and old. Furthermore, instead of cocci, he found the most prevalent form to be a motile, rod-shaped bacillus which forms yellowish, opalescent colonies on nutrient media. Chupp concluded from his experiments that bacteria do not enter the host with the amoebae and that the disease must attain a certain advanced stage before the bacteria can enter. According to him, they are not essential to the development of *P. Brassicae,* but as Sorauer ('08) had previously pointed out, they may act in decomposing the host cell wall and thus liberating the spores.

Naumov ('25) likewise failed to find bacteria in young galls, while Fedotowa ('30) reported that bacteria may be present within one and a half to five months after infection. He found that *P. Brassicae* spores may be easily freed of bacteria by immersing them for five minutes in a .001 per cent corrosive sublimate solution. From diseased roots he isolated one bacillus and two coccus forms which when injected into roots in pure culture produced no signs of hypertrophy or injury. Fedotowa thus showed that bacteria are in no way necessary to spore germination, entrance of the amoebae, or to the nutrition of the plasmodium.

Plasmodiophora Brassicae and Cellular Inclusion of Cancer Cells, Small Pox, and Rabies

At the close of the last century when animal pathologists were actively engaged in trying to prove the parasitic nature of the inclusions found in carcinoma

Fig. 22, 23. Anteriorly biflagellate, heterocont zoospores. Ledingham, '34.

Fig. 24. Resting spores attached to root hair tip, small spherical myxamoeba within the cell and two myxamoeba entering through a swollen gelatinized region of the wall. Rochlin, '33.

Fig. 25. Three parasites lying in a swollen gelatinized region of the root hair wall. Rochlin, l.c.

Fig. 26. Entry of the parasite through the epidermal cell wall of the root of *B. arvensis.* Note other swollen and disorganized regions where additional parasites have entered. Rochlin, l.c.

Fig. 27. Entry of an amoeba in root hair. Honig, l.c.

Fig. 28. Swollen root hair of cabbage with a living myxamoeba. Woronin, l.c.

Fig. 29. Uninucleate myxamoeba in root hair which is locally swollen. Chupp, l.c.

Fig. 30. Division of a myxamoeba by fission. Chupp, l.c.

Fig. 31. Early stage in cell wall penetration by a young plasmodium. Kunkel, '18.

Fig. 32. Later stage. Kunkel, l.c.

Fig. 33. Young plasmodium passing through cell wall. Kunkel, l.c.

Fig. 34. Large living amoeboid plasmodium moving within host cell. Note pseudopods at the anterior and vacuoles in the posterior end. Woronin, l.c.

Fig. 35. Large saprophytic amoebae or plasmodia outside of host. Honig, l.c.

Fig. 36. Root hair filled with meronts, possibly incipient zoosporangia. Chupp, l.c.

Fig. 37. Empty and developing zoosporangia in a root hair which have been formed from a plasmodium. Cook and Schwartz, l.c.

Fig. 38. Uninucleate segment of plasmodium which will develop into a zoosporangium. Cook and Schwartz, l.c.

Fig. 39. First mitosis (meiotic?) in incipient zoosporangium. Cook and Schwartz, l.c.

Fig. 40. Binucleate stage of same. Cook and Schwartz, l.c.

Fig. 41. Zoosporangium with four incipient zoospores. Cook and Schwartz, l.c.

Fig. 42. Same with three fully formed zoospores. Cook and Schwartz, l.c.

Fig. 43. Nonflagellate zoospores from zoosporangium. Cook and Schwartz, l.c.

Fig. 44–46. Encysted myxamoeba and young plasmodia. Milovidov, '31.

Fig. 47. Large segmented plasmodium, the segments of which have encysted. Cook and Schwartz, l.c.

Fig. 48. Binucleate plasmodium with numerous chondriosomes. Milovidov, l.c.

Fig. 49. Resting nuclei of large plasmodium. Cook and Schwartz, l.c.

Fig. 50. Same in young plasmodium. Nawaschin, '99.

Fig. 51. Same in amoebae with centrosomes and astral rays. Milovidov, l.c.

Fig. 52. Early prophase of "promitosis" with chromatin in the form of numerous granules. Nawaschin, l.c.

Fig. 53. Equatorial plate stage of "promitosis" with divided nucleole. Nawaschin, l.c.

Fig. 54. Same stage. Cook and Schwartz, l.c.

Fig. 55, 56. "Double anchor" stage of "promitosis." Nawaschin, l.c.

Fig. 57, 58. Late anaphase and telophase of "promitosis." Nawaschin, l.c.

Fig. 59–61. Successive stages in development of the "akaryote" stage. Cook and Schwartz, l.c.

Fig. 62. Akaryote stage. Cook and Schwartz, l.c.

Fig. 63. Spireme stage of the first sporogonic (meiotic?) mitosis. Lutman, l.c.

Fig. 64, 65. Synapsis and possibly diakinesis, respectively. Milovidov, l.c.

Fig. 66. Early prophase of meiosis. Terby, '24.

Fig. 67. Synapsis. Terby, l.c.

Fig. 68. Strepsitene. Terby, l.c.

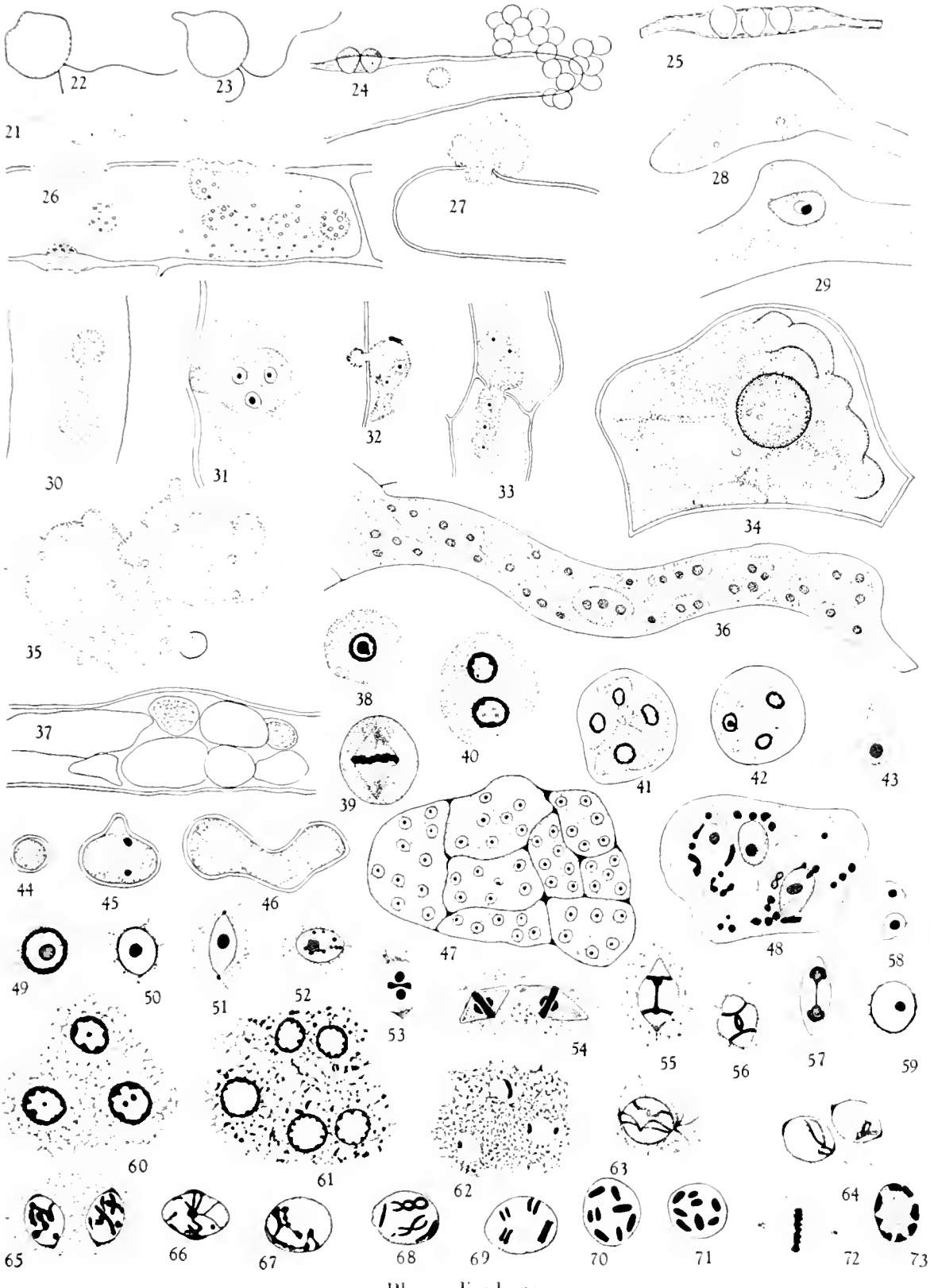
Fig. 69, 70. Diakinesis. Terby, l.c.

Fig. 71. Polar view of equatorial plate stage showing eight chromosomes. Terby, l.c.

Fig. 72. Profile view of equatorial plate stage, first division. Lutman, l.c.

Fig. 73. Polar view of same showing eight large chromatic bodies. Lutman, l.c.

PLATE 3



Plasmodiophora

cells, numerous parallelisms were drawn between cancer and club root of crucifers. The superficial resemblance of the tumors on cruciferous roots to cancerous outgrowths in animals as well as the similarity of the amoeboid stages of *P. Brassicae* to the cellular inclusions (Plimmer bodies, etc.) in cancer cells led some workers to the belief that fungi, particularly the myxomycetes and *Plasmodiophora*, may be associated with, or the cause of cancer in animals. Numerous experiments were accordingly performed in which infected cruciferous tissues were implanted in various kinds of animals. While these studies failed to throw light on the cause of cancer, they nonetheless focused attention on club root from the purely pathological viewpoint and are of considerable historical interest.

In 1898 and 1899 Bebla pointed out the similarity of club root and potato wart to cancer and discussed the possible relation of *Plasmodiophora* and *Synchytrium* to this disease in animals. After having infected animals with these fungi and noted the similarity of their developmental stages to certain inclusions in carcinoma cells, he concluded in 1903 that cancer is caused by a chytridiaceous organism. In 1900 and 1903 Podwyssotzki reported that he had succeeded in producing tumors in guinea pigs and dogs by subcutaneous and intraperitoneal implantations of infected crucifer tissues. These tumors were about the size of a walnut and resembled large-celled sarcoma, endothelioma, or granuloma. They were mesodermic in origin and had arisen through pronounced hypertrophy and repeated division of the connective tissue cells and endothelium of the perivascular fissures. Leucocyte infiltration was quite evident at first but disappeared after 8 to 12 days. Podwyssotzki found further that *P. Brassicae* produced many other changes in animal cells which were similar to those induced in cells of crucifers.

Further attempts to draw analogies between the inclusions of cancerous cells and those produced by *P. Brassicae* in animal cells were made by Feinberg ('02) and Gaylord ('04). The latter succeeded in infecting animals locally with *P. Brassicae,* and from his observations on the tumors produced he pointed out in detail the parallel cellular symptoms of club root and cancer. Gaylord concluded that cancer is caused by an amoeboid organism the developmental stages of which are very similar to *P. Brassicae*. In 1905, however, Lowenthal refuted all previous reports that the club root organism produces typical cancerous tumors in animals. He implanted infected crucifer tissues in the stomach, liver, and kidney of dogs and in the skin of white rats, but failed to get tumors or any other specific reactions in the animals. In the same year Prowazek ('05) made an extensive comparison of *P. Brassicae* and the inclusions of carcinoma cells, particularly the Plimmer bodies, and concluded that except for superficial similarities they have very little in common fundamentally. More recently Levine and Levine ('22) have made a comparison of the tumors of *P. Bras-*

Fig. 74. Simultaneous nuclear division (meiotic?) in a large, somewhat vacuolate plasmodium. Note large starch grains lying in clear regions. Nawaschin, l.c.

Fig. 75. First meiotic division with centrosome-like bodies and astral rays apart from nuclei in the cytoplasm. Terby, l.c.

Fig. 76. Second meiotic division showing four chromosomes. Terby, l.c.

Fig. 77. Second meiotic division showing centrosome-like bodies. Terby, l.c.

Fig. 78. Vacuolate stage of plasmodium prior to cleavage. Lutman, l.c.

Fig. 79. Cleavage furrow at edge of plasmodium. Lutman, l.c.

Fig. 80. Nuclear division in a large cleavage segment. Milovidov, l.c.

Fig. 81, 82, 83. Mitosis and cell division in smaller cleavage segments. Lutman, l.c.

Fig. 84. Fully formed resting spores with chromatin around inner periphery of nucleus. Cook and Schwartz, l.c.

Fig. 85. Mature resting spore with fat droplets. Lutman, l.c.

Fig. 86. Resting spores with chondriosomes. Milovidov, l.c.

Fig. 87. Variations in size and shape of resting spores. Milovidov, l.c.

Fig. 88. Fusion of incipient resting spores. Prowazek, '05.

Fig. 89. Binucleate resting spore. Prowazek, l.c.

Fig. 90. Division of one gametic nucleus. Prowazek, l.c.

Fig. 91, 92. Formation of "reduction bodies." Prowazek, l.c.

Fig. 93. Karyogamy. Prowazek, l.c.

Fig. 94. Centrosome separating from nuclear membrane to become the blephoroplast. Terby, l.c.

Fig. 95. Resting spore with blephoroplast. Terby, l.c.

Fig. 96. Binucleate resting spore. Terby, l.c.

Fig. 97. Host cell filled with resting spores. Woronin, l.c.

P. Diplantherae

Fig. 98. Infected plant of *Diplanthera wrightii* with hypertrophied bead-like internodes. Ferdinandsen and Winge, '14.

Fig. 99. Cleaving plasmodium which fills enlarged host cell and envelops host nucleus. Drawn from photograph. Ferdinandsen and Winge, l.c.

Fig. 100. Host cell filled with resting spores. Drawn from photograph. Cook, '33.

Fig. 101. Normal and collapsed resting spores. Drawn from photograph. Ferdinandsen and Winge, l.c.

P. Fici-repentis

Fig. 102. Gall on branch of *Ficus repens*. Drawn from photograph. Cook, l.c.

P. Halophilae

Fig. 103. Hypertrophied petiole of *Halophila ovalis*. Ferdinandsen and Winge, '13.

Fig. 104, 105. Normal and collapsed resting spores. Ferdinandsen and Winge, l.c.

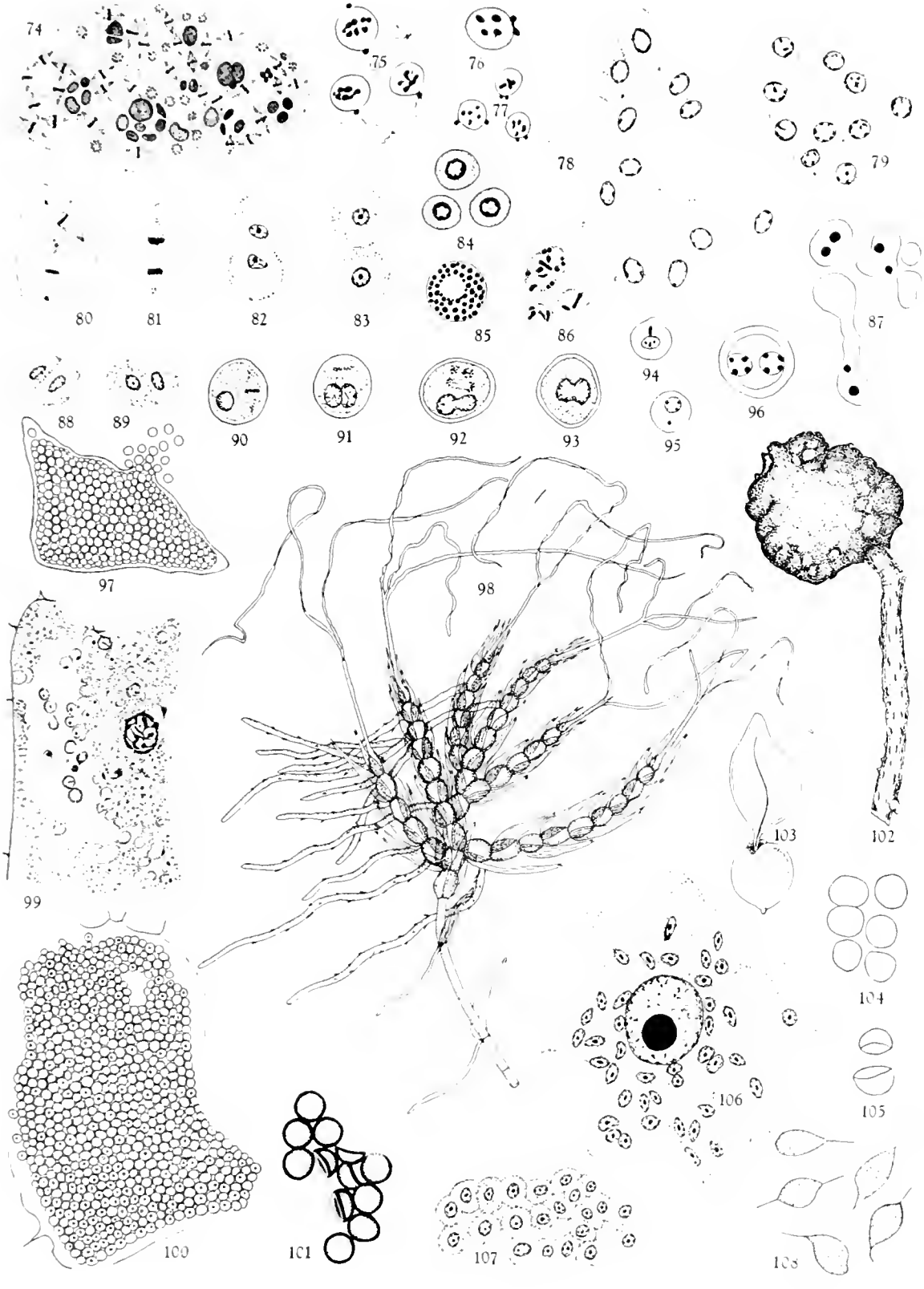
P. bicaudata

Fig. 106. Plasmodium enveloping enlarged host nucleus. Feldmann, '40.

Fig. 107. Cleavage into resting spores. Feldman, l.c.

Fig. 108. Resting spores. Feldman, l.c.

PLATE 4



Plasmodiophora

sicae on crucifers and the malignant neoplasia in animal cancer.

Analogies have also been drawn between the club root organism and the cellular inclusions formed in vaccination against small pox. Gorini ('01) succeeded in producing a slow but marked proliferation of the cornea epithelium in dogs by implanting infected cabbage tissues and found that the intracellular effects were very similar to those caused in vaccination. Certain phases of *P. Brassicae* under these conditions resembled the *Cytorcytes* bodies associated with small pox. Pollacci ('12) pointed out some of the striking resemblances between the early developmental stages of *P. Brassicae* and the Negri bodies of rabies in dogs and believed that there might be a connection between these two cellular structures.

P. DIPLANTHERAE (Ferdinandsen and Winge) Cook, 1932. Hong Kong Nat. Suppl. 1:34.
Ostenfeldiella Diplantherae Ferdinandsen and Winge, 1914. l.c., pl. 45. Fig. 1-4.

Resting spores globose, spherical, 4–4.5 μ, with fairly thick, brown, smooth walls; germination unknown. Zoospores and evanescent zoosporangia unknown. Plasmodium filling host cell, 125–200 μ in diam.; schizogony doubtful; cleaving directly into uninucleate resting spores during sporogenesis.

Parasitic in *Diplanthera wrightii*, St. Croix, West Indies, causing large galls on the stems in the region of the internodes.

This imperfectly known species was found in 1913 by Ostenfeld who turned over his material to Ferdinandsen and Winge for further study. From this scanty and poorly fixed material they created a new genus, *Ostenfeldiella*, for the species at hand. Cook subsequently examined their prepared slides and concluded that the fungus is a species of *Plasmodiophora*. Because its plasmodia cleave directly into resting spores which are not united in cystosori but lie loose and separate in the host cell as in *Plasmodiophora*, there is no reason, on the basis of present-day knowledge, for keeping this species in a separate genus. Cook's disposition of it is accordingly followed here.

Plasmodiophora Diplantherae attacks only the internodes and causes them to enlarge, so that the stem has the appearance "of a string of pearls," as is shown in figure 98. The parasite is restricted to the inner cortex where it leads to marked enlargement of the infected cells. Normal cells measure approximately 35 μ in diameter, while infected ones vary from 125 to 200 μ. In the early stages of infection the young host cells apparently retain their ability to divide, and it is not improbable that the young parasites may be dispersed by division of the host cell. The plasmodium seems to envelop the host nucleus (fig. 99), and with the start of the sporogonic phase the nucleus begins to degenerate. The pressure exerted by the enlarging cells causes tangential stretching of the outer cortical elements, and in cases of unilateral infection the central cylinder becomes laterally displaced. Since the parasite is localized in the inner cortex, infected stems can readily continue to grow and elongate.

Whether schizogony or any other division of the parasite within the host occurs in this species is uncertain. So far none has been observed. Ferdinandsen and Winge nevertheless concluded that the young amoebae divide after each mitosis, because only uninucleate stages are to be found in the meristematic areas of the stem.

P. HALOPHILAE Ferdinandsen and Winge, 1913. Centralbl. Bakt. Parasitenk. II, 37:167. Fig. a-c.

Resting spores yellowish in mass, hyaline when single, globose, 5 μ, with fairly thin smooth walls. Plasmodia one to several in a cell, variable in size and shape, subglobose, elongate, 30–60 μ long. All else unknown.

Parasitic in the petioles of *Halophila ovalis* on the island of Noesa Kembangan near the southern shore of Java, causing conspicuous pea-shaped galls.

The diagnosis of this species is based on a study of dried material which Ostenfeld found in a collection of *H. ovalis* in the Botanical Museum of Huana. He believed that the hypertrophied petioles (fig. 103) were parasitized by a species of *Plasmodiophora* but made no study of the organism. The dried material was subsequently sent to Ferdinandsen and Winge who diagnosed the parasite as a new species. It has never since been collected, nor is anything more known about its structure and development.

The species which Feldman ('36) found on petioles of *Halophila Baillonis* in Guadeloupe, West Indies, may possibly be identical or closely related to *P. Halophilae*. Feldman merely noted its occurrence without describing or identifying the parasite.

P. FICI-REPENTIS Andreucci, 1926. Arch. Bot. 2:26.

Resting spores spherical, 1.55 μ, with thin, hyaline walls; producing pyriform uniflagellate (?) zoospores in germination; flagellum 2.7 μ in length. Thin-walled evanescent zoosporangia unknown. Amoebae and young plasmodia from zoospores variable in shape and size, 6×24 μ, aggregating and fusing into larger plasmodia, which later cleave into irregular segments and finally into spores; sometimes encysting to form hyaline, globular, 9.15–73 μ cysts with granular and sculptured thick walls; cysts producing plasmodia in germination.

Parasitic on the large and small branches of *Ficus repens* in Italy, causing woody, brownish-gray, globular, irregular and coral-like tumors up to 5 cm. in diameter (fig. 102).

This species differs from *P. Brassicae* primarily by its smaller resting spores and the fact that it attacks aerial organs rather than the roots of its host. It has been recorded but once, and Andreucci unfortunately did not illustrate it. However, Cook ('33) examined dried galls, which were unsuitable for cytological study, and described this species as a doubtful member of the genus.

P. BICAUDATA Feldman, 1940. Bull. Soc. Hist. Nat. Afrique Nord 31: 173. Fig. 1, 2.

Resting spores ovoid and slightly spindle-shaped, 3–3.5 $\mu \times 7\ \mu$, thin-walled and bright yellow with a fine attenuated bristle at one or both ends; germination unknown. Plasmodia large, filling host cell completely. Zoosporangia, zoospores, and amoebae unknown.

Parasitic on *Zostera nana* in Mauretania, French West Africa, causing marked swelling and shortening of the internodes.

Feldman's study and diagnosis of *P. bicaudata* were made on material preserved in alcohol sent by Murat from Tanomdert, Mauretania, and many of the developmental stages are thus unknown. Like other members of *Plasmodophora*, this species has a marked effect on its host. Infected internodes may be two to three times the diameter of healthy ones, so that affected stalks have a characteristic monilioid appearance which is even more striking and accentuated than that of *D. wrightii* parasitized by *P. Diplantherae*. This hypertrophy appears to be due entirely to cell enlargement, since division of infected and adjacent healthy cells has not been observed. The enlarged cells of infected internodes are undifferentiated and meristematic, so that the galls produced by the parasite are typically kataplasmic.

The parasite is confined to the cortical and epidermal cells and does not infect the vascular bundles. In infected stalks, however, the latter tissue may be twice its normal diameter, due perhaps to reduction in longitudinal growth. Infected cortical cells may be two to six times their usual size and completely filled with the plasmodium or spores. They apparently do not divide after infection, although Feldman did not study the early developmental stages of the disease. He nevertheless found a few binucleate infected cells, which suggests that mitosis is not completely inhibited. The plasmodium may envelop the host nucleus (fig. 106) which is thereby stimulated to enlarge and becomes six to ten times as large as those of healthy cells. The nucleole likewise enlarges, while the chromatic material becomes more basophyllic and also vacuolate. The nucleus may persist until after the spores are mature, and in greatly enlarged cells with ripe spores it may be 35 to 40 μ in diameter, enucleolate, and looks like a partially empty vesicle containing a small amount of chromatic débris.

BIBLIOGRAPHY: PLASMODIOPHORA

Appel, O. 1928. Handb. Pflanzenkr. 2. Berlin.

———, and E. Werth. 1910. Mitt. Kgl. Biol. Anst. Land.-u. Forstw. 10: 176.

Behla, R. 1898. Centralbl. Bakt. Parasitk. 24: 829.

———. 1899. Zeitschr. Hyg. 32: 133.

———. 1903. Die Pflanzenparasitare Ursache des Krebses und die Krebsprophylaxe. Berlin.

Chupp, C. 1917. Cornell Univ. Agr. Exp. Sta. Bull. 387.

Cook, W. R. I. 1926. Trans. Brit. Mycol. Soc. 11: 196.

———. 1928a. Bull. Soc. Mycol. France 44: 105.

———. 1928b. Ann. Bot. 42: 347.

———. 1933. Arch. Protistk. 80: 179.

———, and E. J. Schwartz. 1930. Phil. Trans. Roy. Soc. Ser. B. 218: 283.

Couch, J. N., J. Leitner, and A. Whiffen. 1939. Jour. Elisha Mitchell Sci. Soc. 55: 399.

Ericksson, J. 1913. Die Pflanzenkr. Landw. Kulturpflanze. Leipzig.

Esmarch, F. 1921. Die Kranke Pflanze 4: 169.

Eyeleshymer, A. C. 1901. Jour. Mycol. 7: 79.

Favorsky, W. 1910. Mem. Soc. Nat. Kieff. 20: 119.

Fedotowa, T. 1930. Phytopath. Zeitschr. 1: 195.

Fedorintschik, N. S. 1935. Summ. Sci. Wk. Inst. Pl. Protect. Leningrad 1935: 69–70.

Feinberg, L. 1902. Ber. Deut. Bot. Ges. 19: 533.

Feldmann, J. 1936. Bull. Bot. Soc. France 83: 608.

Gaylord, 1904. Zeitschr. Krebsf. 1: 93.

Gibbs, J. G. 1931. New Zealand Jour. Agr. 42: 1, 193.

Gleisberg, W. 1923. Nachrbl. Deut. Pflanzensch'dl. 1: 10.

Gorini, D. C. 1901. Centralbl. Bakt. Parasitk. abt. I, 29: 589.

Halsted, B. D. 1893. New Jersey Agr. Exp. Sta. Rept. 1893: 332.

Henckel, A. G. 1923. Bull. Inst. Rech. Biol. Perme, fasc. 2.

Honig, F. 1931. Gartenbauwiss. 5: 116.

Hönigmann. 1926. Pflanzenbau, Halbmschr. f. Saatwesen Anbau u. Pflege d. Kulturpflanz. 2: 290.

Horne, A. S. 1911. Jour. Roy. Hort. Soc. 37: 362.

———. 1930. Ann. Bot. 44: 199.

Jones, P. M. 1928a. Arch. Protistk. 62: 313.

———. 1928b. Bot. Gaz. 81: 146.

Jorstad, I. 1923. Norsk. Havetid. 39: 126.

Kunkel, L. O. 1918. Jour. Agr. Res. 14: 513.

Larsen, R. H. 1934. *Ibid.* 49: 607.

Ledingham, G. A. 1931. Nature 133: 531. 1935, *Ibid.* 135: 3991.

———. 1939. Canadian Jour. Res. C, 17: 38.

Levine, L., and M. Levine. 1922. Jour. Cancer Res. 7: 163. *Ibid.* 7: 171.

Lowenthal, W. 1905. Zeitschr. Krebsf. 3: 16.

Lutman, B. F. 1913. Vermont Agr. Exp. Sta. Bull. 175.

Maire, R., and A. Tison. 1909. Ann. Mycol. 7: 226.

MacLeod, D. J. 1931. Rept. of Dom. Bot. for 1930. Div. Bot. Canad. Dept. Agr. 1931: 181.

Milovidov, P. F. 1931. Arch. Protistk. 73: 1.

Molliard, M. 1909. Bull. Soc. Bot. France 56: 23.

Motte, M. H. 1933. Jour. d'Agr. Prat. 97: 177. 1935, *Ibid.* 99: 93.

Naumov, N. A. 1925. Morbi Plant. 14: 19.

Nawaschin, S. 1899. Flora 86: 104.

———. 1901. Kiev. Zap. Obsch. Jest. 17: 36.

———. 1924. C. R. Acad. Sci. Russie 1924: 173.

Pape, H. 1925. Pflanzenbau 2: 172.

Pinoy, E. 1902. Bull. Soc. Mycol. France 18: 288.

———. 1903. C. R. Acad. Sci. Paris 137: 580.

———. 1905. C. R. Soc. Biol. 58: 1010.

———. 1907. Ann. Inst. Pasteur 21: 686.

Podwyssozki, W. 1900. Centralbl. Bakt. Parasitk. abt. I, 27: 97.

———. 1903. Zeitschr. klin. Med. 47: 199.

Pollacci, G. 1912. Atti Inst. Bot. Univ. Pavia. 2 ser. 15: 291.

Prowazek, S. 1905. Arb. Kais. Gesund. 22: 396.

Rochlin, E. 1933. Phytopath. Zeitschr. 5: 381.

Smith, E. F. 1911. Bacteria in relation to plant disease 2: 169.

Sorauer, P. 1908. Handb. Pflanzenkr. Berlin.

Terby, J. 1923. Mem. Roy. Acad. Belg. 7: 128.

———. 1924a. Bull. Roy. Soc. Bot. Belgique 56: 18.

———. 1924b. Bull. Roy. Acad. Belg. 5 ser. 10: 519.

———. 1932. Mem. Roy. Acad. Belg. 11: 1–20.

Van Wissenlingh, C. 1898. Jahrb. Wiss. Bot. 31: 619.
Vonwiller, P. 1918. Arch. Protistk. 38: 279.
Vouk, V. 1913. Die Naturw. 1: 81.
Walker, J. C. 1939. Jour. Agr. Res. 59: 815.
Wellman, F. L. 1930. U. S. Dept. Agr. Tech. Bull. 781.
Winge, O. 1913. Ark. Bot. 12, no. 9: 1.

EXCLUDED SPECIES

P. ALNI (Wor.) Moeller, 1885. Ber. Deut. Bot. Ges. 3: 102. Figs. 1-4. 1890. Ibid., 8: 215.

In 1866 Woronin found an organism in galls on the roots of *Alnus glutinosa* which he named *Schinzia Alni* and believed to be a hyphomycetous fungus. Gravis discussed its identity in 1879, and in 1885 Moeller placed it in *Plasmodiophora* where it was subsequently retained by Schroeter ('86, '97). Since that time its identity and relationship have been the subject of extended controversy. In 1886 Brunchorst made an intensive study of the galls of *Alnus* species and found no evidence of a plasmodium. Instead, he found a mycelioid fungus with numerous sporangia which he named *Frankia subtilis* and believed to be related to the Mucorales or Saprolegniales. This led Moeller ('90) to restudy the causal organism, after which he retracted his former view and confirmed Woronin's and Brunchorst's observations on the presence of a mycelium in the host cells. Frank confirmed these observations in 1891, but he was uncertain as to the nature of *Frankia subtilis.* While he pointed out that it might well be a form of *Leptothrix,* he was nonetheless inclined to the view that it is a mycorrhizal fungus. In 1900, according to Maire and Tison ('09, p. 242), Chodat studied the organism in question and asserted that it is a species of *Plasmodiophora.* Two years later, after an intensive study of the tubercles on *Alnus* roots, Schibata came to the conclusion that no hyphomycetous fungus was present and that the galls are caused by an organism with bacterium-like filaments which eventually become bacteroid and deformed. In 1904 Björkenheim figured and described fungus hyphae in the galls, but three years later Keissler reported the organism again as *Plasmodiophora Alni.*

Finally, in 1909, Maire and Tison undertook a cytological study of the tubercles and confirmed the observations of Shibata. They found an abundance of partially digested mycelial filaments, the ends of which became vesicular and later segmented into a large number of irregular chromatic bodies. Maire and Tison changed the name of the organism to *Frankiella Alni,* but since that time Keissler and Lohwag ('37) have reported it again as *Plasmodiophora Alni* on *Alnus* species in China.

P. ELAEAGNI Schroeter, 1889. Cohn's Krypt. Fl. Schlesiens 3: 134. 1897, Engler u. Prantl, Nat. Pflanzenf. I: 7.

Schroeter gave this name to an organism which he believed to be the cause of galls on the roots of *Elaeagnus angustifolia.* It seems that he was not aware that Brunchorst ('86) had already found the same organism on *E. angustifolia, E. argentea, E. pungens,* and *Hippophae rhamnoides* and named it *Frankia subtilis.* Maire and Tison ('09) also observed similar galls on the *H. rhamnoides,* and since they found the causal organism to be of the same type as *F. Alni,* they renamed it *Frankiella Elaeagni.* It has subsequently been reported as *Plasmodiophora Elaeagni* by Jaap ('07) and Sydow ('24) from Switzerland and New Zealand.

P. VITIS Viala and Sauvageau, 1892. C. R. Acad. Sci. Paris 114: 1558; Jour. de Bot. 6: 355, pl. 12.

This species was described by Viala and Sauvageau as causing the "brunissure" disease of grape leaves in Europe and the U. S. A. The disease had previously been observed by several workers, and in 1891 Pastre gave an account of its external symptoms. It was subsequently reported in England (Anony., '93; Cooke, '93; Massee, '93), Italy (Briosi, '94; Briosi and Cavara, '94; Cuboni, '94; Solla, '01), Germany (Moritz and Busse, '94; Behrens, '99), Algeria (Debray, '94a, '94b), Holland (Bos, '95), and France (Prilleux, '95; Roze, '99). Cooke believed that the clubbing of vine roots also was due to this organism.

The presence or absence of a causal organism in this disease as well as its identity and relationship have been the subject of much debate. In 1894 Debray pointed out that the organism was more closely related to *Ceratium* than to *Plasmodiophora,* and in the following year he established a new genus, *Pseudocommis,* to include it. In 1895 Massee reported that the plasmodia and amoebae figured by Viala and Sauvageau were nothing but vacuolated tannin vesicles and the reticulate primordial utricle of the host cell. Behrens likewise questioned the presence of a causal organism in this disease, but in the same year Roze reported that *P. Vitis* occurs widely and is almost a universal parasite. Ducomet ('03, '07) confirmed Massee's view that "brunissure" is the result of certain environmental and physiological conditions. Maire and Tison ('09) also reported that no organism was present in the diseased tissues which they examined, and they thus concluded that the so-called plasmodia were products of cell degeneration.

P. CALIFORNICAE Viala and Sauvageau, 1892. C. R. Acad. Sci. Paris 115: 67-69.

Viala and Sauvageau believed this species to be the cause of a vine disease in California, and it was subsequently reported as such by Casali and Ferraris ('00) in Italy. Massee ('95) pointed out that the disease is physiological and that the reported amoebae and plasmodia are nothing more than tannin vesicles and reticulated host protoplasm. Ravaz

('06) also reported that *P. Californiae* is not an organism but only degenerated chlorophyll, and in 1909 Maire and Tison confirmed the findings of both of these investigators.

P. ORCHIDIS Massee, 1895. Ann. Bot. 9: 170, 421-429, pl. 15.

Under this name Massee reported an organism which he believed to be the cause of spot diseases of orchid leaves. Later in the same year after more intensive study he retracted this view and showed that what he had previously believed to be spores were nothing more than tannin vesicles.

P. TOMATI Abbey, 1895. Jour. Hort. 3rd ser. 30: 369.

Abbey stated in a letter to the Journal of Horticulture that this organism is probably the cause of a disease of tomatoes, but as far as the present writer is aware he never gave a description of the parasite. Massee ('95, p. 427) believed that Abbey's disease is not due to a parasitic organism but to certain rapid changes in environmental conditions.

P. HUMULI Kirk, 1906. Rept. Dept. Agric. New Zealand 11: 337.

On the roots of hops in New Zealand Kirk found galls which were similar to those produced by *P. Brassicae* on crucifers, and without seeing the causative agent, he assumed the disease to be caused by a species of *Plasmodiophora* to which he gave the name *P. humuli*. In 1922 Nicholls reported a disease of hops in Tasmania which showed the "take all" symptoms, and he assumed it to be caused by the same organism without examining the tissues microscopically. Later, in his correspondence with Miss McLennan ('31, p. 12) he stated that he had found a myxomycete which he took to be *P. humuli*. In studying the "take all" disease of hops more intensively in Tasmania, McLennan concluded that it may be caused by a virus. In some of the diseased plants, however, she found a plasmodial organism which was later isolated and grown in pure culture and turned out to be a proteomyxean species, *Leptomyxa reticulata* var. *humuli*. She also examined preserved material of diseased hops labelled *P. humuli* which belonged to the Department of Agriculture, Melbourne, but found no evidence of a myxomycete. Miss McLennan accordingly concluded that the tumors described by Kirk were crown galls caused by *Pseudomonas tumefaciens* and that *P. humuli* is no longer valid.

P. VASCULARUM Matz, 1920. Jour. Dept. Agric. Porto Rico 4: 45, figs. 7-9.

Ligniera vascularum M. T. Cook, 1929. *Ibid.* 13: 19. Pls. 3-6.

Sorosphaera vascularum M. T. Cook, 1937. *Ibid.* 21: 81. Pls. 5-7.

This species was described by Matz ('20, '21, '22) as causing the dry top rot of sugar cane, *Saccharum officinari*, in Porto Rico, and in 1934 it was reported on the same host in Venezuela by Chardon and Toro. M. T. Cook transferred the organism to the genus *Ligniera* because it does not cause hypertrophy of the host. Later ('32), W. R. I. Cook made an intensive study of the organism from material sent by M. T. Cook and found that the disease was caused by two protozoan parasites to which he gave the names *Amoebosporus vascularum* and *A. Saccharinum*. M. T. Cook later ('37) transferred it to the genus *Sorosphaera*.

P. TABACI Jones, 1926. Bot. Gaz. 81: 416, pls. 34-37. Fig. 1, 2.

P. tabacum Jones, 1926. Phytopath. 16: 67.

In tobacco, potato and tomato plants affected with mosaic-like symptoms and leaf roll, Jones found a plasmodiaceous organism which he believed to be a species of *Plasmodiophora*. Infected cells become necrotic and adjacent ones hyperplastic, and all tissues except bast fibers and xylem are invaded by plasmodia which pass from cell. Jones found only plasmodia in the host plants, but when these are cultured in Knop's solution, they give rise to amoebae and uniflagellate organisms, both of which may or may not encyst. The amoebae which continue to develop discharge chromidia from the nucleus into the cytoplasm, and these chromidial bodies soon aggregate to form daughter nuclei, thereby making the enlarged amoebae multinucleate. Such amoebae give rise to motile uniflagellate isogametes which fuse shortly, forming a uniflagellate zygote. The zygote then divides into two zoospores which in turn form amoebae.

The flagellate cells and amoebae which encyst produce amoebae on germination, and these fuse to form the plasmodium. According to Jones, hundreds of amoebae may flow together in this manner and make a huge plasmodium which creeps along rapidly like a giant amoeba ingesting food in its path. Sporogenesis in this species is unlike that of any known member of the Plasmodiophoraceae. As the plasmodium moves along, oval and spherical spores are delimited in rows and left behind. The nuclei of these spores soon enlarge, discharge chromidia, and eventually disappear, while the chromidia in the cytoplasm aggregate and form daughter nuclei. Walls develop around these nuclei, and in this fashion 3 to 15 endogenous spores are formed. *Plasmodiophora tabaci* has a very complex life history, according to Jones, but he was not certain as to the sequence of stages. Since he also found certain flagellate and amoeboid stages which could not be fitted into any known life cycle, it is not improbable that he may have had more than one organism at hand. Miss McLennan ('31) believed that the plasmodial stage may relate to a proteomyxean-like organism of the *Leptomyxa reticulata* type. In 1931 Cook expressed a similar opinion in stating that *P. tabaci* is probably a species of amoeba which had temporarily entered the tobacco leaf, but in 1933 he suggested that

the stages which Jones found in mosaic diseased plants may relate to excitation and degeneration products of the kind described by Kunkel, Goldstein, Holmes, Sheffield, and others.[1] At any rate *P. tabaci* has but little in common with other known members of genus, and the author agrees with Cook that there is little if any justification at present for including it in the Plasmodiophoraceae.

Since he was able to produce mosaic-like symptoms in plants by inoculation with cultures, Jones believed his organism relates to the cause of tobacco mosaic. Later in the same year, however, he, Link, and Taliaferro ('26) found that the organism could be cultivated from healthy as well as diseased plants. Furthermore, upon inoculation, mosaic-like symptoms appeared only when the amoebae and plasmodia came from diseased plants. They accordingly concluded that *P. tabaci* is not the cause of mosaic but may be a carrier of the causative agent.

In 1937 Jones retracted his previous views about *P. tabaci* and redescribed it as the soil amoeba, *Naegleria gruberi,* which he claimed is not an amoeba proper but a stage in the life cycle of a myxomycete. He excluded it from the Plasmodiophorales on the grounds that: (1) several amoebae fuse and form a large multinucleate plasmodium; (2) the nuclei divide promitotically as in an amoeba; (3) the plasmodium forms an aggregate of separate resting spores; and (4) it does not parasitize plants. He furthermore reported that *N. gruberi* may be an alternate host for the mosaic producing organism in tobacco. Jones' above-cited reasons for excluding this organism from *Plasmodiophora* are obviously no more critical than those presented previously for including it in this genus. The additional data which he has presented do not clarify its taxonomic position or relationship.

P. SOLANI Brehmer and Bärner, 1930. Arb. Biol. Reichsanst. f. Land-u. Forstwirtsch. 18: 1-54, pl. 1, fig. 1-32.

Brehmer and Bärner gave this name to an oval, 4.35–5.5 $\mu \times$ 2.9 μ, amorphous, pale yellowish-green organism with a distinct refringent sheath which they found in older portions and parenchymatous tissues of potato stems showing leaf roll, mosaic, and other degenerative symptoms. The thallus divides into as many as eight daughter cells, and these in turn give rise to vesicles or spores which are subsequently liberated by the breakdown of the daughter cells. The spores produce a filamentous zoospore, 5 μ in length and a fraction of a micron in diameter. Brehmer and Bärner found all of these stages in filtered juices of diseased plants as well as in plants to which virus symptoms had been communicated by grafting and concluded therefrom that *P. Solani* is the cause of potato virus. These authors pointed out the similarities of their organism to Jones' parasite *Calkinsi* and the so-called "X" bodies of various investigators and considered it to be either an independent amoeboid entity capable of spore formation or a plasmodium living in symbiotic relationship with the plastids. They regarded it as a member of a hitherto unknown group of the Archimycetes allied to the Plasmodiophoraceae. It has subsequently been reported by Moesz ('38) on potatoes in Hungary.

There is little in the life cycle of this organism, as described by Brehmer and Bärner, which indicates relationship to *Plasmodiophora,* and it is accordingly excluded from the genus.

[1] Kunkel, L. O. 1921. Hawaiian Sugar Planters' Assoc. Bot. Ser. 3: 108.
Goldstein, B. 1927. Bull. Torrey Bot. Club 54: 285.
Holmes, F. O. 1928. Bot. Gaz. 86: 50.
Sheffield, F. M. L. 1931. Ann. Appl. Biol. 18: 471.

P. THEAE Fitzpatrick, 1930. The lower fungi-Phycomycetes. New York.

See *Sorosphaera Theae* Spesehnew.

BIBLIOGRAPHY: EXCLUDED SPECIES

Anony. 1893. Gard. Chron. 74: 217.
Behrens, J. 1899. Weinb. u. Weinh. no. 33.
Björkenheim, C. G. 1904. Zeitsch. Pflanzenkr. 14: 129.
Bos, R. 1895. Zeitschr. Pflanzenkr. 5: 286.
Briosi, G. 1894. Boll. Notiz. Agr. 16: 522.
———, and F. Cavara. 1894. Essiccati, delineati e descritti. Fasc. X.
Brunehorst. 1886. Untersuch. Bot. Inst. Tübingen 2: 151.
Casali, C., and T. Ferraris. 1900. Giorn. Vitic. Entolog. 8: 10.
Chardon, C. E., and R. A. Toro. 1934. Monogr. Univ. Porto Rico, Phys. Biol. Sci. ser. B, no. 2: 75.
Cook, M. T. 1929a. Phytopath. 19: 91.
———. 1929b. Ann. Rept. Insular Exp. Sta. Dept. Agr. Labor. Porto Rico 1927/'28. 1929: 59.
Cook, W. R. I. 1931. Ann. Protist. 3: 197.
———. 1932: Jour. Dept. Agr. Porto Rico 16: 407.
Cooke, M. C. 1893. Gard. Chron. 13: 711.
Cuboni, G. 1894. Boll. Notiz. Agr. 16: 378.
Debray, F. 1894a. C. R. Acad. Sci. Paris 119: 110.
———. 1894b. Rev. Viticult. nos. 35, 38.
———. 1895. C. R. Acad. Sci. Paris 120: 943.
Duconet, V. 1903. C. R. Assn. Franc. avanc. Sci. 32 sess. Angiers 2nd pt. pp. 697–707.
———. 1907. Ann. Fecole d'Agric. Rennes 1: 1–284. 1908, *Ibid.* 3: 1–70.
Frank, B. 1891. Ber. Deut. Bot. Gesell. 9: 244.
Gravis, A. 1879. Bull. Roy. Soc. Bot. Belgique 18: 50.
Jaap, O. 1907. Ann. Mycol. 5: 246.
Jones, P. M. 1928. Arch. Protistk. 62: 307.
———. 1937. Amer. Nat. 71: 488.
Keissler, K. 1907. Ann. Mycol. 5: 220.
———, and H. Lohwag. 1937. Fungi Symb. Sinicae 2: 2.
Link, G. K., P. M. Jones, and W. H. Taliaferro. 1926. Bot. Gaz. 81: 403.
Maire, R., and A. Tison. 1909. Ann. Mycol. 7: 242.
Massee, G. 1893. Gard. Chron. 14: 282.
Matz, J. 1921. Esta. Exp. Insular Porto Rico 8: 63.
———. 1922. Jour. Dept. Agr. Porto Rico 6: 28.
McLennan, E. I. 1931. Australian Jour. Exp. Biol. Med. Sci. 8: 9.
Moesz, G. 1938. Ann. Hist. Nat. Mus. Hungary 31: 58.
Moritz, J., and W. Busse. 1894. Zeitschr. Pflanzenkr. 4: 257.
Nicholls, H. M. 1922. Ann. Rept. Govt. Mierobiol. Dept. Agr. Stock, Tasmania 1920–21 and 1921–22.
Prillieux, E. 1895. Maladie des Plantes Agric. 1: 17.
Ravaz. 1906. Rapp. Caise Rech. Paris 1906: 173.

Roze, M. E. 1899. Bull. Soc. Mycol. France 15: 37.
Shibata, K. 1902. Jahrb. wiss. Bot. 37: 643.
Speschnew, N. N. 1907. Die Pilzparasiten des Teestrauches. Berlin.
Solla. 1901. Zeitschr. Pflanzenkr. 11: 228.
Sydow, H. 1924. Ann. Mycol. 22: 293.
Woronin, M. 1866. Mem. Acad. Sci. St. Petersburg 7 ser. 10, no. 6: 13.

TETRAMYXA

Goebel, 1884, Flora 67: 517

(PLATE 5, FIG. 1–26)

Molliardia Maire and Tison, 1911, Ann. Mycol. 9: 236.

Resting spores usually in tetrads but often separating and lying singly, or in diads and triads; variously shaped, giving rise to a single nonflagellate (?) and amoeboid cell in germination. Plasmodia usually small, becoming parietal in the host cell at maturity and cleaving into uninucleate spore-mother cells or sporonts which usually divide twice to form tetrads of resting spores. Zoosporangia and zoospores unknown.

Tetramyxa is the second plasmodiophoraceous genus to be recognized as such, and although it has been known for many years, our knowledge of its critical developmental stages is meager. It includes at present two species and possibly a third one, which is so imperfectly known that its inclusion in *Tetramyxa* is very problematical. While this genus appears to be comparatively rare in occurence, it is widely distributed and has been reported from Finland, Germany, Great Britain, France, Morocco, Japan, and California, U. S. A. Further studies may show it to be world wide in distribution.

No zoosporangia or zoospores have been observed in *Tetramyxa*, and the resting spores are reported to give rise to amoeboid, nonflagellate cells. More careful and intensive studies under optimum developmental condition, however, may show that these cells are biflagellate and heterocont and that zoosporangia also are developed.

T. PARASITICA Goebel, l.c. Pl. 10. Hissinger, 1888. Meddel. Soc. Fauna et Flora Fenn. 14: 53. Pl. 1–10. Maire and Tison, 1911, Ann. Mycol. 9: 228. Pl. 10.
Thecaphora Ruppiae Setchell, 1924. Mycologia 16: 243. Pl. 18. Fig. 3, 4.

Resting spores spherical, 3.5–7 μ, and angular, with smooth hyaline walls, giving rise to nonflagellate (?) amoeboid cells upon germination. Plasmodia usually several in a cell, small, 15–30 μ, or almost filling host cell.

Parasitic in the stalks of *Ruppia rostellata, Zannichellia polycarpa,* and *Z. palustris* in Finland and Germany (Hissinger and Goebels, l.c.); *Ruppia* sp. and *Z. palustris* in Great Britain (Boyd, '97; Schwartz, '11; Haddon, '22); *R. rostellata* in France (Maire and Tison, '10, '11), *Potamogeton panormitanus* in Morocco (Maire, '17; Maire and Werner, '37), and *Ruppia maritima* var. *rostrata* in California, U. S. A. (Setchell, l.c.), causing small or large, up to 15 mm. in diam., greenish and later whitish brown galls on the stalks, peduncles, and margins of the leaves.

The galls (fig. 1) are primarily due to increased cell multiplication. The infected and adjacent cells do not increase much in size, but are stimulated by the parasite to divide rapidly. This is particularly true of the infected cells, and by such means the meronts or portions of the plasmodium are passively distributed to the respective daughter cells (fig. 4). This appears to be the primary means of dispersal of the parasite within the host tissue, although Cook believed that the amoebae are capable of migrating from cell to cell. The relation between host and pathogen is very intimate, according to Maire and Tison, and no antagonism is exhibited. The cytoplasm of both often appears to be confluent, and it is frequently impossible to determine the boundaries between them. Although the nuclei of the host cells may be enveloped by the plasmodium (fig. 2), they do not become enlarged and deformed or divide amitotically as in *Triglochin palustre* parasitized by *T. Triglochinis*. When young, the infected cells contain a fairly large amount of starch, but this usually disappears after the sporulation of the parasite. The nucleus remains intact for some time later, but eventually degenerates.

Cook ('33) reported that this species had been collected by Boyd and Haddon in Scotland and England and that a diseased specimen of *R. rostellata* in the Father Reader Herbarium, University of Bristol, had been collected as early as 1885 in Hampshire. Maire and Tison ('11) found *T. parasitica* in abundance on *R. rostellata,* which grew in close association with *Z. palustris* var. *pedicellata.* The latter host was not infected. Maire and Tison accordingly expressed doubt about Hissinger's report of the parasite's occurrence on *L. polycarpa*, because many authors regard this species as only a form of *Z. palustris.*

T. TRIGLOCHINIS Molliard. 1909. Bull. Soc. Bot. France 56: 24.
Molliardia Triglochinis (Moll.) Maire and Tison, 1911, Ann. Mycol. 9: 236. Pl. 12, fig. 47–63; Pl. 13, fig. 65–67.

Resting spores, zoospores, and zoosporangia unknown. Plasmodia small, usually numerous in a host cell; undergoing multiple division into several oval, elongate, sickle-shaped uninucleate meronts which grow in size, and during the two- to eight-nucleate stage function in turn as schizonts. All else unknown.

Parasite on the stems, flowering stalks, stamens, ovaries, but rarely on the leaves of *Triglochin palustra* and *T. maritimum* in France (Molliard, Maire and Tison, l.c.) and *T. maritimum* in England (Cook, '33), causing small fusiform, oval and irregular galls.

No resting spores have been observed in this species, so that its relationship to the other members of the Plasmodiophoraceae is obscure. Because of the lack of resting spores, Maire and Tison regarded

it as representative of a new genus, but as Cook pointed out, there are no good reasons for introducing a new genus until more is known about the life history of this species. It is accordingly retained provisionally in *Tetramyxa*. Maire and Tison's cytological study, nonetheless, indicates its similarity to *T. parasitica* in the type of vegetative nuclear division and the presence of centrosomes and astral rays.

The effects of this species on the host are striking. According to Molliard, the parenchyma cells of the stem and flowering stalk are greatly hypertrophied and divide irregularly, while development of the sclerenchyma is inhibited Likewise the flowers in an infected region are sterilized. The infected cells may enlarge to four times their normal diameter, while their nuclei become enormous and deformed (fig. 20). The nucleoli also increase markedly in size and become deeply basophyllic. At the same time numerous deep-staining, chromosome-like chromatic bodies develop in the nuclear cavity. Furthermore, infected cells may often become multinucleate (fig. 19) as the result of amitosis, according to Maire and Tison. The nuclei of adjacent cells may also become enlarged and deformed. The presence of the parasite further stimulates starch formation in the cells surrounding infected regions.

T. ELAEAGNI Yendo and Takase, 1932. Bull. Sericult. Silk—Ind. Japan 4, no. 3: 5.

Plasmodium inter- and intracellular, segmenting into uninucleate spore mother cells which divide twice to form tetrads of resting spores. Amoebae formed from germinating resting spores. Sporangia and zoospores unknown.

In the roots of *Elaeagnus multiflora* in Japan.

This species causes tubercles or nodules which in exceptional cases may attain the size of a man's fist on old trees. The parasite occurs most abundantly in the cortex and causes marked hypertrophy of the infected cells as well as enlargement and distortion of the nuclei. Yendo and Takase found that the percentage of nitrogen in the nodules was almost twice that of the normal cortex, and for this reason they believed that there is a definite symbiotic relationship between host and parasite.

So little is known about this species that its validity as a member of the Plasmodiophoraceae is very doubtful. Yendo and Takase reported that the plasmodium spreads over the host cells and fills the intercellular spaces. Furthermore, the resting spores are said to be capable of forming fine, curled, nonseptate, branched germ tubes or filaments instead of amoebae. The formation of germ tubes suggests that Yendo and Takase may have had spores of another fungus at hand.

ADDITIONAL BIBLIOGRAPHY: TETRAMYXA

Cook, W. R. I. 1932. Hong Kong Nat. Suppl. no. 1: 38.
———. 1933. Arch. Protistk. 80: 216.
Maire, R., and A. Tison. 1910. C. R. Acad. Sci. Paris 50: 1768.
Schwartz, E. J. 1911. Ann. Bot. 25: 794.
Winge, O. 1913. Ark. f. Bot. 12, no. 9: 26.

PLATE 5

Tetramyxa parasitica

Fig. 1. Galls on stems of *Ruppia rostellata*. Goebel, l.c.; Maire and Tison, '11.

Fig. 2. Multinucleate plasmodium surrounding host nucleus. Maire and Tison, l.c.

Fig. 3. Plasmodium consisting of two multinucleate meronts which appear to be fusing; nuclei dividing in one and at rest in the other. Maire and Tison, l.c.

Fig. 4. Division of infected cell by which the meronts have been passively divided and distributed. Maire and Tison, l.c.

Fig. 5. Equatorial ring stage of "promitosis" in which distinct chromosomes are evident. Maire and Tison, l.c.

Fig. 6. Anaphases of same. Maire and Tison, l.c.

Fig. 7. Plasmodium becoming parietal and cleaving into uninucleate spore-mother cells or sporonts. Centrosomes and astral rays present at poles of some nuclei. Maire and Tison, l.c.

Fig. 8, 9. Prophases of meiosis in sporonts. Maire and Tison, l.c.

Fig. 10. Equatorial plate stage of first meiotic division. Maire and Tison, l.c.

Fig. 11. Binucleate sporont with conspicuous astral rays. Maire and Tison, l.c.

Fig. 12. Equatorial plate stage of second meiotic division. Maire and Tison, l.c.

Fig. 13. Cleavage into tetrads.

Fig. 14. Tetrad of resting spores.

Fig. 15. Enlarged host cell with resting spores isolated and single, in linear series, in diads, triads and tetrads. Large resting spores binucleate. Maire and Tison, l.c.

Fig. 16. Four resting spores in linear series. Goebel, l.c.

Tetramyxa Triglochinis

Fig. 17. Galls on *Triglochin palustre* caused by *T. Triglochinis*. Maire and Tison, l.c.

Fig. 18. Enlarged host cell with spherical multinucleate and fusiform uninucleate meronts. Maire and Tison, l.c.

Fig. 19. Enlarged host cell with uninucleate meronts in vacuoles. Host cell tetranucleate; nuclei with numerous densely chromatic bodies. Maire and Tison, l.c.

Fig. 20. An enlarged, deformed host nucleus. Maire and Tison, l.c.

Fig. 21. Uninucleate fusiform meronts. Maire and Tison, l.c.

Fig. 22–24. Equatorial plate, anaphase and telophase stages of "promitosis."

Fig. 25, 26. Bi- and multinucleate thalli. Maire and Tison, l.c.

Octomyxa Achlyae

Fig. 27. Habit sketch of *Achlya glomerata* showing effects of parasite on the hyphae. Couch, *et al.*, '39.

Fig. 28. Early infection stage showing large parasite nucleus in host cell.

Fig. 29. Binucleate thallus surrounded by host protoplasm; nuclei dividing "promitotically."

Fig. 30. Large plasmodium in a vacuolate area of hyphal tip.

Fig. 31. Sporangiosorus of nearly mature zoosporangia.

Fig. 32. Zoosporangia with emerging zoospores.

Fig. 33, 34. Biflagellate heterocont zoospores.

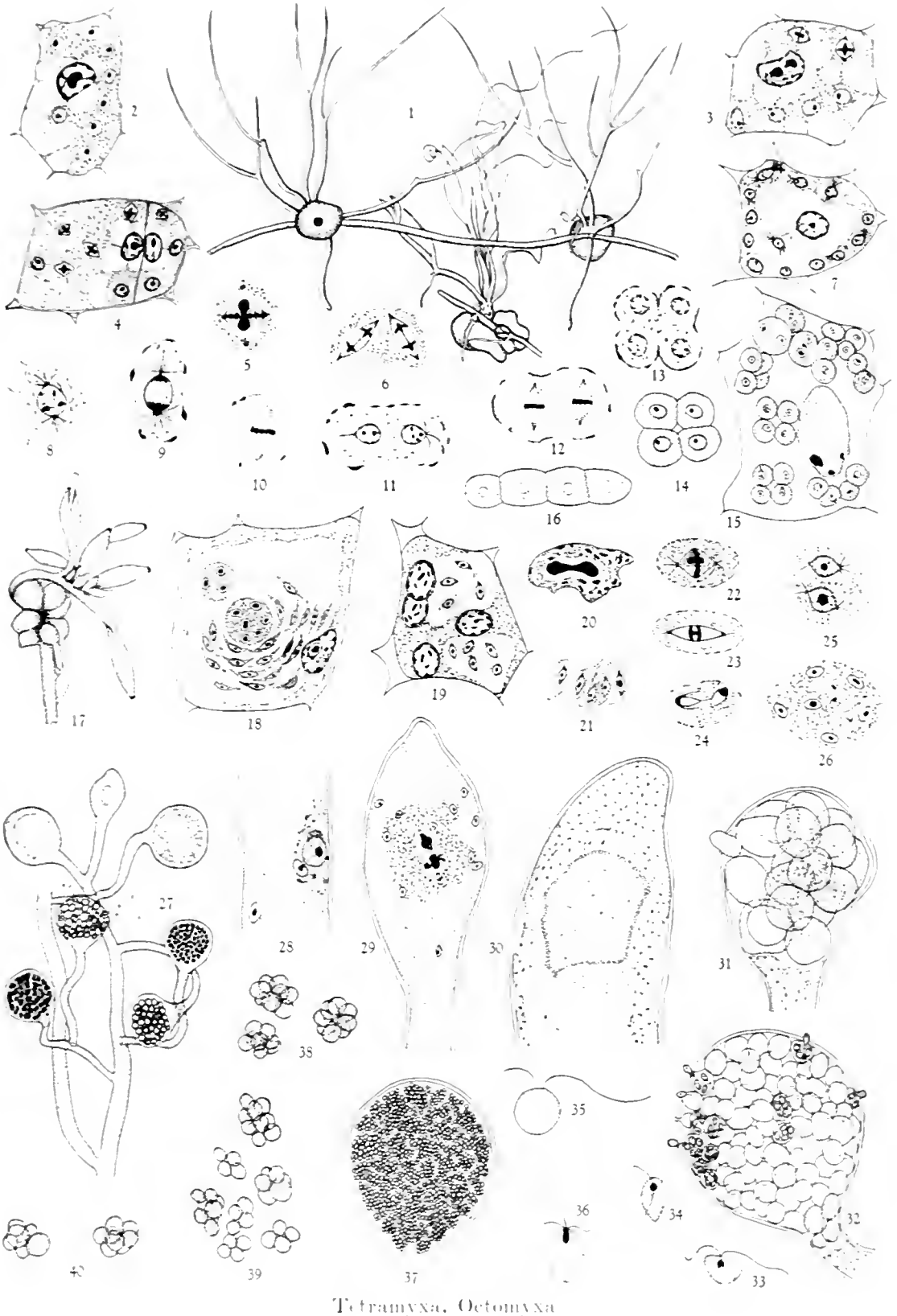
Fig. 35. Zoospore killed in osmic acid fumes and stained with gentian violet. Drawn from photomicrograph.

Fig. 36. Large tetraflagellate zoospore.

Fig. 37. Sorus of resting spores.

Fig. 38–40. Groups of resting spores.

PLATE 5



Tetramyxa, Octomyxa

OCTOMYXA

Couch, Leitner, and Whiffen, 1939. Jour. Elisha Mitchell Sci. Soc. 55: 400. Whiffen, 1939. *Ibid.* p. 243.

(PLATE 5, FIG. 27–40)

Resting spores usually adhering in groups of eight, sometimes in groups of six to nine; forming zoospores which infect the host and develop into vegetative plasmodia. Such plasmodia cleaving into sporangiosori composed of numerous small zoosporangia, which are sometimes conjoined by narrow isthmuses; exit papillae lacking on some zoosporangia. Zoospores anteriorly biflagellate and heterocont. Sporogenous plasmodia cleaving into small segments which in turn divide into eight uninucleate spores.

This monotypic genus is characterized by resting spores which are grouped usually in clusters of eight (fig. 38). As in other genera, the zoospores enter the host hyphae directly and completely without leaving a spore case on the outside. Infection may occur at any place along the hyphae, but hypertrophy of the host occurs only at or near the tip (fig. 27). The young, naked parasite is surrounded by the host protoplasm (fig. 28–29) and soon develops into a multinucleate plasmodium. As the latter develops, the hyphal tip swells and attains its maximum size before the parasite is completely mature. As a result, the plasmodium lies in a vacuolate region (fig. 30) of the swelling, surrounded by radiating strands of host protoplasm along which small particles may be seen moving toward the parasite. The latter thus lives within and in intimate contact with the host protoplasm, and in the early stages of development the two protoplasts are indistinguishable. The plasmodium usually develops from a single zoospore, but Couch *et al.* believed several small plasmodia may fuse to form a large one.

The mature plasmodium, however formed, may give rise to sporangesori or cytosori, but the latter do not usually appear until the cultures are several days old. The zoosporangia (fig. 31) are delimited as globose or ovoid masses which soon develop thin, hyaline walls. Sometimes cleavage may be incomplete, so that several sporangia are joined by narrow isthmuses. As the sporangia mature, exit papillae are formed on those adjacent to the host wall and on some in the center of the group or sorus. As a result, the zoospores may be discharged (fig. 32) directly to the outside or within the host cell. They emerge from the zoosporangia singly and slowly, and after moving about sluggishly for a few seconds at the mouth of the exit papillae swim away. The two unequal flagella are attached at or near the anterior end, and during motility the shorter one extends forward while the longer projects backward. Occasional zoospores with four flagella occur (fig. 36), which appear to be the result of incomplete or unequal cleavage instead of fusion.

The plasmodia which give rise to the resting spores are indistinguishable from the zoosporangial plasmodia until after cleavage begins. Miss Whiffen ('39) reported that the two are to be distinguished cytologically by the fact that the nuclei of the resting spore plasmodia pass through the so-called akaryote stage and undergo reduction division. However, she has not yet counted the chromosomes present during the two meiotic divisions. The sporogenous plasmodia cleave into a number of comparatively large masses, as in *Tetramyxa,* and these in turn usually divide into eight uninucleate segments which soon encyst in groups of two tetrads of resting spores. This grouping, however, may frequently vary from six to nine. Four normal-sized spores and two larger ones may sometimes occur, while nine and seven may be found in other groups. After a short dormant period, the resting spores germinate, each one giving rise to a single zoospore. The structure, type of flagella, and method of swimming of these zoospores are unknown.

O. ACHLYAE Couch, *et al.*, l.c., Pl. 47, 48.

Resting spores spherical, 2.4–3.2 μ, with smooth, slightly thickened walls. Zoosporangia spherical, ovoid, sometimes flattened by mutual pressure, 6–16 μ in diameter, hyaline and thin-walled; single exit papilla on sporangia adjacent to host wall and in the center of gall; deeper lying sporangia often discharging zoospores through the peripheral sporangia. Zoospores 6–14 in a sporangium, discharged directly to the outside and also within the host wall; oval; flagella attached to or near the anterior end, the shorter one extending forward and the larger one backward during swimming.

Parasitic in *Achlya glomerata* in North Carolina, U. S. A., causing marked enlargement of the hyphal tips.

This species appears to be an obligate parasite of *A. glomerata.* Couch, *et al.,* attempted to transfer it to *Saprolegnia ferax, S. megasperma, A. imperfecta, A. flagellata, A. colorata, A. racemosa, A. deBaryana, Aphanomyces stellatus, Apodachlya brachynema, A. minima,* and *Allomyces arbuscula,* but all results were negative. So far, this is the only known species of the Plasmodiophoraceae parasitic in a fungus.

The life cycle of *O. Achlyae* seems to be almost identical with that of *Woronina polycystis* as far as both species are known at present, and it is not improbable that the two may prove to be related. According to Couch, *et al., O. Achylae* differs from *W. polycystis* by the fact that it usually causes spherical swellings and does not lead to septation of the host hyphae. Furthermore, its cystosori are hyaline instead of brown, and the resting spores are usually grouped in clusters of eight rather than in spherical, oval, elongate, and irregular masses. The first difference cited above is not very significant, since the shape of the swellings is not a very fundamental diagnostic character. What seems more significant is that the sporangia and resting spores of *W. polycystis* give a definite cellulose reaction, while those of *O. Achylae* do not.

SOROSPHAERA

Schroeter, J. 1866. Cohn's Krypt. Fl. von Schlesien 3: 135.

(PLATE 6)

Cystosori one to several in a cell, predominantly of the shape of hollow spheres or ellipsoids, but often extremely variable in size and shape; presence of common enveloping membrane doubtful. Resting spores oval, ellipsoidal, pyriform, pyramidal and urn-shaped with yellowish-brown to brown, thin, smooth or verrucose walls; with or without apical collar; producing a single biflagellate, heterocont zoospore in germination. Evanescent thin-walled zoosporangia small. Plasmodia one to several in a cell, large or small; schizogony present (?) or lacking; producing a single cystosorus.

This genus includes at present only two species which have been reported in moist and damp localities in Europe, England, and the U. S. A. Of these two, *S. Veronicae* appears to be more common and has been frequently studied cytologically. Nonetheless, many of its critical developmental stages are still imperfectly known, and there has been considerable controversy relative to many of its cytologieal details. Germination of the resting spores had not been observed until very recently. Blomfield and Schwartz ('10) reported the presence of amoebae in a sterile infusion of *Veronica* leaves which had been inoculated with portions of dead tumors. Since this infusion was thus no longer sterile and soon became invaded with bacteria, molds and other organisms from the tumors, the uninucleate amoebae which they found after fourteen days in the bottom of the test tube may not relate to *S. Veronicae* at all. In *S. radicalis*, Cook and Schwartz likewise failed to observe germination, but among diseased root hairs they found anteriorly uniflagellate zoospores which they assumed relate to their species. However, they did not follow the development of these zoospores into mature thalli. On the other hand, Barrett found that the zoospores from sporangia are distinctly biflagellate and heterocont. He also succeeded in germinating the resting spores, but has not yet determined the number of flagella on such zoospores. Ledingham ('39, p. 43) found that zoospores from resting spores of *S. Veronicae* also are biflagellate and heterocont. Cook ('33) stated that the resting spores form a single amoeba or zoospore, but it is quite probable that the multinucleate spores reported by Maire and Tison (fig. 49–51) may give rise to several zoospores.

As the primary uninucleate amoebae of *S. Veronica* (fig. 9) increase in size within the host cell, their nucleus divides, and multinucleate plasmodia are soon formed (fig. 11–15, 22). By the time the eight-nucleated stage has been reached, the plasmodia may function as schizonts and split off uni- and multinucleate meronts (fig. 23, 24), according to Maire and Tison. The multinucleate meronts may in turn undergo schizogony into uninucleate segments before further mitoses occur. The uninucleate meronts are equivalent to the primary amoebae and may thus begin the cycle anew, while the schizont from which they are derived passes into the sporogonic phase of development, in the opinion of Maire and Tison.

It is to be noted, however, that these workers have never observed the actual splitting off of meronts, and their reports on the presence of schizogony are based only on the appearance of constricted plasmodia (fig. 23, 24) and the great abundance of uninucleate amoebae in infected cells. The latter may well be the result of multiple infection, while stages such as are shown in figures 23 and 24 may possibly represent, as Maire and Tison earlier interpreted them, fusions of uni- and binucleate amoebae with multinucleate plasmodia. While the author readily admits the possibility of schizogony, he does not consider the evidence so far presented as sufficiently reliable to have conclusively settled the problem. In this connection it is significant to note that schizogony has not been recorded in species, such as *S. radicalis*, where the process if present could be readily observed in living material.

The vegetative phase is terminated by the so-called transitional stage after which follow cleavage and meiosis, as has been described in Chapters II and III. The plastic cleavage segments or incipient resting spores become associated in a globular mass (fig. 44) and resemble myxamoebae in a pseudoplasmodium. By mutual readjustment they soon move to the periphery (fig. 44) and thus form a hollow sphere or ellipsoid. At this early stage the center of the mass is filled with a viscuous fluid, doubtless a residue of the plasmodium which is not used up in cleavage. Whether or not this substance represents extraneous waste material which is dumped into a central vacuole in the dedifferentiation of the protoplasm preparatory to sporogenesis as in various proteomyxean species is not certain. Maire and Tison stated that it has an osmotic coefficient and exerts centrifugal pressure on the spores whereby they are pushed to the periphery of the mass.

Shortly after their arrival there, the individual spores develop delicate walls which thicken and turn brown with maturity and often become verrucose. No evidence of cellulose or pectin was found in these walls by Maire and Tison. By mutual compression the spores usually become pentagonally and hexagonally pyramidal in shape with convex external and slightly concave internal surfaces. According to Winge, a collar is formed at the apex or external surface (fig. 48), but this structure has not been recorded by other workers. Occasional bi- and trinucleate spores occur (fig. 49–51), which may have arisen by incomplete cleavage or by subsequent division of the spore nucleus (fig. 50).

It is to be particularly noted that in none of the figures and descriptions of Blomfield and Schwartz or Maire and Tison which illustrate the aggregation of incipient resting spores and their transformation into cystosori is there evidence of a distinct, common enveloping membrane around the sorus. Likewise, it

is lacking in Rostrup's, Winge's, and Palm and Burk's figures of cystosori. Cook's ('33, Pl. 6, fig. 9) own photographs of *S. Veronicae* fail to show a distinct membrane. Nonetheless, he has often contended that it is present and has used ('33) the presence of this structure as one of the distinguishing generic characters of *Sorosphaera* as well as *Sorodiscus*. In the original diagnosis of the genus, Schroeter described the cystosori as being surrounded by a common cuticle, and this may be partly responsible for Nemec's ('11) and Cook's contention as to the presence of a membrane. Webb described it as being formed after the spores had developed their individual walls, but he gave no figures of its development. Winge ('13, p. 30) denied its existence, while Blomfield and Schwartz as well as Maire and Tison, who have so far made the most extensive study of the genus, said nothing about it. It is quite probable that the adjacent lateral walls of the spores become more or less fused by mutual pressure as they develop, and this prevents the spores from separating readily at maturity. The best cytological data in the literature to date do not, therefore, support Cook's view on the presence of a membrane, and the use of this structure as a diagnostic generic character is at present open to serious question.

The cystosori of *S. Veronicae* are predominantly hollow spheres and ellipsoids, but numerous variations in shape have been noted by Maire and Tison, Trotter, Webb, and others. Palm and Burk in particular found them to be unusually variable in galls on *V. americana* collected in Colorado, U. S. A. In this material they found the cystosori to be three principal shapes: hollow spheres, flattened ellipsoids, and irregular sponge-like masses, and between these types all degrees of variations and intergradations were observed. As is shown in figures 52 to 57, the *Sorosphaera*- or hollow-sphere type predominated, but two-layered flattened discs as in *Sorodiscus* (fig. 53, 54), spongy masses with narrow or wide channels as in *Spongospora* (fig. 55, 56), and irregular masses of indeterminate shape as in *Ligniera* (fig. 57) were not uncommon. Likewise within the same sorus, spores with smooth and verrucose walls were present (fig. 53, 55, 57). Palm and Burk accordingly concluded that the shape of the cystosorus and the relative arrangement of the spores are governed largely by environmental conditions and that the size and shape of the host cell are determining factors. They furthermore concluded that since sori typical of those of *Spongospora, Sorodiscus, Ligniera, Ostenfeldiella, Clathrosorus,* and *Membranosorus* may all be found in *S. Veronicae,* these genera should be regarded as synonyms of *Sorosphaera.* Fitzpatrick ('30) believed that *Ligniera* also should be incorporated in *Sorosphaera* on the grounds that the only difference between the two is that the former causes no hypertrophy of the host.

In 1907 Speschnew (p. 22, Pl. 2, fig. 7–12) described a species on tea leaves in the Caucasus which he named *Sorosphaera theae.* Two years later, however, Ducommet ('09) reported that no organism is present in the leaves and that the so-called spores are only tannin deposits in the cells. Fitzpatrick referred to this species as *Plasmodiophora Theae.*

S. VERONICAE Schroeter, l.c.

Tubercinia Veronicae Schroeter. 1877. Beitr. Biol. Pflanz. 2: 383.

Sorosporium Veronicae Winter. 1884. Die Pilze Deutschlands, Oesterreich und der Schweiz 1: 103.

Cystosori bright brown, one to several in a cell, variable in size and shape, predominantly in the form of hollow spheres, 18–42 μ, occasionally elongate, flat and disc-shaped, irregular and indeterminate, compact or loose and spongy with numerous ramifying channels, composed of from four to 64 spores. Resting spores ovoid, pyramidal, urn-shaped 4–5 $\mu \times$ 8–9 μ, with brown, smooth or verrucose outer walls, often surmounted by an apical collar. Zoospores biflagellate and heterocont. Plasmodia one to several in a cell, 20–30 μ, schizogony doubtful; producing a single cystosorus. Zoosporangia unknown.

Parasitic in *Veronica hederaefolia, V. chamaedrys,* and *V. triphyllos* in Germany (Schroeter, '77, '86, '97; Winter, l.c.; Diedicke, '11; Grevillius, '13); *V. saxatilis, V. officinalis, V. hederaefolia, V. scutellata, V. Beccabunga, V. Anagallis, V. aquatica, V. serpyllifolia,* and *V. Chaemaedrys* in Finland, Norway and Sweden (Lagerheim: see Winge, '13; Palm, '08); *V. hederaefolio* in Schleswig-Holstein and Denmark (Hennings, '91; Rostrup, '94); *V. Chamaedrys* in France (Maire and Tison, '08, '09, '10, '11; Maire, '10); *V. Chamaedrys* in England (Blomfield and Schwartz, '10; Cook and Schwartz, '29); *V. arvensis* and *V. hederaefolia* in Italy (Trotter, '04, '16); *V. americana* and *V. arvensis* in the U. S. A. (Palm and Burk, '33; Donald, '34), causing tumors up to 5 mm. in diameter on the stems, petioles, and midrib of leaves.

This species was first described by Schroeter in 1877 as a member of the Ustilaginales under the name *Tubercinia Veronicae,* and in 1884 Winter transferred it to the genus *Sorosporium.* In 1886, however, Schroeter created the genus *Sorosphaera* for it and transferred it to his newly established Phytomyxinae. Rostrup found it in Denmark in 1894 and replaced it in the Ustilaginales, and according to Winge, Lagerheim found it in Norway and Sweden on a large number of *Veronica* species, and as early as 1901, "and knew the correct systematic position of *Sorosphaera.*" Trotter discovered it in Italy in 1904, and while he questioned its inclusion among the smuts, he also doubted that it is a member of the Mycetozoa. The subsequent studies of Maire and Tison and Blomfield and Schwartz clearly showed that it belongs in the Plasmodiophoraceae in close relation to *P. Brassicae.*

The tumors caused by *S. Veronicae* vary from pinhead size to 5 cm. in diameter and are usually composed of a mass of healthy and infected undifferentiated cells among which are interspersed a few spiral and annular vessels. The galls are the result of

both cell multiplication and cell enlargement with the latter process playing the dominant role in the later stages of development. Since the parasite has a predilection for the provascular strands in the apical meristem, the tumors may involve the entire stem in instances of severe infection. In such cases the primordia of the stems and leaves are reduced to a mass of cells in which pith, cortex, etc., are indistinguishable. In less extensive infections only small portions of the stem become involved, and the normal growth of the plant is not seriously affected. According to Lagerheim, the development of the vascular ring is suppressed in the region of infection, while in the outer cortex the collenchyma is still present. The remaining cortical cells become tangentially oriented in growth and greatly enlarged. The epidermal cells become isodiametric, and the guard cells of the stomata are often considerably enlarged, with the pore itself abnormally wide.

Although infection has not been observed, *S. Veronicae* appears to make its initial entrance in the apical meristem, because the youngest plasmodia and smallest galls occur in or near the apex. Blomfield and Schwartz succeeded in producing tumors on *Veronica* seedlings by spraying them with water containing crushed cystosori and found single, isolated infected cells close to the growing point. The amoebae of the parasite are apparently unable to pass through the walls into adjacent cells. According to Blomfield and Schwartz, and Cook ('33), they are passively distributed by the repeated division of infected provascular cells. If the young plasmodia undergo schizogony, as Maire and Tison reported, the number of amoebae is greatly increased, and by repeated division of infected cells, large diseased areas are soon formed. In the early stages of the disease the presence of the parasite apparently does not inhibit cytokinesis of the host cells, but later on after they have become enlarged the latter lose the ability to divide. The enormously enlarged nuclei, however, undergo several mitoses with the result that the infected cells become multinucleate (fig. 3). Division of the host nuclei is greatest at the close of the vegetative stage of the parasite, but with the onset of the sporogonic phase mitosis ceases. At this stage the host nuclei become distorted (fig. 5), more densely stainable, (fig. 4), and eventually disintegrate (fig. 6). By the time the cystosori are mature, only atrophied and degenerated nuclei are to be found, according to Blomfield and Schwartz. On the other hand, Maire and Tison reported that the nuclei as well as plastids and starch grains may persist long after the sori have matured.

In the early stages of infection only slight enlargement of the host cells occurs, but as the plasmodia increase in size, marked expansion takes place. In exceptional cases infected cells may enlarge to 20 times their normal diameter. *Sorosphaera Veronicae* accordingly not only causes enormous cell enlargement but also prevents cell differentiation. Adjacent healthy cells as well as stomatal guard cells may also be stimulated to enlarge by the presence of the parasite. As is shown in figure 2, there appears to be no visible antagonism between the protoplasm of the host and pathogen. The latter lies embedded in the host cytoplasm and in the young stages may be closely associated with the host nucleus. Infected cells may contain numerous plastids and starch grains, but these are not so abundant as in the adjacent healthy cells. According to Lagerheim, epidermal cells in the infected regions are richer in crystals than those in healthy portions of the stem.

Slugs frequently feed on the galls, and it is believed that they play a significant role in spreading the disease. Most tumors soon soften and decay, liberating the cystosori into the soil, where the resting spores germinate. When new plants push up through the soil, their apices apparently become infected. *Sorosphaera Veronicae* has never been found parasitizing the roots.

Nematodes also may cause galls on *Veronica* which are strikingly like those produced by *S. Veronicae* and may easily be mistaken for them. For this reason Cook ('33) regarded with suspicion the reports of Lagerheim and Winge of the presence of the parasite in a large number of *Veronicae* species in Norway and Sweden.

S. RADICALIS Cook. 1933. Arch. Protistk. 80: 201. Pl. 7, fig. 10, 11.

S. radicale Cook and Schwartz. 1929. Ann. Bot. 43: 86. Pl. 2.

Cystosori single and partly filling host cell, hollow, rarely spherical, 20 μ, usually oval, ellipsoidal and elongate, 16–20 $\mu \times$ 20–57 μ, bright yellowish-brown; including up to 500 spores. Resting spores oval, 3 $\times$ 4 μ, with thin yellowish-brown, smooth walls; producing one zoospore in germination. Zoospores oval and spherical, 2–3 μ, with an anterior flagellum (?) 4–6 μ long. Evanescent zoosporangia unknown. Plasmodia single and partly filling host cell, 20–60 μ in diameter, producing one cystosorus; schizogony lacking (?).

Parasitic in the root hairs only of *Poa fluitans Monilia caerulea, Catabrosa aquatica,* and other grasses in England, causing localized enlargement of the infected cells.[1]

This species is distinguishable from *S. Veronicae* primarily by its oval, ellipsoidal and elongate cystosori which are also much larger and composed of a greater number of small resting spores. In addition, its nuclei are considerably smaller. While *S. radicalis* may occur in the same vicinity with and infect some of the hosts of *Ligniera Junci* as well as *L. verrucosa* and *L. pilorum,* Cook and Schwartz maintained that it is quite distinct. However, it is to be noted here that these *Ligniera* species may also occur in locally

[1] In recent correspondence with the author, Prof. J. T. Barrett, College of Agriculture, California University, reported that he had found what he believes to be *S. radicalis* in roots of *Poa annua* on the college campus. In addition to cystosori and resting spores, he observed thin-walled sporangia which produce biflagellate, heterocont zoospores. Barrett thus confirms Ledingham's previous report of such zoospores in *Sorosphaera.*

swollen root hairs and occasionally form almost spherical, oval and elongate hollow cystosori.

Sorosphaera radicalis has been found only in root hairs and does not attack the other tissues of the root. Hence no external symptoms of the disease are visible on the host plant except a slight reddening of the stem and leaf bases. When the infected root hairs decay, the cystosori are liberated into the soil. Infection by zoospores apparently occurs during the early developmental stages of the root hairs.

Although Cook and Schwartz failed to count the chromosomes, they nonetheless believed that meiosis occurs during the first of the two last divisions preceding sporogenesis. No evidence of gametic fusion has been observed.

Cook and Schwartz reported that at the conclusion of promitosis "a wall is now secreted around the plasmodium, and the whole mass passes into a sporing stage." If this statement and observation are true, it is obvious that *S. radicalis* differs radically in this respect from all other known species of the Plasmodiophorales.

PLATE 6

Sorosphaera Veronicae

Fig. 1. *Veronica chamaedrys* with galls caused by *S. Veronicae*. Winge, '13.

Fig. 2. Hypertrophied host cell with six plasmodia. Note relative sizes of healthy and infected cells. Blomfield and Schwartz, '10.

Fig. 3. Hypertrophied host cell with five plasmodia. Four host nuclei in telophases of division. Blomfield and Schwartz, l.c.

Fig. 4. Nucleus of parasitized cell with numerous nucleoli. Blomfield and Schwartz, l.c.

Fig. 5. Lobed and distorted nucleus of an infected cell. Maire and Tison, '09.

Fig. 6. Old host cell with four cystosori; protoplasm almost completely gone. Blomfield and Schwartz, l.c.

S. radicalis

Fig. 7. Hypertrophied root hair with cystosorus in surface view. Cook and Schwartz, '29.

Fig. 8. Median longitudinal section of an ellipsoidal cystosorus. Cook and Schwartz, l.c.

S. Veronicae

Fig. 9. Uninucleate stage of thallus. Maire and Tison, l.c.

Fig. 10. Resting nucleus of young parasite. Blomfield and Schwartz, l.c.

Fig. 11. Beginning of promitosis of a 4-nucleate plasmodium with centrosomes and astral rays. Maire and Tison, l.c.

Fig. 12. "Saturn stage" of promitosis. Maire and Tison, l.c.

Fig. 13. Early anaphases. Maire and Tison, l.c.

Fig. 14. "Double-anchor" stage of promitosis. Maire and Tison, l.c.

Fig. 15. Late anaphases with centrosomes and asters. Maire and Tison, l.c.

Fig. 16. Late prophase of vegetative nuclei in plasmodium with four chromosomes. Webb, '35.

Fig. 17. Later stage showing four split chromosomes. Webb, l.c.

Fig. 18. Four chromosomes arranged in a ring around constricting nucleole. Webb, l.c.

Fig. 19. Metaphase; daughter chromosomes beginning to separate. Webb, l.c.

Fig. 20. Early anaphase. Two rings of four chromosomes each moving apart. Webb, l.c.

Fig. 21. Later anaphase. Webb, l.c.

Fig. 22. Telophases of promitosis. Maire and Tison, l.c.

Fig. 23, 24. Schizogony of plasmodium; uni- and binucleate segments respectively being split off. Maire and Tison, l.c.

Fig. 25. Beginning of akaryote stage; chromatin passing out into cytoplasm. Blomfield and Schwartz, l.c.

Fig. 26. Akaryote stage; nuclei clear and vacuole-like. Blomfield and Schwartz, l.c.

Fig. 27. Reconstructed nuclei following akaryote stage. Blomfield and Schwartz, l.c.

Fig. 28. Later stage showing reappearance of nucleoli and chromatin. Maire and Tison, l.c.

Fig. 29. So-called "garland" stage of reconstructed nuclei. Maire and Tison, l.c.

Fig. 30. Same stage highly magnified. Winge, l.c.

Fig. 31, 32. Synezesis (?). Maire and Tison, l.c.

Fig. 33. Beginning of cleavage into spore mother cells; appearance of nuclei suggestive of diakinesis. Maire and Tison, l.c.

Fig. 34. Early diakinesis. Webb, l.c.

Fig. 35. Diakinesis with four pairs of homologous chromosomes. Webb, l.c.

Fig. 36. Equatorial plate stage of heterotypic division during sporogenesis. Cleavage into spore mother cell complete. Maire and Tison, l.c.

Fig. 37. Same stage. Winge, l.c.

Fig. 38. Late anaphases of meiotic division; first division of spore mother cells beginning. Maire and Tison, l.c.

Fig. 39. First division of spore mother cells complete. Maire and Tison, l.c.

Fig. 40. Late prophase nucleus of second or homeotypic division with four chromosomes. Webb, l.c.

Fig. 41. Equatorial plate stage of second division during sporogenesis. Maire and Tison, l.c.

Fig. 42. Second cell division into incipient resting spores.

Fig. 43. Incipient resting spores aggregating into a globular mass; initial stage in formation of cystosorus. Maire and Tison, l.c.

Fig. 44. Later stage in cystosorus development; spores arranged at periphery with a viscous substance in the center. Maire and Tison, l.c.

Fig. 45. Young cystosorus in median section with well-defined walls around spores; remants of viscous substance in center. Maire and Tison, l.c.

Fig. 46. Cystosorus in median section. Blomfield and Schwartz, l.c.

Fig. 47. Portion of a cystosorus in surface view. Blomfield and Schwartz, l.c.

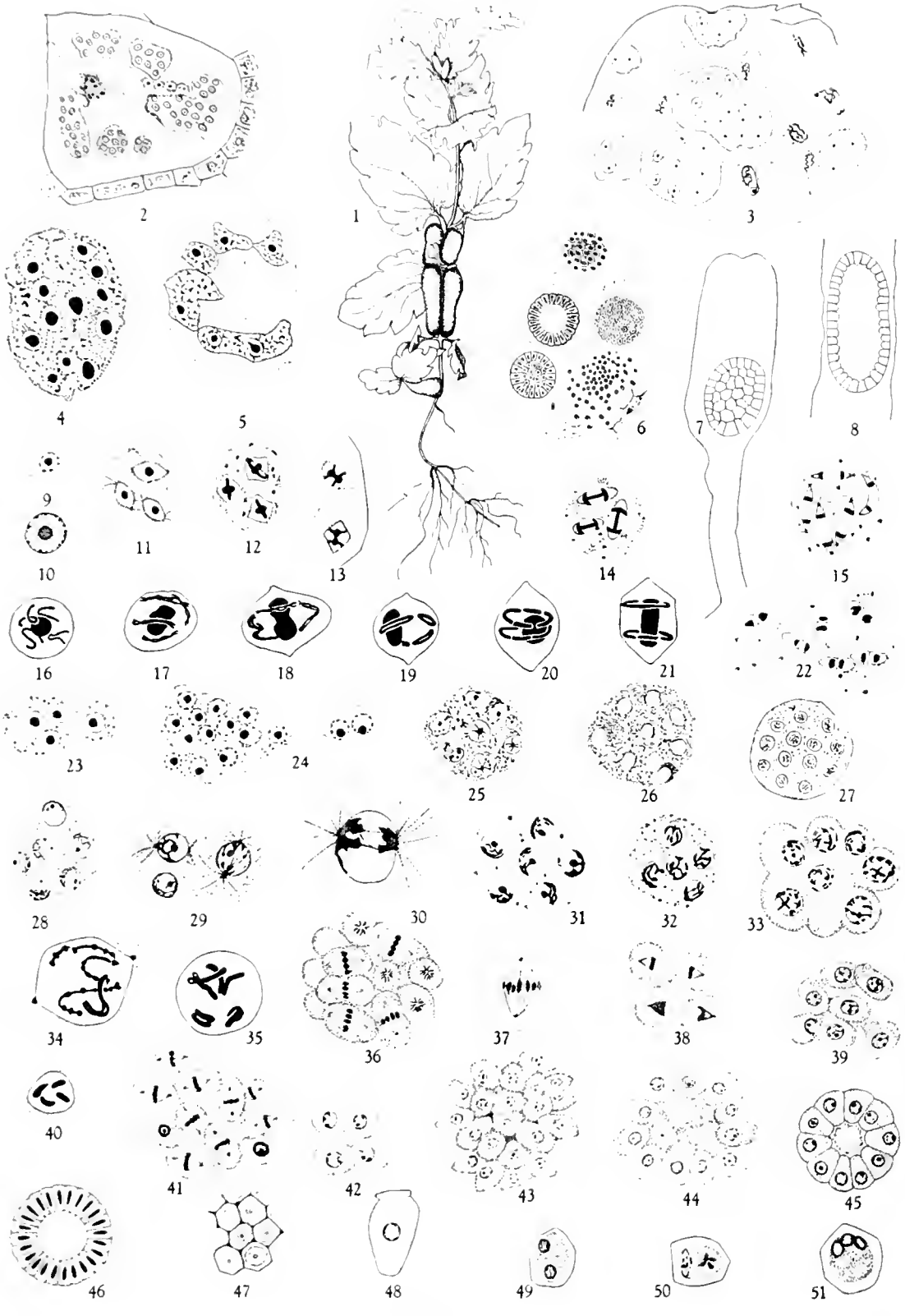
Fig. 48. Urn-shaped resting spore with apical collar. Winge, l.c.

Fig. 49. Binucleate resting spore. Maire and Tison, l.c.

Fig. 50. Division of nuclei in binucleate resting spore. Maire and Tison, l.c.

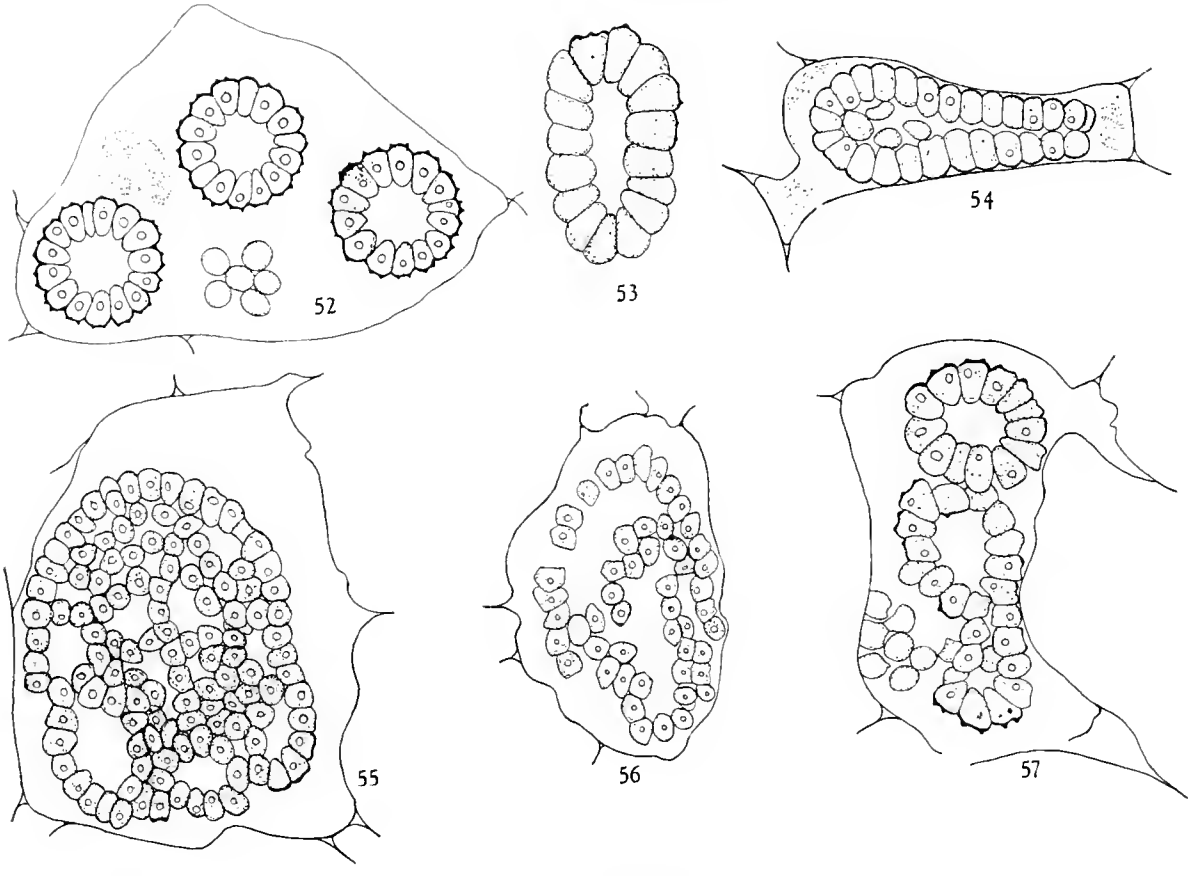
Fig. 51. Trinucleate resting spore. Maire and Tison, l.c.

PLATE 6



Sorosphaera

PLATE 6—*Continued*



Sorosphaera Veronicae

Fig. 52–57. Variations of the cystosori of *S. Veronicae*. Palm and Burk, '33.

Fig. 53. Typical hollow *Sorosphaera-like* cystosori with smooth and verrucose spores.

Fig. 54. Flattened *Sorodiscus*-like cystosorus.

Fig. 55. Spongy *Spongospora*-like cystosorus.

Fig. 56. Loose, spongy *Clathrosorus*-like cystosorus.

Fig. 57. Irregular *Ligniera*- and globular *Sorosphaera*-like cystosori.

ADDITIONAL BIBLIOGRAPHY: SOROSPHAERA

Blomfield, J. E., and E. J. Schwartz. 1910. Ann. Bot. 24: 35.
Diedicke, H. 1911. Mitt. Thür. bot. Ver. n. f. 28: 83.
Donald, L. 1934. Phytopath. 24: 843.
Ducomet, V. 1909. Ann. l'ecole Nat. d'Agric. Rennes 2: 1.
Grevillius, A. Y. 1913. Abh. Ver. Nat. Erfors. Niederrheins 1: 165.
Henning, P. 1891. Schrift. Nat. Ver. Schleswig-Holstein 9: 235.
Ledingham, G. A. 1939. Canadian Jour. Res. C 17: 43.
Maire, R. 1909. Ann. Mycol. 7: 226.
———. 1910. Bull. Soc. Linn. Normandie 6 ser. 2: 57.
———. 1911. Ann. Mycol. 9: 226.
———, and A. Tison. 1908. C. R. Acad. Sci. Paris 147: 1110.
Palm, B. T. 1908. Svensk. Bot. Tids. 2: 48.
———, and M. Burk. 1933. Arch. Protistk. 79: 263.
Rostrup, E. 1894. Bot. Tidsskr. 19: 201.
Schwartz, E. J. 1914. Ann. Bot. 28: 229.
Smith, A. L., and J. Ramsbottom. 1917. Trans. Brit. Mycol. Soc. 6: 231.
Speschnew, N. N. 1907. Die Pilzparasiten des Teestrauches, p. 22. Berlin.
Trotter, A. 1904. Ann. Mycol. 2: 536.
———. 1916. Marcellia 15: 58.
Webb, P. C. R. 1935. Ann. Bot. 49: 41.
Winge, O. 1913. Ark. f. Bot. 12, no. 9: 4.

SORODISCUS

Lagerheim and Winge, 1913. Ark. f. Bot. 12: 23

(PLATES 7, 8)

Cystosori usually flat, oval, disc-shaped and composed of two layers of spores pressed closely together; often variable in size and shape, rarely hollow spheres, occasionally an elongate and irregular linear series of spores or reduced to tetrads, triads, diads and rarely monads; soral membrane doubtful or lacking. Resting spores polygonal, angular and urn-shaped or oval and almost hemispherical with hyaline smooth or spiny outer walls; apical collar and cap present or lacking; remaining attached together or separating at maturity; producing one or

possibly more than one zoospore in germination. Zoosporangia unknown. Plasmodia one to several in a cell, large or small; schizogony lacking or doubtful in some species.

Sorodiscus includes at present three or possibly four species (see *Membranosorus* in this connection) which have been reported from Russia, Norway, Sweden, South Africa and the United States. They occur under fairly moist and aquatic conditions, parasitize algae and higher plants, and cause marked hypertrophy of the host in the form of galls or tumors which may be uni- or multicellular. Since most of these species have been studied only from fixed and stained material many of the critical developmental stages are poorly known, and the various claims concerning the presence of sexuality, karyogamy, meiosis, alternation of haploid and diploid generations, etc., are obviously based on inadequate cytological data.

Furthermore, the outstanding character of the genus, namely, oval and almost circular, flat and disc-shaped cystosori composed of two closely pressed layers of resting spores, has been seriously questioned. In the type species, *S. Callitrichis*, the cystosori may sometimes be hollow spheres, while in *S. karlingii* they may vary from hemispherical multinucleate monads, diads, triads, tetrad, flat discs, and elongate linear series of spores to almost hollow spheres (Pl. 8, fig. 11–21). In all of the species, however, the majority of cystosori are flattened and disc-like. While Winge ('13) regarded *Sorodiscus* as a distinct genus he nonetheless pointed out that its similarity to *Sorosphaera* is so great "that it would seem most reasonable to unite them into one genus." Later, however, in a communication to Cook ('31, p. 318) he said "that the spore masses are so characteristic in *Sorodiscus* that it would be wrong to put it in the same genus as *Sorosphaera*." Palm and Burk ('33), on the other hand, regarded *Sorodiscus* as a synonym of the latter genus.

Schizogony has not been observed in *Sorodiscus*, although Winge believed that the widespread distribution of amoebae in the galls formed by *S. Callitrichis* suggests its occurrence. Whether or not a common enveloping membrane is present around the cystosori in all species is uncertain at present. Furthermore, little is known about the origin and development of this membrane in the species in which it has been reported to occur. In *S. Callitrichis*, according to Winge, the resting "spore-wall divides into two layers of which the outer one merges into that of the neighboring spores (fig. 31, 32) so that it gives one the impression of the spores being deposited in a common substance." According to this statement no distinct and separate wall is formed, and the spores are merely adherent by the outer layer of their walls. Figure 33, however, shows an enveloping membrane. Cook considered Winge's interpretation incorrect and stated that in *S. radicicolus* a distinct wall is laid down around the cystosori. He did not, however, present any evidence about its origin—whether it consists of the original bounding membrane of the plasmodium present at the time of cleavage or is deposited subsequently by the maturing resting spores. Furthermore, his figures 23 and 24 of mature spore cakes do not show a separate common wall around the spores. In *S. karlingii* no evidence of an enveloping membrane has yet been observed (fig. 11–21). The presence of such a membrane in the genus as a whole is thus still open to serious question, and if present its origin and method of development are certainly in need of intensive cytological study.

Winge and Cook differed also in their observations relative to sporogenesis and the stage at which meiosis occurs. In *S. Callitrichis* numerous binucleate segments or spore mother cells are formed by progressive cleavage of the plasmodium (fig. 27), and these segments (fig. 28 and 29) then divide once to form groups of spores in twos (fig. 30), according to Winge. These groups of incipient resting spores soon aggregate together, deposit two-layered walls (fig. 28, 29), and thus form the characteristic cystosori (fig. 33). In *S. radicicolus*, however, according to Cook ('33, p. 207), the primary cleavage segments or spore mother cells (fig. 20) divide twice to form four instead of two incipient resting spores. Cook did not show clearly how these united to form the cystosorus and an enveloping wall. It may be that the two species actually differ in this respect, but further study is necessary to determine this point. If Winge's and Cook's accounts are correct *Sorodiscus* shows marked similarity to *Sorosphaera* by the presence of spore mother cells which divide into diads and tetrads and subsequently aggregate into sori.

S. CALLITRICHIS Lagerheim and Winge, l.c., p. 23. Pl. 1, fig. 9, 10; Pl. 2; Pl. 3, fig. 43–63.

Cystosori up to 10 in a cell, usually circular, flat and disc-shaped, 30–45 μ × 40–65 μ × 12–14 μ, rarely spherical and hollow; composed of up to 200 resting spores usually arranged in two layers and closely pressed together; outer layer of spore walls continuous (?). Resting spores urn-shaped in longitudinal section and hexagonal in cross section, 4–5 μ × 6–7 μ, with smooth hyaline walls surmounted at the apex by a collar; germination unknown. Zoosporangia and zoospores unknown. Plasmodia one to several in a cell, large, 40–60 μ in diam., each forming one cystosorus; schizogony doubtful or lacking; cleaving at maturity into binucleate segments or spore mother cells which divide once (?) into two resting spores.

Parasitic in *Callitriche vernalis* in Norway (Lagerheim and Winge, l.c.) and *C. autumnalis* in Russia (Kareltschikoff and Rosanoff, '70) and Sweden (Ostenfeld), causing globular galls up to 3×7 mm. on the primary and secondary axes.

This species was first recorded in 1870 by Kareltschikoff and Rosanoff who mistook the cystosori for cystoliths and compared them with those present in the Urticaceae, although Rosanoff was of the opinion that they might be remnants of a parasitic mycelium. According to Winge, Lagerheim collected

this species on *C. vernalis* in Norway in 1893 and 1900, and although he fixed, sectioned and studied his material he published nothing but passed the material on to Winge. In 1907 Rosenfeld (Anonymous, '08) discovered the fungus on *C. autumnalis* in Sweden, and since that time it has not been reported.

Sorodiscus Callitrichis has a marked effect on the host. All parts of the stem except the outermost cortical tissues and epidermis are attacked, and the vascular bundles become displaced and lie scattered about in the tumors or are completely destroyed. Infected cells may often enlarge to 10 times their normal diameter, but whether or not they and adjacent healthy ones are stimulated to divide by the fungus is unknown. It is not improbable, however, that the galls are due to both cell enlargement and cell multiplication. The nucleus of the host cell apparently enlarges also and forms several conspicuous nucleoli. So far nothing is known about the site and method of infection.

S. RADICICOLUS Cook, 1931. Ann. Mycol. 29: 321. Pls. 1, 2.

Cystosori one to several in a cell, usually flat and disc-shaped; composed of up to 50 resting spores usually arranged in two layers and closely pressed together; enveloped in a delicate membrane which later disintegrates and frees the individual spores. Resting spores oval, rectangular and polygonal in section, 3.8–4.2 μ × 3.2–3.6 μ, with smooth walls, the outer layer of which may be extended to form blunt spines; separating at maturity and giving rise to zoospores in germination. Zoospores oval pyriform, 2.5–3.5 μ, soon becoming amoeboid. Zoosporangia unknown. Plasmodia one to several in a cell, small 15–30 μ in diameter; schizogony doubtful or lacking; each producing a single cystosorus; at maturity cleaving into uninucleate segments or spore mother cells which divide twice into four resting spores.

Parasitic in the roots of *Gynandropsis pentaphylla* near Pretoria, South Africa, causing convoluted, coral-like galls, 3–15 mm. in diameter.

Cook's study of this species was based entirely on prepared slides and fixed material sent by Dr. E. M. Doidge from South Africa. It has accordingly never been examined in the living state. Many of the critical developmental stages such as resting spore germination, fusion of gametes, schizogony, alternation

PLATE 7

Sorodiscus

(Fig. 1, 2, 9–15, 23–33 after Winge, '13. Remaining figures after Cook, '31. Fig. 8 drawn from portion of a photograph.)

Fig. 1. *Callitriche vernalis* with numerous galls caused by *S. Callitrichis*.

Fig. 2. Enlarged gall.

Fig. 3. Diagrammatic sketch of cross section through the root and gall of *Gynandropsis pentaphylla* showing the progressive developmental stages of parasite from the root to the opposite side of tumor. *S. radicicolus*.

Fig. 4. Anteriorly flagellate zoospore or gamete. *S. radicicolus*.

Fig. 5. Amoeboid stage of same. *S. radicicolus*.

Fig. 6. Fusion of gametes. *S. radicicolus*.

Fig. 7. Young binucleate plasmodium after first division of zygote nucleus. *S. radicicolus*.

Fig. 8. Large multinucleate plasmodium. *S. radicicolus*.

Fig. 9. Resting nucleus of plasmodium. *S. Callitrichis*.

Fig. 10. Prophase, showing differentiation of "tropho" and "idiochromatin." *S. Callitrichis*.

Fig. 11. Equatorial plate stage of "promitosis" (?). *S. Callitrichis*.

Fig. 12. Similar stage from a young plasmodium showing chromatin aggregated into chromosome-like bodies. *S. Callitrichis*.

Fig. 13. Anaphase with conspicuous astral rays. *S. Callitrichis*.

Fig. 14. Late telophase showing differentiation of "tropo-" and "idiochromatin." *S. Callitrichis*.

Fig. 15. Daughter nuclei with well-marked centrosome-like bodies and astral rays. *S. Callitrichis*.

Fig. 16. Beginning of chromatin discharge into cytoplasm from nucleus during chromidial stage. *S. radicicolus*.

Fig. 17. Later stage showing karyosome broken up into granules which lie at the inner periphery of nucleus. *S. radicicolus*.

Fig. 18. Final akaryote stage with all stainable chromatin discharged from nucleus. *S. radicicolus*.

Fig. 19. Prophase of meiosis, the so-called "garland stage." *S. radicicolus*.

Fig. 20. Equatorial plate stage of meiosis with four chromosomes. Plasmodium segmenting into spore mother cells. *S. radicicolus*.

Fig. 21. Binucleate spore mother cell *S. radicicolus*.

Fig. 22. Second meiotic division with two chromosomes. *S. radicicolus*.

Fig. 23–24. So-called "garland" stages in *S. Callitrichis*.

Fig. 25. Equatorial plate stage of the first (homeotypic) division. *S. Callitrichis*.

Fig. 26. Equatorial plate stages of meiosis. Plasmodium cleaving into segments. *S. Callitrichis*.

Fig. 27. Paired daughter nuclei in cleaving plasmodium. *S. Callitrichis*.

Fig. 28, 29. Binucleate segment of plasmodium. *S. Callitrichis*.

Fig. 30. Four incipient resting spores resulting from cleavage of two binucleate segments. Nuclei quite large. *S. Callitrichis*.

Fig. 31. Mature spores with two-layered walls, the outer layer merging with that of neighboring spores. *S. Callitrichis*.

Fig. 32. Young spores with outer and inner walls. *S. Callitrichis*.

Fig. 33. Side view of cystosorus of *S. Callitrichis* showing common enveloping membrane.

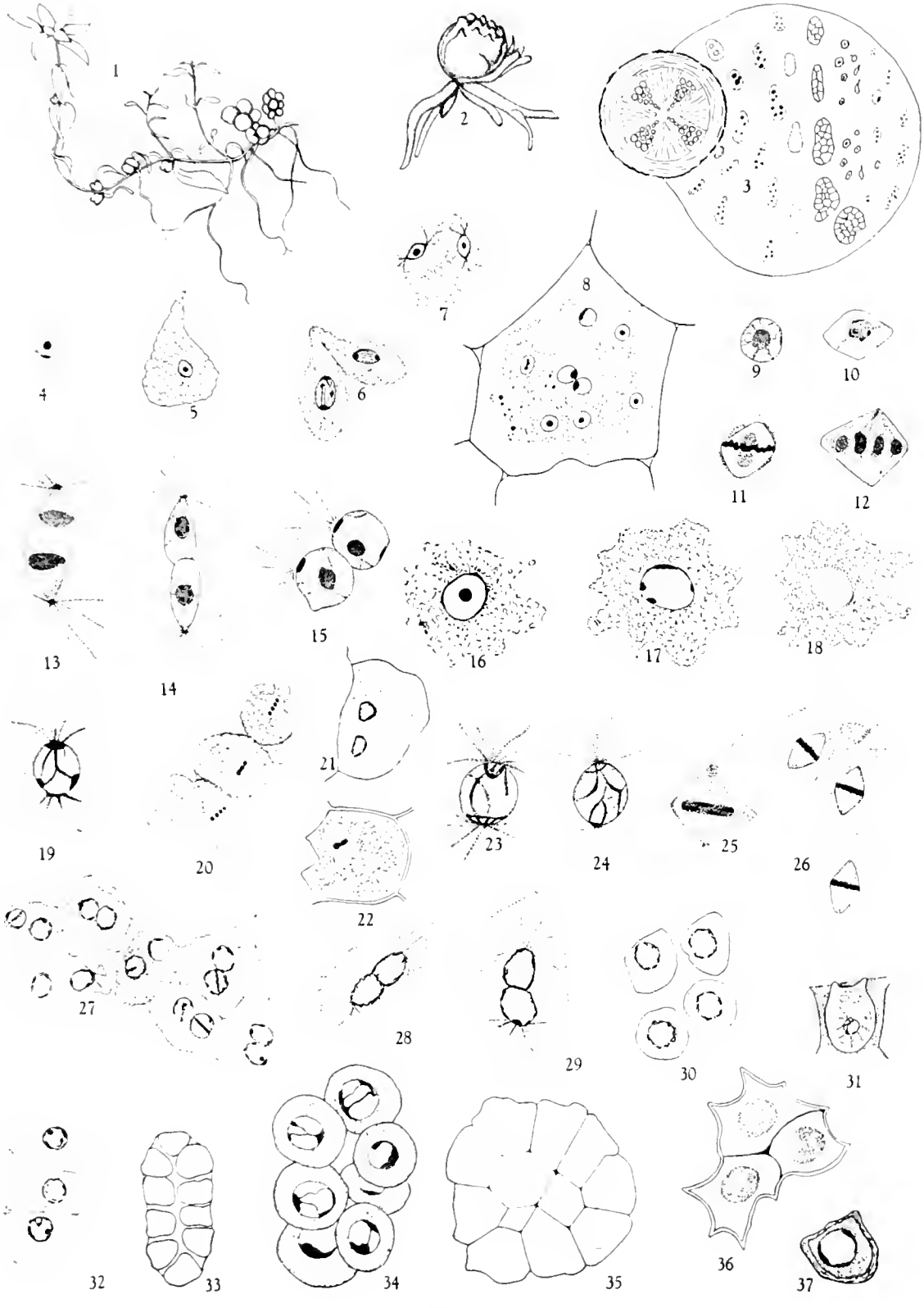
Fig. 34. Young spore with incompletely formed walls. *S. radicicolus*.

Fig. 35. Surface view of small cystosorus. *S. radicicolus*.

Fig. 36. Spiny resting spores. *S. radicicolus*.

Fig. 37. Thick-walled resting spore broken away from cystosorus. *S. radicicolus*.

PLATE 7



Sorodiscus

of haploid and diploid generations, etc., are thus in need of further investigation.

The method of infection has not been observed, but Cook believed that the amoeboid zoospores or amoebae are capable of passing through the walls from cell to cell and even to the outside of the host where they may infect other roots. As is shown in figure 3 at least two generations of the parasite may occur in large galls during the course of one season, but the host plants are not seriously affected by the presence of the galls and fungus. The central cylinder of the roots apparently is not attacked, and the galls seem to originate in the cortex, although Cook was not at all clear about their origin. Infected cells do not enlarge greatly, but their nuclei eventually become disorganized and degenerate. The presence of the fungus may possibly stimulate cytokinesis or at least does not prevent division of infected and adjacent healthy cells. The galls are therefore doubtless due to both cell enlargement and cell multiplication.

S. KARLINGII Cook, 1933, l.c. p. 207. Karling, 1928. Am. Jour. Bot. 15: 485. Pl. 32, fig. 1–9.

Cystosori numerous, up to 400 in a cell, quite variable in size and shape, often oval, elongate and disc-shaped, 15–30 $\mu \times$ 45–70 μ, occasionally almost spherical, 10–35 μ in diameter, irregular, or reduced to tetrads, triads, diads and rarely monads; consisting of from 1 to 200 spores; enveloping membrane unknown. Resting spores polygonal and angular, 4–9 μ, when pressed together in large sori, spherical, oval and ellipsoidal when single or in small groups, 5–23 μ in diameter, uni- or multinucleate with hyaline smooth walls and surmounted by one and occasionally two fairly thick caps; germination unknown. Plasmodia one to several in a cell, multinucleate, and up to 90 μ in diameter; schizogony unknown. Zoosporangia and zoospores unknown.

Parasitic in *Chara contraria* and *C. delicatula* in New York City, causing marked hypertrophy of the infected cells.

This is the only known species which parasitizes algae. Because of the great variation in the size and shape of its cystosori and the lack of a common surrounding membrane, it is a doubtful member of *Sorodiscus,* and until more is known about its life cycle it is retained only provisionally in this genus. Its effect on the host is quite marked and extensive, and all cells appear to be equally susceptible. Hypertrophied stipules, leaflets, spicules, internodal and cortical cells have frequently been found. As is shown in figures 1 and 2 infected cells may swell to many times their normal diameter and have the appearance of spherical, oval and elongate green blisters.

The presence of the plasmodia has no visible effect at first on the streaming of the host protoplasm and are continually carried along passively with the host nuclei and cytoplasm. Individual hypertrophied cortical cells have been removed from the leaves and kept alive in hanging drops for ten days, during which period the plasmodia, host nuclei and cytoplasm rotated continually. The streaming begins to slow down in about 12 days and ceases entirely within 20 days, after which the cell soon dies. As is shown in figure 3 the host nuclei and cytoplasm appear normal during the actively streaming period, and in spite of the extension which it has undergone the cell wall remains normal in thickness. Later, the host protoplasm is reduced to a thin layer. The presence of the parasite may also often lead to the formation of an abundance of storage starch grains in the plastids.

The cystosori, which were previously ('28) called sporangesori by the author, are quite variable in size and shape, and those consisting of a few large multinucleate and several small uni-nucleate spores (fig. 15, 21) have possibly arisen by unequal and incomplete cleavage of the plasmodium. The unusually large multinucleate spores (fig. 19) are probably the result of the encystment of the entire plasmodia which failed to segment. Since such spores are multinucleate, it is not improbable that they form several zoospore in germination.

ADDITIONAL BIBLIOGRAPHY: SORODISCUS

Anonymous. 1908. Bot. Tidsskr. 28: XXVII.
Cook, W. R. I. 1933. Arch. Protistk. 80: 203.
Kareltschikoff, S., and S. Rosanoff. 1870. Mem. Soc. Sci. Nat. Cherbourg. 2nd ser. 5: 124.
Palm, B. T., and M. Burk. 1933. Arch. Protistk. 79: 271.
Schwartz, E. J. 1944. Ann. Bot. 28: 230.

PLATE 8

Sorodiscus karlingii

(All figures after Karling)

Fig. 1. Hypertrophied internodal cell of *C. delicatula* which has burst the sheath of cortical cells.

Fig. 2. An extreme case of parasitism of the cortical cells of *C. contraria.*

Fig. 3. Longitudinal section of an enlarged cortical cell with twenty-six cystosori and seven plasmodia surrounded by the host protoplasm. The six host nuclei appear normal.

Fig. 4–6. Uni-, bi- and tetranucleate stages of the parasite.

Fig. 7. A multinucleate vacuolate plasmodium in surface view.

Fig. 8. Similar plasmodium in edge view.

Fig. 9. Large irregular plasmodium.

Fig. 10. Cleavage of plasmodium to form cystosorus.

Fig. 11. Surface view of a large flattened cystosorus consisting of approximately 200 spores.

Fig. 12. An almost spherical cystosorus.

Fig. 13. Flattened cystosorus in end view.

Fig. 14. Tetrad of resting spores.

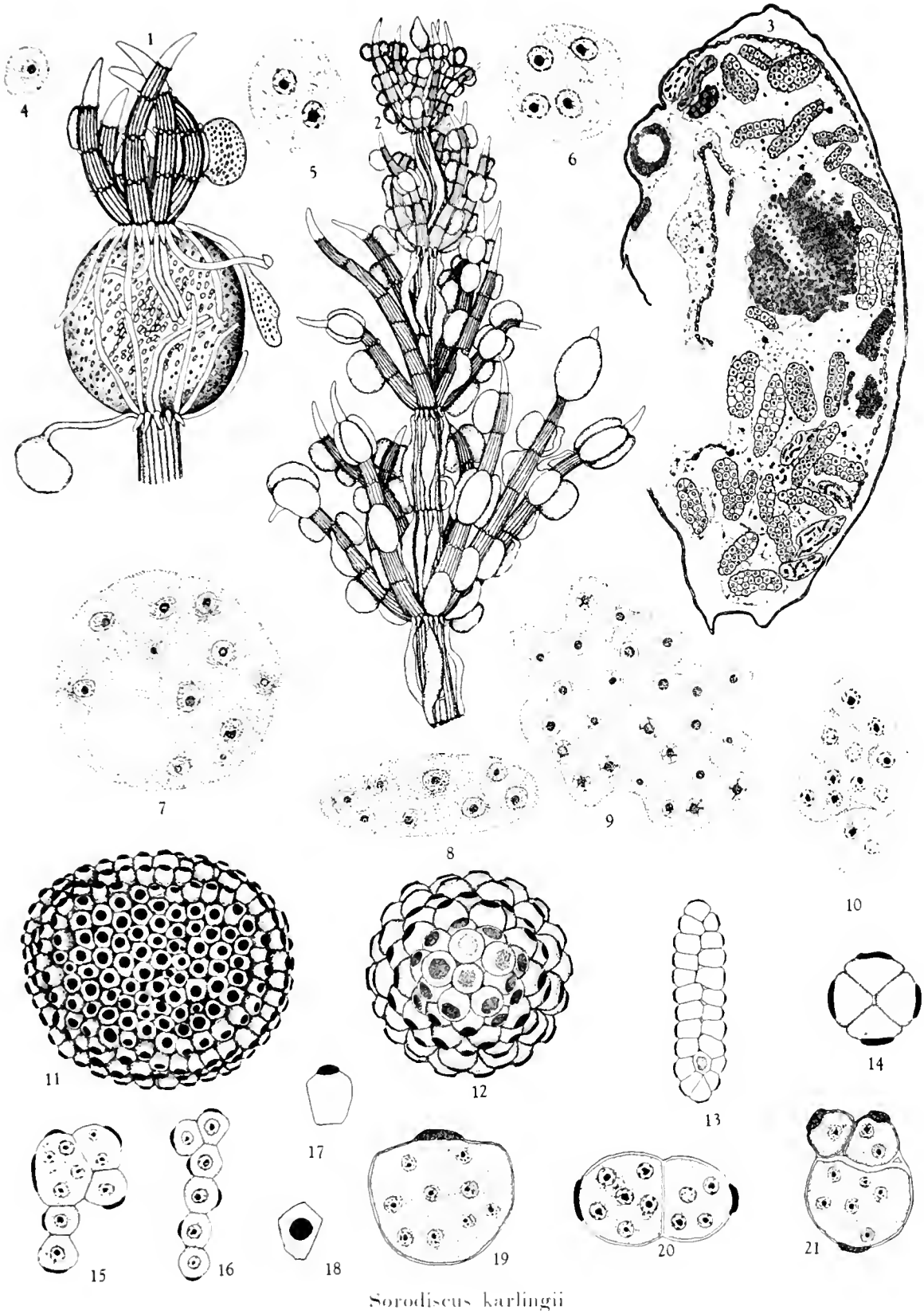
Fig. 15, 16. Further variations in size and shape of cystosori.

Fig. 17, 18. Small resting spores in side and surface views showing the apical caps.

Fig. 19. Large isolated multinucleate spore.

Fig. 20, 21. Cystosori consisting of two and three spores.

PLATE 8



Sorodiscus karlingii

The genus *Membranosorus* has been regarded as a synonym of *Sorodiscus*, but inasmuch as its inclusion in this genus as well as in *Sorosphaera* is highly questionable at present it seems advisable for the time being to discuss it separately.

MEMBRANOSORUS

Ostenfeld and Petersen, 1930. Zeitschr. f. Bot. 23: 17.

(PLATE 9)

Cystosori one or more in a cell, variable in size and shape; frequently a hollow, single-layered structure which covers the inner periphery of the host cell and conforms to the latter's size and shape; often oval, disc-like and single-layered, rarely double-layered, occasionally composed of an irregular mass of loosely attached spores or a row of spores arranged in a linear series. Resting spores slightly variable in size and shape; germination unknown. Plasmodia one or more in a cell, variable in size and shape; often in the form of a parietal layer around the host protoplasm; schizogony unknown. Zoosporangia and zoospores unknown.

In light of present-day knowledge *Membranosorus* is obviously a doubtful genus which should perhaps be discarded entirely, but until more is known about the Plasmodiophoraceae as a whole its inclusion in any of the other genera is open to serious question. Wernham's observations have shown that the outstanding character described by Ostenfeld and Petersen, namely, hollow single-layered cystosori which line the inner periphery of the host cell and conform to the latter's size and shape, is too variable (fig. 11–18) to be of significant diagnostic value. The incorporation of *Membranosorus* in *Sorosphaera* or *Sorodiscus* is equally questionable if the present-day concepts of these genera are to be maintained, because only occasionally are cystosori in the form of hollow spheres or double-layered discs developed. By the extreme variability of its cystosori this genus resembles perhaps more closely *Ligniera* and *Polymyxa*. Ostenfeld and Petersen regarded it as closely related to *Sorosphaera* and *Tetramyxa*, while Wernham implied that it should be incorporated with *Sorodiscus*. Palm and Burk regarded it as a synonym of *Sorosphaera*. Cook apparently overlooked its existence entirely in his monograph of the Plasmodiophorales.

M. HETERANTHERAE Ostenfeld and Petersen, l.c., fig. 1–6.

Sorodiscus Heteranthera, Wernham, 1935. Mycologia 27: 272. Pl. 17, 18, fig. 1, 2.

Resting spores always aggregated in multiples of four, Globose, ovoid, angular, 3.5–5 μ in diameter, hyaline and buff-brown, with smooth, 0.6–1.0 μ thick walls; apical ring, collar or operculum lacking. Plasmodia oval, ellipsoidal, 8 μ in diameter, or disc-like, flat and often ribbon-shaped, 28–70 μ in length, and encircling the host protoplasm.

Parasitic on *Heteranthera dubia* in Ontario and Quebec, Canada; Vermont and New York, U. S. A., causing marked hypertrophy of adventitious and true roots.

Whether or not the species described by Ostenfeld and Petersen, and Wernham, respectively, are identical is not absolutely certain, but since they have the same habitat and distribution, cause the same symptoms, infect the same tissues of identical hosts, and agree closely as to spore size and shape, they are listed herewith as synonymous. The chief differences so far relate to spore color and variations in the size and shape of the cystosori. Since Ostenfeld's and Petersen's material was very scanty they may have missed most of the variations later observed by Wernham. Likewise, although Wernham never found a single-layered cystosorus completely lining a host cell, his figure 2 shows that the type of sorus described by Ostenfeld and Petersen was often approximated in his material. There is accordingly good evidence that they may have had the same species at hand.

Nothing is known about the method by which this parasite gets into the roots, but entrance appears to

PLATE 9

Membranosorus Heterantherae

(Fig. 1–3, 6, 19 and 20 after Ostenfeld and Petersen; remainder after Wernham; fig. 5 and 18 drawn from photographs.)

Fig. 1. Portion of infected stem of *H. dubia* with 10 swollen and 5 normal roots.

Fig. 2. Early infection stage with small granular parasite attached to host nucleus.

Fig. 3. Young bi- and trinucleate parasites in daughter host cells.

Fig. 4. Young parasite with three nuclei.

Fig. 5. Large parietal plasmodium which almost completely envelopes host protoplasm.

Fig. 6. Large multinucleate plasmodium enveloping the host nucleus.

Fig. 7. Plasmodium with nuclei dividing promitotically.

Fig. 8. Plasmodium in which nuclei are about to undergo reduction division.

Fig. 9. Plasmodium with nuclei which have just undergone reduction division.

Fig. 10. Second meiotic divisions. Plasmodium cleaving into resting spores.

Fig. 11. Cystosorus of young thin-walled resting spores.

Fig. 12. Flat, almost circular cystorus composed of a single layer of resting spores.

Fig. 13. Similar cystosorus with one resting spore projecting beneath.

Fig. 14. Flat, two-layered cystosorus.

Fig. 15. Cystosorus with resting spores in a row.

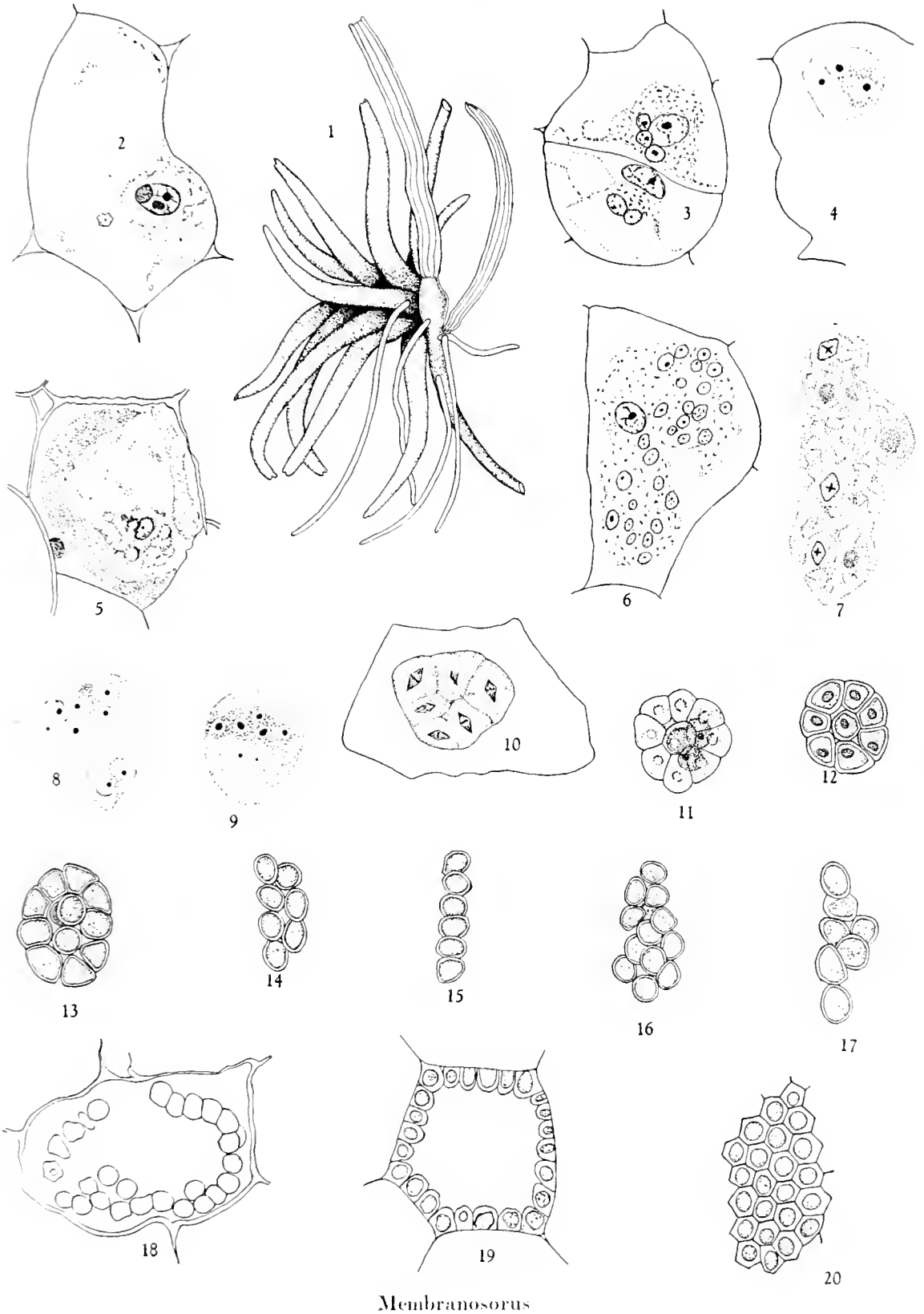
Fig. 16 and 17. Irregular cystosori with loosely attached resting spores.

Fig. 18. Single-layered cystosorus incompletely lining inner periphery of host cell.

Fig. 19. Similar cystosorus completely lining inner periphery of the host cell.

Fig. 20. Surface view of similar cystosorus.

PLATE 9



Membranosorus

be effected at or near the tip. Cells of the periblem are more frequently attacked, and the fungus occurs most abundantly in a region approximately 0.5 cm. back of the root tip. The cells of the central cylinder apparently are never infected. According to Ostenfeld and Petersen, the fungus first appears as a small plastic granular body close by or attached to the host nucleus (fig. 2), and as it grows in size and becomes multinucleate it may envelop the host nucleus and cytoplasm (fig. 3, 5, 6). There is thus a close association of the protoplasts of host and pathogen, and in Ostenfeld and Petersen's drawings it is difficult to distinguish between them. The parasite causes the infected cells to enlarge somewhat but apparently does not stimulate cell division. Figure 3, however, suggests that infected cells may divide, whereby the parasites are passively distributed to the daughter cells.

The mature plasmodia vary greatly in size, and the large extensive ones may often line the inner periphery of the host cell (fig. 5) as in *Tetramyxa*. According to Wernham, cruciform nuclear divisions occur (fig. 7) during the vegetative phase of the plasmodium, and the nuclei undergo meiosis in the first of the two divisions prior to cleavage into resting spores. Although he stated that he had observed numerous meiotic stages and counted four to six pairs of chromosomes, his figures (fig. 8, 9, 10) show nothing of the process.

SPONGOSPORA

Brunchorst, 1887. Bergens Mus. Aarsberet. 1886: 225.

Clathrosorus Ferdinandsen and Winge, 1920. Ann. Bot. 34: 468.

(PLATE 10)

Resting spores usually arranged in hollow or irregularly-channeled spongy, globose balls or cystosori. Resting spores loosely or fairly closely packed together, spherical, oval, pentagonal, hexagonal in optical section with hyaline, yellowish to yellowish-green, smooth, thin or fairly thick walls; each spore producing a single (?) zoospore; such zoospores giving rise to either plasmodia or zoosporangia. Plasmodia usually large, irregular, amoeboid and multinucleate; partly or completely filling the host-cell; forming one or more spore balls. Zoosporangia single or in clusters, variously-shaped. Zoospores from resting spores and zoosporangia similar, small, biflagellate and heterocont; flagella attached at or near anterior end.

Spongospora includes at present three species, one of which is poorly known and doubtful. The type species, *S. subterranea*, has been repeatedly studied morphologically and cytologically, but there is still considerable disagreement concerning some of the critical stages of its life history. As noted in Chapter III, these controversies have centered primarily around the stages at which plasmogamy and karyogamy occur, and the manner by which the parasite invades and spreads in the host tissue. Johnson ('07) described the resting spores as one- to eight-nucleate and giving rise to a corresponding number of zoospores in germination, but subsequent workers including Massee ('08), Kunkel ('15), Cook ('33)

PLATE 10

Spongospora subterranea

(Fig. 7–9, 11 and 23 drawn from photographs)

Fig. 1. Potato with shallow powdery scab lesions.

Fig. 2. Malformed potato with deep cankerous lesions and excrescences.

Fig. 3. Powdery scab galls on roots of potato.

Fig. 4. Enlarged host cell with eight spongy spore balls or cystosori. Osborn, '11.

Fig. 5. Section through a mature cystosorus. Osborn, l.c.

Fig. 6. Uninucleate resting spores. Osborn, l.c.

Fig. 7, 8. Zoospores from germinated resting spores. Ledingham, '35.

Fig. 9. Tetraflagellate zoospore. Ledingham, l.c.

Fig. 10. Dividing amoeba. Massee, '08.

Fig. 11. Irregular zoosporangium. Ledingham, l.c.

Fig. 12. Uninucleate amoebae surrounded by host cytoplasm. Osborn, l.c.

Fig. 13. Host cell with three amoebae and numerous starch grains. Osborn, l.c.

Fig. 14. Dividing host cells with passively distributed amoebae. Osborn, l.c.

Fig. 15. Hypertrophied cells of *S. warsewiczii* which have divided: amoebae aggregated around host nuclei. Melhus, *et al.*, '16.

Fig. 16. Group of infected enlarged tomato cells; typical "Krankheitsherde." Melhus, *et al.*, l.c.

Fig. 17. Infecting plasmodium pushing down between host cells. Kunkel, '15.

Fig. 18. Plasmodium entering host cell and enveloping nucleus. Kunkel, l.c.

Fig. 19. Coalescence of amoebae to form plasmodium; host nucleus enlarged, irregular, and densely chromatic. Osborn, l.c.

Fig. 20. Plasmogamy of two amoebae derived from germinated resting spores. Cook, '33.

Fig. 21. Karyogamy. Cook, l.c.

Fig. 22. Zygote. Cook, l.c.

Fig. 23. Saprophytic plasmodium (?) grown on nutrient agar. Kunkel, l.c.

Fig. 24, 25. Vegetative nuclei degenerating and extruding chromidia into cytoplasm. Osborn, l.c.

Fig. 26. Akaryote and chromidial stage. Osborn, l.c.

Fig. 27. Reconstructed nuclei emerging on new sites. Osborn, l.c.

Fig. 28, 29. Reconstructed nuclei pairing and fusing. Osborn, l.c.

Fig. 30. Late stage in karyogamy. Osborn, l.c.

Fig. 31. Diploid nuclei. Osborn, l.c.

Fig. 32. Late prophase of meiosis with eight chromosomes. Horne, '30.

Fig. 33. Contraction stage and beginning of pairing of homologous chromosomes. Horne, l.c.

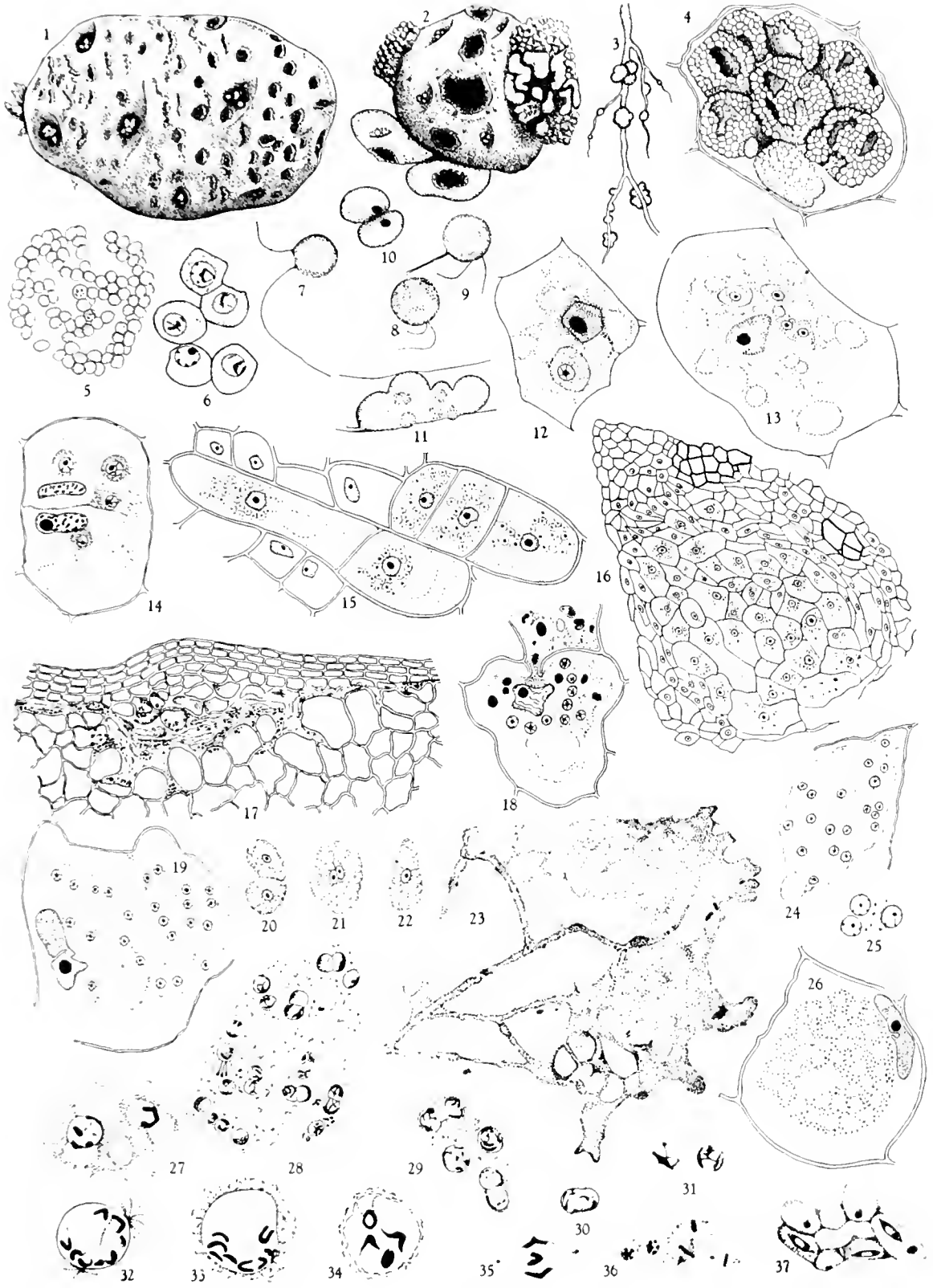
Fig. 34. Diakinesis. Horne, l.c.

Fig. 35. Metaphase, first division, showing three of the chromosome pairs. Horne, l.c.

Fig. 36. Equatorial plate, second division, showing seven chromosomes. Osborn, l.c.

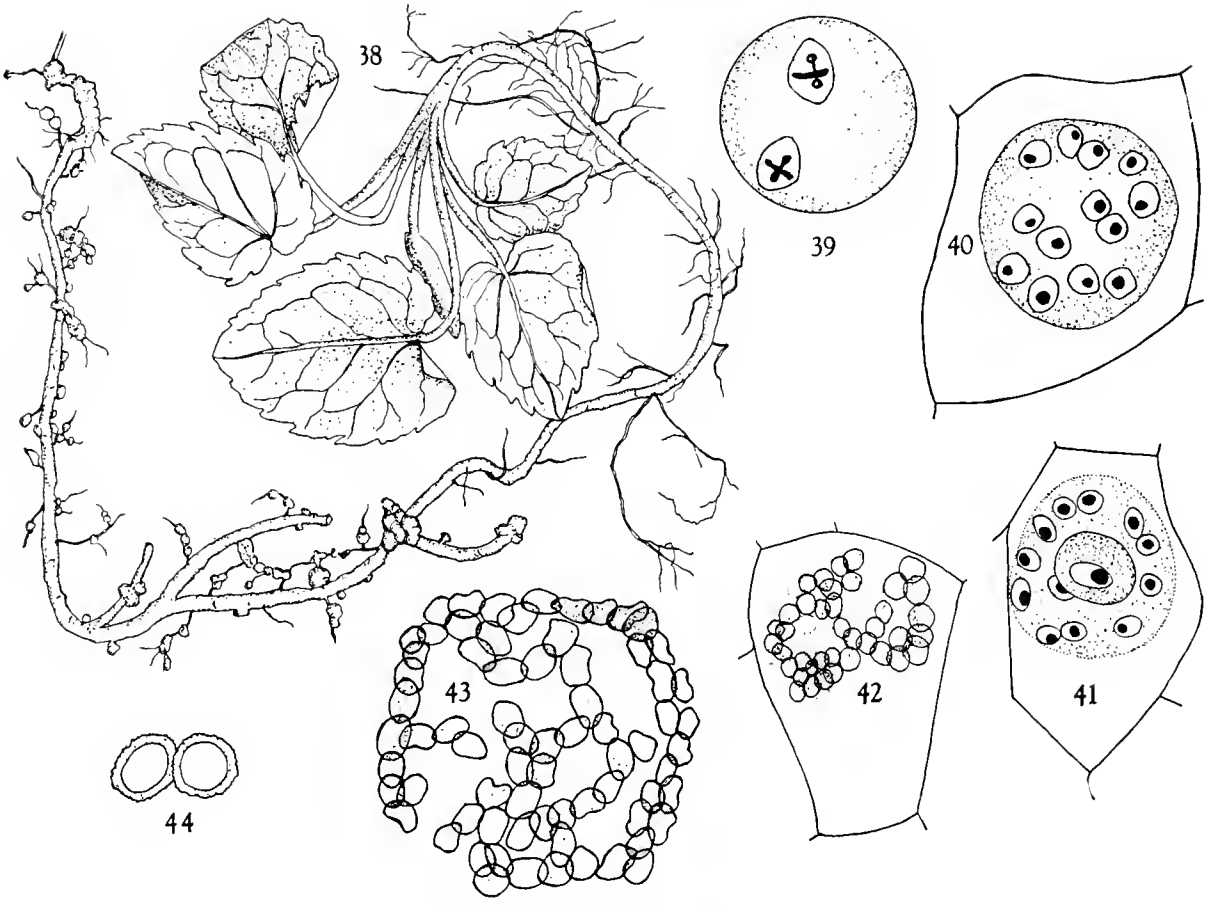
Fig. 37. Anaphase, second division, and cleavage. Osborn, l.c.

PLATE 10



Spongospora

PLATE 10—*Continued*



Spongospora Campanulae

Spongospora Campanulae

Fig. 38. *Campanula rapunculoides* with numerous galls and nodules on roots. Ferdinandsen and Winge, '20.

Fig. 39. Young parasite with nuclei dividing "promitotically." F. and W., l.c.

Fig. 40. Multinucleate plasmodium. F. and W., l.c.

Fig. 41. Plasmodium enveloping host nucleus. F. and W., l.c.

Fig. 42. Irregular cystosorus. F. and W., l.c.

Fig. 43. Section through a cystosorus. F. and W., l.c.

Fig. 44. Section through two resting spores showing finely punctate warty walls. F. and W., l.c.

and Ledingham ('35) observed only one zoospore. Furthermore, all earlier investigators figured and described the zoospores as uniflagellate, but Ledingham demonstrated conclusively that they are biflagellate and heterocont (fig. 7, 8). Whether the flagella are attached at or near the anterior end is not definitely known. Massee, Kunkel, Osborne ('11) and Horne ('30) held that the plasmodium is formed by the fusion of several amoebae (fig. 19), but they were not certain whether such amoebae arise by division of a single amoeba within the infected host cell or are the result of infection by several amoebae. Cook ('33), on the other hand, contended that the plasmodium is initiated by the fusion of gametes in pairs (fig. 20–22).

There is also difference of opinion about infection and spread of parasite in the host tissue. Massee and Cook in particular held that the amoebae have the ability to penetrate the host cell walls and thus pass from cell to cell, spreading the infection. Osborne and Horne, in contrast, maintained that the amoebae are incapable of boring through the walls and are distributed passively and fortuitously by division of the infected cell (fig. 14). Kunkel, however, reported that the primary infection of young tubers as well as secondary infection of tissues around old sori occurs by invasion of the plasmodium. The latter passes through and between the epidermal cells, and once beneath the epidermis it spreades out in all directions (fig. 17). Johnson ('09) believed that the plasmodium may migrate from the diseased parent tubers into the stem and stolons of the young plants, and eventually infect the young tubers. Massee thought that the plasmodium might encyst during the cold winter season and renew its activities when the tubers began to sprout. Wild ('29) considered the

lenticels, instead of the unbroken epidermis, to be the principal avenue of initial infection, with some penetration through wounds.

According to Kunkel, the resting spores germinate readily on nutrient agar and form plasmodia in culture. By weekly transfers, such plasmodia may be kept in an active growing condition on synthetic media for a long time, and under these conditions they are strikingly similar in appearance, shape, behavior, and locomotion to the plasmodia of the Myxomycetes (fig. 23). When subjected to drought they encyst or sclerotize, and if transferred to fresh media the plasmodia may often break up into smaller masses which move away and form stalked fruiting structures like those of *Dictyostelium* and *Polysphondylium*. The erect, single or branched sporophores bear sori of rod-shaped spores like *Dictyostelium*, and in germination give rise to myxamoebae which later aggregate to form pseudoplasmodia. These in turn form sporophores again. Kunkel's observations have not been confirmed, and since species of the Acrasiales occur in soils with *Spongospora* it is not improbable that he may have introduced contaminants of this type in his cultures. It is to be *particularly* noted, however, that the plasmodium which he photographed looks like a true myxomycetous plasmodium. Since it has none of the characteristics of an acrasiaceous pseudoplasmodium in which the individual myxamoebae retain their individuality as cells, it is difficult to conceive how Kunkel got *Dictyostelium*- and *Polysphondylium*-like soroearps from a plasmodium of the type shown in figure 23. His photographs and descriptions suggest that he may have had more than one type of plasmodium at hand. The possibility that *S. spongospora* may form large plasmodia on nutrient agar remains thus to be proven by pure culture studies.

S. SUBTERRANEA (Wall.) Lagerheim, 1891, Journ. Mycol. 7: 104.

Erysibe subterranea Wallroth, 1842a, Linnaea 16: 332. 1842b, Beitr. zur. Bot. 1: 118.

Protomyces Tuber-Solani Martius, 1842a, Die Kartoffelepidemie der letzten Jahre order die Stockfäule und Räude der Kartoffeln, München. 1842b, C. R. Acad. Sci. Paris 15: 314.

Rhizosporium Solani Rabenhorst, 1843, Arch. Pharm. 83: 300, 1844, Krypt. Fl. Deutsch. Oesterr., und der Schweiz 1.

Tuberculina scabies Berkeley, 1846, Jour. Hort. Soc. London 1: 33, Pl. 4, fig. 30–31.

Sorosporium scabies Waldheim, 1877, Aperçu System. des Ustilag. Paris, p. 33.

Spongospora Solani Brunchorst, l.c.

S. scabies Massee, 1908, Jour. Bd. Agric. England 15: 594, Fig. 1–12.

S. subterranea radicicola Blattný, 1935, Rec. Inst. Rech. Agron. Rép. tchécosl. 137: 21.

S. subterranea tubericola Blattný, l.c.

Resting spore clusters or balls oval, elongate, irregular, 19–85 μ in diameter, somewhat spongy with numerous irregular channels. Resting spores loosely packed together, angular, polygonal, spherical, 3.5–4.5 μ, with smooth, thin, yellow to yellowish-green walls. Plasmodia unusually large, up to 70 μ or more in length, amoeboid, irregular; giving rise to one or more spore walls. Zoosporangia single or in clusters, up to a dozen or more in a cell, spherical, oval, elongate, lobed and irregular, hyaline and thin-walled; opening by the rupture of a small papilla which bursts through the host cell wall emitting the zoospores. Zoospores from resting spores and zoosporangia oval, spherical, 2.5–3.5 μ, with two unequal flagella.

Parasitic on *Solanum tuberosum*, *S. warscewiczii*, *S. haematoclodum*, *S. mamosum*, *S. marginatum*, *S. ciliatum*, *S. commersomi*, *S. nigrum*, and *Lycopersicon esculentum*, causing scabby lesions and cankers on the tubers, and galls on the roots and stems. A further account of the distribution and hosts of this species is given in Chapter VI.

Spongospora subterranea causes the disease of potatoes commonly known as powdery or corky scab. While it is chiefly a parsite of the potato, it may also infect close relatives of this host. In extensive inoculation experiments Melhus, *et al.* ('16), found that it will infect all but one of the hosts listed above but not *S. nigrum*, *S. mauritianum*, *S. duplosumaton*, *S. Lobelii*, *S. heteracanthum*, *S. seaforthianum*, *S. lanciniatum*, *S. torvum*, and *Solanum* sp. Ferdinandsen ('23), however, reported that it is transmissible to *S. nigrum* in Denmark. Weber ('22) and Ledingham ('35) also found it on tomatoes in Denmark and Canada, respectively. It has also been reported by Rybakova and Nedoshivinia ('36) on *Ullucus tuberosus* of the Chenopodiaceae in Russia. Truscott ('34) found a *Spongospora*-like organism in the roots of strawberries in Canada, but he was not certain about its identity. Blattný's distinction of two forms of *S. subterranea* on the roots and tubers, respectively, does not seem justified. The two forms may be transferred readily from one organ to another and do not differ greatly in size and color of their spore balls. Blattný, nevertheless, believed that the root form may be mycorrhizal instead of parasitic. Rybakova and Nedoshivinia also described an aberrant form near Moscow which differs from the normal type by the occurrence of its spore balls outside of the host cells. These balls are faintly brown instead of yellowish-green in color, plicate or irregularly crumpled on the surface, and may be aggregated in a common mass. They vary in size from 20–25 μ by 13–19 μ and show no cellular structure. Khrobrykh ('38) experimented with various forms of *S. subterranea* from different potato varieties of different geographical origin and concluded that these forms are not biotypes or geographical races but ecotypes dependent on the host variety, height, and size of the pustules. In this connection it may also be noted that Sharples ('23) described a disease of the petioles and leaf stalks of the cocoanut palm in Malaya which appeared to be associated with a species of *Spongospora*, but he was not certain about the identity of the causal organism. It probably does not relate to *Spongospora* at all.

Spongospora subterranea was the first species of the Plasmodiophoraceae to be reported in the literature, but it was not recognized as a member of this family until about fifty years later. It was first reported, in part by Wallroth in 1842, but he had apparently found it the year before as is indicated in Bartling's (1841) discussion. As is shown in the synonomy above, it was rediscovered a number of times shortly afterwards in connection with other fungi in scabby lesions of potatoes, and included in various genera. It was not until 1886, however, that Brunchorst first recognized it as a species of the Plasmodiophoraceae. For a considerable number of years a long controversy raged about its identity and synonymy, which has been fully reviewed by Lagerheim, Massee, Pethybridge and Cook, and need not be discussed further here.

S. CAMPANULAE (Ferdinandsen and Winge) Cook, 1933. Arch. Protistk. 80: 215.

Clathrosorus Campanulae Ferdinandsen and Winge, l.c. Pl. 21.

Spore clusters or balls irregular, rounded or elongate, 25–50 μ in diameter with large irregular channels. Spores spherical, 4–5.5 μ, oval, irregular, truncate, with fairly thick and slightly verrucose walls. Plasmodia solitary in the host cell and only partly filling it, multinucleate, irregular, 30–50 μ in diameter, when mature; segmenting into resting spores which remain attached in a fairly loose spore ball. Zoosporangia and zoospores unknown.

Parasitic on the roots of *Campanula rapunculoides* in Denmark, causing numerous single or confluent, tubercle-like galls.

This species has been reported but once. Whether it belongs in *Spongospora*, as Cook believed, or represents a new genus is obviously questionable in light of present-day knowledge, but since its spore clusters are reported to be loose, irregular, round or elongate balls (fig. 38), it may be conveniently included here for the time being. It occurs in the cortex of the roots (fig. 38), and although the central cylinder may be distorted, it is never parasitized. The infected cells are only slightly if at all enlarged (fig. 40–42) and do not divide, but the presence of the parasite nonetheless stimulates adjacent healthy cells to divide. The galls are thus almost entirely the result of cell multiplication. The nucleus of the host cell is often enveloped by the parasite (fig. 40), but it does not become greatly enlarged.

According to Ferdinandsen and Winge, meiosis occurs during the last two nuclear divisions in the plasmodium preceding spore ball formation. They did not, however, count the number of chromosomes nor observe plasmogamy and karyogamy, so that their conclusions are not based on adequate observations.

Another species of *Spongospora* was recently reported and described by J. T. Barrett in a brief paper presented before a joint meeting of the American Mycological and Phytopathological Societies at Philadelphia, Pennsylvania, December 30, 1940. Dr. Barrett has not completed his study of this species, but he has graciously allowed me to include a few notes on the essential features of its life cycle. This species parasitizes *Cotula australis* in California and causes conspicuous galls or nodules on its roots. Barrett accordingly named it *S. Cotulae*. In germination each resting spore produces a single zoospore with two unequal flagella as in *S. subterranea*. The zoospores infect the host and eventually give rise to zoosporangia which in turn form motile cells of the same type and character as the zoospores produced by the resting spores. Barrett found fusion stages of the zoospores or gametes from the sporangia in fixed and stained material, but he has not yet observed plasmogamy in living material. Whether or not the sporogenous plasmodium is thus zygotic in origin is uncertain at present. The spore balls or cystosori and resting spores, nevertheless, usually follow the sporangial stage and thus complete the cycle of development.

ADDITIONAL BIBLIOGRAPHY: SPONGOSPORA

Bartling, E. 1841. Versammelung. Deut. Nat. u. Aerzte zu Braunschweig im September 1841. Vieweg und Sohn, 1842.
Ferdinandsen, C. 1923. Tidsskr. f. Landokonomi. 1923.
Horne, A. S. 1930. Ann. Bot. 44: 199.
Johnson, T. 1907. Econ. Proc. Roy. Dublin Soc. 1: 345.
———. 1909. Sci. Proc. Roy. Dublin Soc. n. s. 12: 165.
Khrobrykh, N. D. 1938. Summ. Sci. Res. Inst. pl. protect. for 1936–1938: 27.
Kunkel, L. O. 1915. Jour. Agr. Res. 1: 265.
Ledingham, G. A. 1935. Nature 135: 394.
Massee, G. 1908. Jour. Bd. Agric. England 15: 594.
Melhus, I. E., J. Rosenbaum, and E. S. Schultz. 1916. Jour. Agr. Res. 7: 213.
Osborn, T. G. B. 1911. Ann. Bot. 25: 271, 327.
Rybakova, S., and H. Nedoshivina. 1936. White Russ. Acad. Sci. Inst. Biol. Sci. Minsk 1936: 57.
Sharples, A. 1923. Malayan Agric. Jour. 11: 267.
Truscott, J. H. L. 1934. Canad. Jour. Res. 11: 1.
Weber, A. 1922. Tomatsygdomme. Copenhagen.
Wild, N. 1929. Phytopath. Zeitschr. 1: 367.

LIGNIERA

Maire and Tison, 1911. C. R. Acad. Sci. Paris 152: 206.

(PLATE 11)

Resting spores not consistently aggregated in cystosori of characteristic shape and structure; variously-shaped with relatively thin hyaline or colored, smooth or verrucose walls. Plasmodium relatively small, partly or completely filling the host cell; segmenting into either zoosporangia or one or more cystosori; schizogony reduced or lacking (?). Zoosporangia numerous in a cell and usually grouped together, small and variously-shaped; opening by a rupture of the wall. Zoospores from sporangia pyriform. Germination of resting spores doubtful or unknown at present.

This genus was established by Maire and Tison for all plasmodiophoraceous species characterized by loosely and variously aggregated resting spores, little or no schizogony of the plasmodium, complete development within a single host cell, and which cause no hypertrophy of the host. As such, it is a very questionable genus and should perhaps be discarded, since none of its characters are very distinctive and diagnostic. In the first place the shape and character of the resting spore clusters or cystosori are too variable to be of much generic value. Secondly, none of the species has yet been studied intensively and sufficiently well to determine whether or not schizogony is well developed, reduced, or lacking entirely. Furthermore, it is not certain that the parasite completes its entire life cycle within one host cell. Finally, the presence or absence of host hypertrophy is not a structural or cytological character of the parasite itself, but relates to the reactions of host and pathogen. Even if this latter character were tenable, it would not be diagnostic for the group as a whole, because *L. pilorum*, according to Fron and Gaillat, causes marked local enlargement of the root hairs of *Poa annua*. On the basis of present-day knowledge, *Ligniera* appears thus to be scarcely more than a convenient dumping ground for species which cause little or no hypertrophy. Further intensive studies, however, may reveal a more fundamental basis of distinction.

The pyriform uninucleate zoospores of *Ligniera* have been described by Cook as anteriorly uniflagellate (fig. 1), but more careful study will doubtless show them to be biflagellate and heterocont as in *Plasmodiophora*, *Polymyxa*, *Spongospora*, and *Octomyxa*.[3] After penetrating root hairs and epidermal cells, they may become flagellate and actively motile again in the host protoplasm (fig. 2B), according to Cook. The flagellum soon disappears, however, and the parasite becomes amoeboid in shape and motion (fig. 3). Nuclear divisions occur as the amoebae increase in size (fig. 4, 8, 9), until a multinucleate plasmodium is formed. One or more amoebae and plasmodia may be present in a host cell, but so far no conclusive evidence has been presented to show that they coalesce to form a larger structure. As noted before Cook ('33) reported that the zoospores are isogametes which fuse in pairs to form zygotes, but his evidence of plasmogamy or karyogamy is not very conclusive.

Host cells usually contain only one plasmodium, which fills them almost completely (fig. 12). Very little is known about the feeding habits of the intramatrical plasmodia. They apparently absorb the host cytoplasm, envelop the nucleus, and lead to the disappearance of the starch grains, so that the infected regions of the roots appear quite pale in color. Maire and Tison ('11), however, reported that the plasmodium is capable of engulfing large food particles, and figured a young parasite with five engulfed algal cells (fig. 5).

Whether or not schizogony occurs in *Ligniera* is still questionable. Schwartz ('10) reported that the young plurinucleate plasmodia of *L. Juncі* function as schizonts, splitting off smaller uni- or multinucleate daughter segments. Maire and Tison ('11b), on the other hand, believed that schizogony may be lacking entirely or is reduced to the formation of two or three meronts (fig. 10). Subsequent workers also have doubted its occurrence or claimed that, if present at all, it takes place only in the young developmental stages.

With the doubtful exception of *L. pilorum* (fig. 25, 26, 38), species of *Ligniera* have no noticeable effect on the host tissues, according to most accounts in the literature. No galls are produced, and the infested cells are not markedly enlarged or stimulated to divide. Schwartz ('11), however, reported that parasitized cells of *Poa annua* are often considerably elongated, due either to the failure of transverse wall development or the absorbtion of such walls by the parasite. Subsequent workers, however, have not confirmed these observations. The relation between the protoplasts of the host and pathogen appears to be very intimate, and no marked antagonism is exhibited, according to the figures and descriptions in the literature. Schwartz reported that the amoebae are strongly attracted to the host nucleus and migrate toward it as soon as they have entered the cell (fig. 6, 7) like in *S. subterranea*. The amoebae shown in these figures, however, look like nuclei of the parasite, which suggests that the host nucleus in these instances might possibly be enveloped by a multinucleate plasmodium. Complete envelopment of the host nucleus and a mixing of the two protoplasts has also been shown in root hairs of *Juncus articulatus* (Schwartz, '10, fig. 5). Figure 37 shows a host nucleus inside a mature cystosorus. The nucleus of infected cells shows little or no enlargement, and no conspicuous signs of degeneration are evident until the parasite is almost mature. Maire and Tison's figures (14, 37) suggest that as degeneration progresses the nucleus becomes more chromatic and densely stainable.

According to Cook ('26, '28) the mature plasmodium may form either zoosporangia or cystosori. In the former event uninucleate segments (fig. 20) are delimited by progressive cleavage, and their nucleus divides twice and occasionally three times. Cook claimed that the first division is heterotypic and the second homeotypic, but his evidence is not at all conclusive (see Chap. III). In resting spore development uninucleate amoebae may sometimes undergo two nuclear divisions (fig. 22) and form tetrads of resting spores, according to Maire and Tison ('11b). It is not improbable, however, that their figures may relate instead to sporangia and zoospore development. In larger plasmodia, the reconstructed nuclei following the so-called "akaryote" stage also divide twice. Following these divisions the plasmodium cleaves into uninucleate segments, which round up,

[3] This is suggested by Barrett's discovery of biflagellate heterocont zoospores in *Rhizomyxa hypogea* which is regarded as a combination of *Ligniera* sp. and another fungus species.

become invested with a wall (fig. 31), and mature into resting spores. These spores usually remain attached to each other and form cystosori of variable sizes and shapes (fig. 28–40) in accordance usually with the size of the plasmodium and the shape of the host cell.

L. JUNCI (Schwartz) Maire and Tison, l.c.

Sorosphaera Junci Schwartz, 1910. Ann. Bot. 24: 512. Pl. 40.

S. graminis Schwartz, 1911. *Ibid.* 25: 791. Pl. 61.

L. graminis (Schwartz) Winge, 1913. Ark. f. Bot. 12, no. 9: 15.

L. radicalis Maire and Tison, l.c.; 1911. Ann. Mycol. 9: 233. Pl. 11, fig. 24–38.

L. Bellidis Schwartz, 1914. Ann. Bot. 28: 232. Pl. 12, fig. 7–8.

L. Menthae Schwartz, l.c. Pl. 12, fig. 1–6.

L. Alismatis Schwartz, l.c., p. 233.

Resting spores rarely in tetrads, sometimes end to end in a linear series; more often in irregular masses, solid or hollow, flat, globose or ellipsoidal, cylindrical and elongate cystosori. Resting spores spherical oval, angular and polyhedral when compressed together, 4–7 μ in diameter, with relatively thin hyaline smooth walls; apparently giving rise to zoospores which infect the host cell. Plasmodium partly or completely filling the host cell; segmenting into either zoosporangia or one or more masses of resting spores, schizogony questionable or reduced. Zoosporangia oval, subglobose, spherical, angular and polyhedral, 15–20 μ in diameter, with thin hyaline smooth walls; method of dehiscence unknown. Zoospores from sporangia 4 to 8 in number, pyriform, 3.5×4.5 μ.

Parasitic in the root hairs and roots of *Juncus articulatus, J. obtusiflorus, J. bufonius, J. lamprocarpus, Poa annua, Bellis perennis, Mentha pulegium, Alisma Plantago, Callitriche stagnalis, Potomgeton natans, Polygonum Hydropiper, Iris pseudocorus, Ranunculus circinatus, R. aquatilis, Plantago major, Cerastium vulgatum, Veronica Beccabunga,* and *Chrysanthemum leucanthemum* in England (Schwartz, '10, '11, '14; Cook, '26, '27, '28, '33); *Callitriche stagnalis* and *Poa annua* in France (Maire and Tison, l.c., '11b); *Alisma Plantago* in New York, U. S. A. (Karling, '34).

Cook ('26) made extensive cross inoculation experiments involving 165 individuals of different species, 151 of which became infected with *L. Junci* after four months. These plants included the hosts of Schwartz's *L. graminis, L. Bellidis, L. Menthae,* and *L. Alismantis,* and since Cook found no essential differences between these *Ligniera* species and *L. Junci,* he concluded that they are identical. The species which he found in *Callitriche stagnalis* was likewise capable of infecting the same hosts; and for this reason he ('33) later concluded that *L. radicalis* described by Maire and Tison in *C. stagnalis* in France is also identical to *L. Junci.* The resting spores of *L. radicalis,* however, are only 4–5 μ in diameter, while those of *L. Junci* range from 5 to 7 μ. This difference is not very great and may not

PLATE 11

Ligniera

Fig. 1. Zoospore highly magnified (*L. Junci;* Cook, '28).

Fig. 2a. Zoospore outside of root hair; 2b, after entering host cell (Cook, '26).

Fig. 3, 4. Developmental stages of amoebae and young plasmodium (*L. graminis;* Schwartz, '11).

Fig. 5. Young thallus with five engulfed algal cells (*L. radicalis;* Maire and Tison, '11).

Fig. 6. Two amoebae approaching a central host nucleus (*L. graminis;* Schwartz, '11).

Fig. 7. Amoebae clustered around host nucleus (*L. graminis* Schwartz, l.c.).

Fig. 8. Young amoeboid plasmodium (*L. graminis;* Schwartz, l.c.).

Fig. 9. Young plasmodium in root hair; nuclei with large karyosome and abundant chromatin (*L. Junci;* Cook, '26).

Fig. 10. Possibly schizogony of plasmodium (*L. radicalis;* Maire and Tison, l.c.).

Fig. 11. "Promitosis" of vegetative nuclei (*L. graminis;* Schwartz, l.c.).

Fig. 12. Single large plasmodium in a host cell. Nuclei entering akaryote stage (*L. graminis;* Schwartz, l.c.).

Fig. 13. Akaryote stage; nuclei appear as clear spaces (*L. graminis;* Schwartz, l.c.).

Fig. 14. Akaryote state; cytoplasm with numerous chromatic granules; host nucleus densely chromatic in base of cell (*L. radicalis;* Maire and Tison, l.c.).

Fig. 15–18. Successive stages of extrusion of chromatin from the nucleus (*L. Junci;* Cook, '33).

Fig. 19. Prophase of heterotypic division (?) in a reconstructed nucleus (*L. Junci;* Cook, '28).

Fig. 20. Cleavage of plasmodium into zoosporangia; the two large mitotic figures in upper left segments are equatorial plate stages of the first heterotypic division (?); the remainder relate to homeotypic division (?) (*L. Junci;* Cook, '26).

Fig. 21. Cleavage into zoospores (*L. Junci:* Cook, '28).

Fig. 22. Second mitoses prior to resting spore formation (*L. radicalis;* Maire and Tison, l.c.). May possibly relate to sporangia and zoospore development like in figure 20.

Fig. 23. Zoosporangia (*L. Junci;* Cook, '28).

Fig. 24. Empty zoosporangia (*L. Junci;* Cook, '28).

Fig. 25. Plasmodium in swollen root hair tip (*L. pilorum;* Fron and Gaillat, l.c.).

Fig. 27. Cluster of empty resting spores in swollen root hair tip (*L. graminis;* Schwartz, l.c.).

Fig. 28, 29. Small groups of resting spores (*L. Menthae;* Schwartz, '14).

Fig. 30, 31. Types of resting spore clusters (*L. graminis;* Schwartz, '11).

Fig. 32. Single resting spore (*L. Junci;* Cook, '28).

Fig. 33. Resting spore ball filling host cell (*L. Isoetes;* Palm, '18).

Fig. 34. Cross section of a similar hollow resting spore ball (*L. Isoetes;* Palm, l.c.).

Fig. 35. Loose chain of resting spores (*L. Isoetes;* Palm, l.c.).

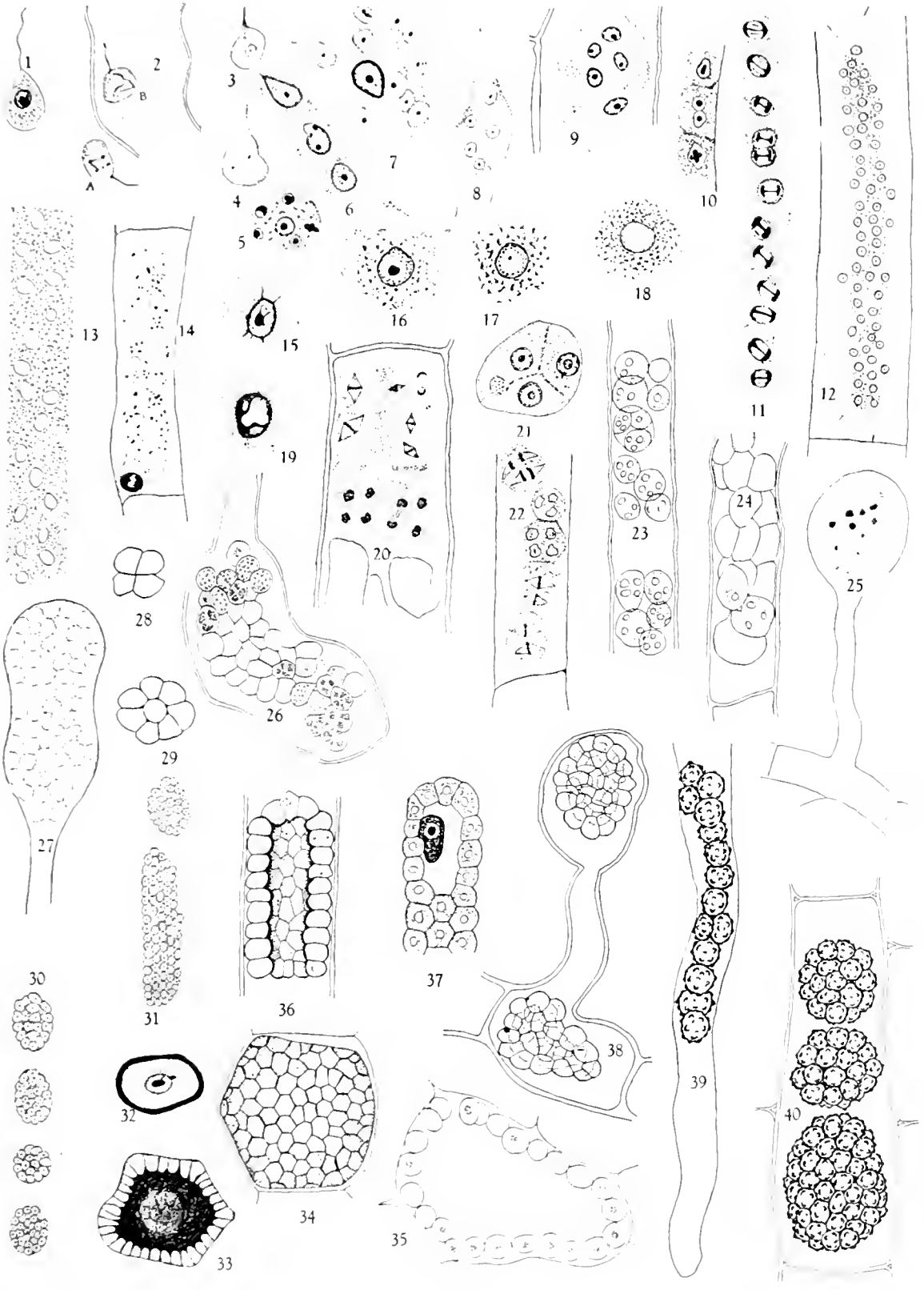
Fig. 36. Longitudinal section of hollow cylindrical resting spore cluster (*L. radicalis;* Maire and Tison, l.c.).

Fig. 37. Cluster of resting spores with host nucleus inside (*L. radicalis;* Maire and Tison, l.c.).

Fig. 38. Resting spore clusters of *L. pilorum* in swollen base and tip of root hair (Fron and Gaillat, l.c.).

Fig. 39, 40. Types of resting spore clusters (*L. verrucosa;* Maire and Tison, l.c.).

PLATE 11



Lignicra

be sufficient reason for separating the two species. Light appears to be the dominant factor in infection. No infection occurs in roots exposed to light even when other environmental conditions are optimum, according to Cook ('27).

In this connection it may be noted that Hildebrand ('34, Pl. 1, fig. 5) observed cystosori of indefinite size and shape in diseased rootlets of strawberries in Canada. Whether or not these resting spores relate to *Ligniera* or another genus is uncertain at present, since Hildebrand made no further study of the organism in question.

L. PILORUM Fron and Gaillat, 1925. Bull. Soc. Mycol. France 41:390. Pl. 10.

Resting spores aggregated into globose and irregular clusters or cystosori, or lying end to end in a linear series; oval, spherical, 4–6 μ, or angular and polyhedral when compressed together, with thin hyaline smooth walls. Plasmodium filling the enlarged base or tip of the host cell; schizogony questionable; plasmodium apparently segmenting into either zoosporangia or resting spores. Zoosporangia (?) oval, spherical, angular and compressed, 4–6 μ (?) with thin, smooth hyaline walls, opening by the rupture of a thin localized area. Zoospores small, pyriform, up to 1 μ (?) in diameter; flagellum of same length as spore body.

Parasitic in the root hairs of *Poa annua* in France, causing marked local hypertrophy (?).

Fron and Gaillat's drawings and descriptions of the developmental stages of this species are very brief and inadequate, and it is not clear whether the zoospores arise from germinating resting spores or zoosporangia like those described by Cook ('26) for *L. Junci*. The latter view seems more plausible because figures 7 and 8 by Fron and Gaillat show what appears to be several zoospore initials within a single unit of the aggregate; whereas the resting spores of most plasmodiophoraceous species are now rather generally believed to form but one zoospore apiece. If Fron and Gaillat's measurements are correct, this species is characterized by unusually small zoospores. Cook ('26, '33) regarded *L. pilorum* as synonymous with *L. Junci*, because it also occurs in *Poa annua* and agrees with the latter in life cycle and resting spore size. The chief differences are zoospore size and the fact that *L. pilorum* causes hypertrophy of the host cell, according to Fron and Gaillat. Cook maintained that such hypertrophy is not due to the stimulus of the parasite but that *L. pilorum* may fortuitously infect root hairs which are already swollen. In further support of his belief that the two species are identical, he pointed out that *L. Junci* occasionally attacks swollen hairs also. Schwartz ('14) likewise observed that normally swollen root hairs (fig. 27) may sometimes become infected with *L. Junci*. It seems almost too accidental, however, that all the infected root hairs shown in Fron and Gaillat's (fig. 1) are greatly enlarged at the tip. Nevertheless, it is not entirely improbable that *L. Junci* and *L. pilorum* are identical, but until more is known about the latter species and host range, its identity and validity will remain questionable.

L. VERRUCOSA Maire and Tison, l.c. 1911, Ann. Mycol. 9:235. Pl. 11, fig. 39–41; pl. 12, fig. 42–46.

Resting spores occasionally aggregated in a linear series, more often in globular, ellipsoidal solid, rarely flattened, and disc-shaped, or hollow balls; resting spores oval, spherical, 4–5 μ in diameter, angular and polyhedral when compressed, with fairly thin, hyaline verrucose walls. Apparently giving to rise to zoospores in germination, which infect the host. Plasmodium partly or completely filling the host cell; giving rise to one or more cystosori; schizogony reduced or lacking entirely. Zoosporangia and zoospores unknown.

Parasitic in the root hairs and roots of *Veronica arvensis* (Maire and Tison, l.c.), *Beta vulgaris, Chenopodium album, Bromus sp.,* and *Festuca sp.* in France (Guyot, '27), without causing hypertrophy of the host tissue.

This species is imperfectly known at present, and many of its critical stages remain to be studied. As is sometimes true of the previous species, the shape and structure of the cystosori depend to a large degree on the character of the host cell. When the cystosori occur in elongate narrow root hairs, they may consist of a linear series of resting spores, but if they develop in the cortical parenchyma cells, they usually have the form of more or less solid, globose and ellipsoidal balls.

Guyot regarded this species as a variety of *L. Junci*, because the characters of his specimens of *L. verrucosa* seemed to merge imperceptibly with those of *L. Junci*. Cook ('33), after examining material submitted by Guyot, and Maire and Tison found no difficulty in distinguishing *L. Junci* and *L. verrucosa*. However, the warts on Guyot's specimens were found to be much less pronounced than those on Maire and Tison's material. Palm and Burk did not regard the presence of warts as a specific character, since in a single species of *Sorosphaera* on *Veronica americana* they found both smooth and warty spores with all degrees of gradation between the two types. Hence, they regarded *L. verrucosa* as identical to *L. radicalis* or *L. Junci*. The development of smooth and warty spores in a single species is not at all uncommon among fungi, and Palm and Burk were probably right in their conclusions. More intensive study of the development, variations, and host range of *L. verrucosa* is, however, essential.

L. ISOETES Palm, 1918. Svenska Bot. Tidsskr. 12:228. Fig. 1–3.

Resting spores sometimes in more or less loosely aggregated clusters, more often in hollow balls which fill the host cell and conform with the latter's shape. Resting spores oval, almost spherical, angular and polyhedral when compressed, 5×6–8 μ, with thin, smooth brownish-colored walls. Plasmodia partly or

completely filling the host cell. Zoosporangia and zoospores unknown.

Parasitic in the leaves and roots of *Isoetes lacustris* in Sweden (Palm, l.c.) and New Jersey, U.S.A. (Karling, '34), causing large, dark spots in the host tissue but no hypertrophy.

This species is so little known at present that its identity is very doubtful. As Cook pointed out, it may well be identical to *L. Junci*, but some of the resting spore clusters figured by Palm are strikingly like those of species of *Sorosphaera* and *Membranosorus*. The present writer's observations on this species in 1934 were very limited, and since then he has not added any further data on its structure and development.

L. VASCULARUM (Matz) M. T. Cook ('29) does not appear to be a valid species. See *Plasmodiophora vascularum*.

ADDITIONAL BIBLIOGRAPHY: *Ligniera*

Cook, W. R. J. 1926. Trans. Brit. Mycol. Soc. 11: 196. 1927, *Ibid.* 12: 282.
———. 1928a. Bull. Soc. Mycol. France 44: 105.
———. 1928b. Ann. Bot. 42: 347.
———. 1932a. Hong Kong Nat. Suppl. No. 1: 29.
———. 1932b. Jour. Dept. Agr. Porto Rico 16: 409.
———. 1933. Glamoran County Hist. Nat. Hist. 1: 213.
———. 1934. Watson's Microscope Record —: 3, 9.
Guyot, A. L. 1927. Rev. path. Ent. Agr. 14: 176.
Hildebrand, A. A. 1934. Canadian Jour. Res. 11: 21.
Karling, J. S. 1934. Torreya 34: 13.
Palm, B. T., and M. Burk. 1933. Arch. Protistk 79: 263.
Smith, A. L., and J. Ramsbottom. 1917. Trans. Brit. Mycol. Soc. 6: 231.

POLYMYXA

Ledingham, 1933. Phytopath. 23: 20.

(PLATE 12, FIGS. 1–22)

Cystosori or resting spore clusters indefinite in size and shape, without a common membrane; formed by cleavage of a naked multinucleate plasmodium. Resting spores few or numerous, variable in shape. Zoosporangia conjoined in a more or less linear series; formed by the septation of an elongate, lobed, irregular and tubular thallus, which may extend through one or more host cells; exit tubes one or more, variable in length, and septate. Zoospores from resting spores and zoosporangia biflagellate and heterocont.

Polymyxa is a monotypic genus, and like *Spongospora, Ligniera, Plasmodiophora*, etc., includes zoosporangial and naked plasmodial stages in its life cycle. The zoospores apparently penetrate the host cell wall directly (fig. 6, 7) and lie in the host protoplasm as small globose bodies. As is shown in figure 8, they soon begin to increase in size and elongate, and as growth continues they become lobed (fig. 9, 10), branched, irregular, and tubular, and sometimes extend through the host walls into adjacent cells. In this manner large septate thalli are developed which are completely surrounded from the beginning by a thin hyaline wall and closely resemble the thalli of *Septolpidium, Lagenidium, Myzocytium*, etc. The segments of the thalli become zoosporangia (fig. 11) and develop one or more septate exit tubes of variable length. The protoplasm then undergoes cleavage into zoospores which exhibit considerable movement within the zoosporangia before emerging. When mature, they emerge fully formed in succession from the exit tubes, become amoeboid for a few moments, and swim away.

The zoospores are pyriform and ovate in shape, usually uninucleate, and possess a long and short flagellum attached to the nucleus near the anterior end of the spore body (fig. 1–4). A few binucleate zoospores with four flagella have been found, but Ledingham was not certain whether they were the result of unequal cleavage or fusion of two biflagellate spores. During active swimming the flagella may extend out in front, but the zoospores are usually propelled from behind, according to Ledingham. They rotate on their axes or roll over in swimming, and their motility appears to be somewhat slower than that of most chytrid zoospores. After an active swimming stage of about two to three hours, the flagella disappear, and the zoospores become amoeboid again (fig. 5). In this state they move about by pseudopodia, and may often engulf small food particles or objects. These amoeboid zoospores may penetrate and reinfect host cells, but it is not certain from Ledingham's account whether they give rise to another crop of zoosporangia or develop into large multinucleate plasmodia. Apparently they possess both potentialities.

The thallus from which the resting spore cluster is formed begins in the host cell as a naked uninucleate amoeba (fig. 12), and at no time does it possess a membrane or wall. As it increases in size, repeated nuclear divisions occur, and a multinucleate plasmodium is soon formed. Its shape changes constantly as it moves about in the host cell. It may frequently be long and tenuous, extending the full length of the host cell, or form a crescentric mass around the host nucleus with long thread-like, radiating pseudopodia. These pseudopodia are later retracted as the protoplasm becomes denser, and the plasmodium may then segment into a number of portions or meronts (fig. 18) which often lie in rows or closely packed groups in the tracheal and cortical cells. Occasionally fusion of several separate plasmodia may occur in the same host cell (fig. 14), but Ledingham was not certain whether these were thalli of opposite sex or merely meronts derived by division of a common schizont. He was unable to count the chromosomes in the nuclear divisions preceding resting spore formation and accordingly found no evidence of meiosis at this stage.

In the early stages of growth the plasmodium is very vacuolate, but as development proceeds the vacuoles decrease in size. As a result the thallus becomes more granular and refringent in texture and

appearance. Very shortly thereafter progressive cleavage (fig. 15) begins and delimits the individual resting spores which remain in continuity as clusters (fig. 16). The resting spores are usually uninucleate, and in germination each gives rise to one zoospore (fig. 21) which is similar in size, shape and structure to those formed in the zoosporangia.

Polymyxa is strikingly similar to *Ligniera* in size and shape of its cystosori, size, shape and arrangement of resting spores, and by its failure to cause hypertrophy of the host. It differs primarily by the shape and size of its zoosporangia, but this difference may be only specific instead of generic. The lack of schizogony in *Polymyxa,* which Ledingham cited as an additional difference, may not prove to be of great significance, since its presence in *Ligniera* also is still quite doubtful.

P. GRAMINIS Ledingham, l.c.; 1939. Canadian Jour. Res. C, 17: 50. Pl. 1–3.

Resting spores spherical, polygonal, 4–7 μ; contents hyaline and refringent; inner wall hyaline, outer wall smooth, yellowish-brown. Zoosporangia lobed, oval, uteriform and irregular; exit tubes of variable length. Zoospores broadly spindle-shaped, ovate, pyriform, 4–5 μ in diameter; flagella 16–20 μ and 4–5 μ long respectively; zoospores emerging fully formed and swimming directly away; rolling over and over while in motion, intermittently amoeboid. Plasmodium variable in size and shape, often filling host cell, amoeboid in shape and motion.

Parasitic in the roots of *Triticum aestivum, T. durum, Hordeum vulgare,* and *Secale cereale* in Canada.

Ledingham found similar resting spores in roots of species of *Agropyron, Scolochloa, Rumex,* and *Impatiens,* but since no sporangia were present, he was uncertain about the relation of this fungus to *P. graminis.* He reported further that species of *Juncus* and *Poa* in which *Ligniera* parasites occur failed to become infected when grown with parasitized wheat roots. He accordingly regarded *P. graminis* as an obligate parasite. Truscott ('34) also reported what he believed to be *P. graminis* in roots of strawberries in Canada.

DOUBTFUL GENERA

Under this title are presented four genera about which there has been much disagreement and controversy. *Rhizomyxa, Sorolpidium,* and *Anisomyxa* occur in the roots of higher plants, do not cause hypertrophy, and form cystosori of indefinite size and shape. In these characters they resemble *Ligniera* and are regarded by most recent investigators as synonyms of this genus. *Trematophlyctis,* however, parasitizes leaves and petioles and causes marked hypertrophy. There is very little evidence in Patouillard's account to warrant inclusion of this genus in the Plasmodiophoraceae, but inasmuch as Palm subsequently reported it to be "an undoubted member of this family" a brief description of its life cycle is herewith presented. The present writer is in agreement with Maire and Tison's, Guyot's, Cook's, and Barrett's interpretation of *Rhizomyxa, Sorolpidium* and *Anisomyxa,* but further intensive study may possibly reveal distinct generic differences. For this reason they are described and figured separately, so that research students may judge independently the evidence of identity and relationships of these genera.

RHIZOMYXA

Borzi, 1884. Rhizomyxa, nuova ficomicete, Messina.

(PLATE 12, FIG. 23–30)

Plasmodia partly or completely filling host cell, variable in size and shape; forming at maturity either single large zoosporangia or sporangiosori composed of small zoosporangia, or cystosori (?). Cystosori and resting spores poorly known or doubtful.

R. HYPOGEAE Borzi, l.c., pl. 1, 2.

Sporangiosori one or more in a cell, spherical, ovoid, irregular, elongate, sometimes made up of lin-

PLATE 12

Polymyxa graminis

(All figures, except 20 and 21, after Ledingham; fig. 6, 7, 8, 17, 19 and 22 drawn from photographs.)

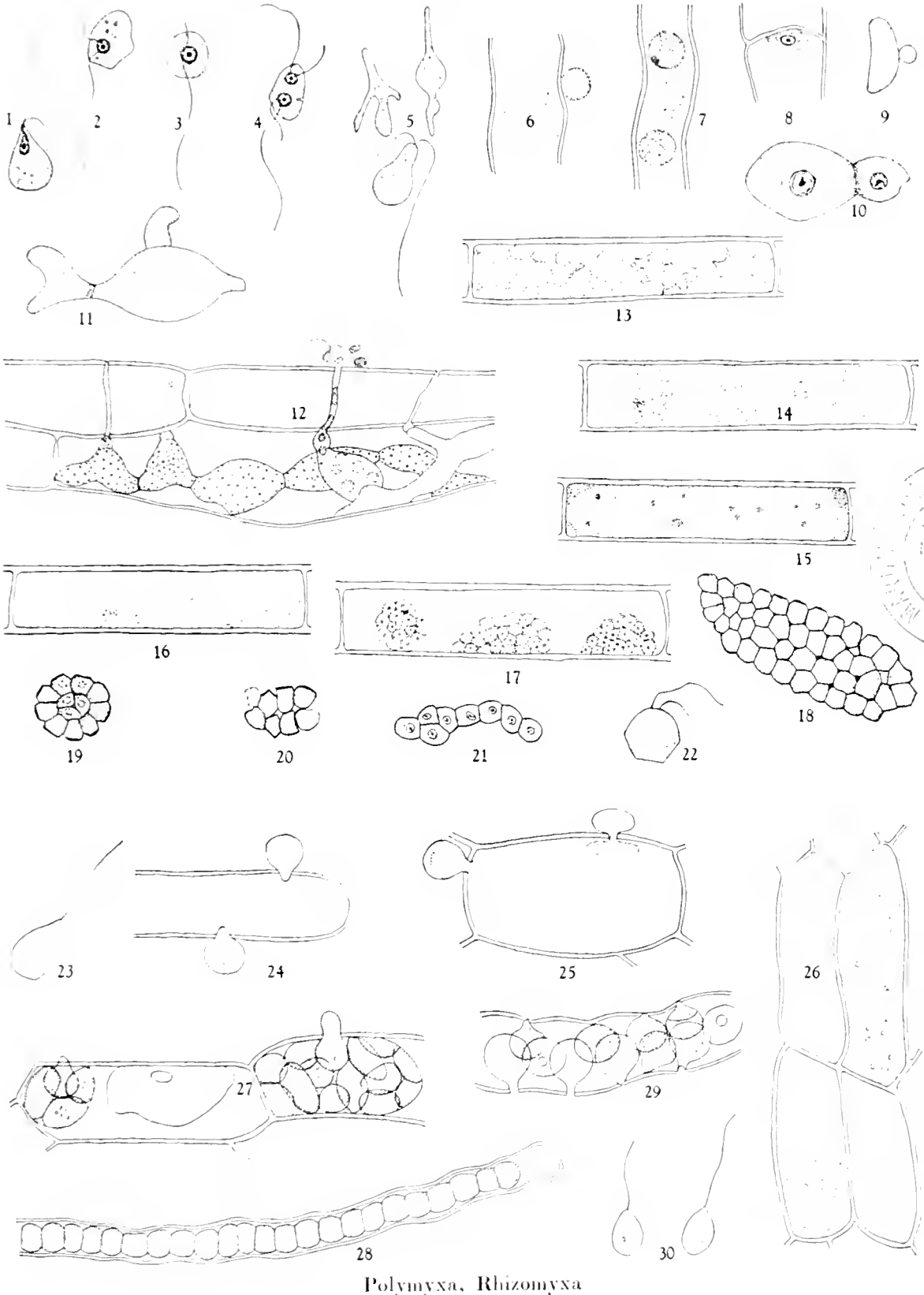
Fig. 1–3. Biflagellate heterocont uninucleate zoospores.
Fig. 4. Large binucleate tetraflagellate zoospore.
Fig. 5. Living, amoeboid zoospores.
Fig. 6. Zoospore on surface of root hair.
Fig. 7. Zoospores after entrance into root hair.
Fig. 8. Stained zoospore inside of cortical cell shortly after penetration.
Fig. 9–11. Stages in development of zoosporangial thalli.
Fig. 12. Mature zoosporangia with exit tubes passing through adjacent cells.
Fig. 13. Naked myxamoeba during period of active growth.
Fig. 14. Segments or meronts formed by division of plasmodia.
Fig. 15. Same cell as in fig. 13 after meronts have coalesced to form a large plasmodium.
Fig. 16. Plasmodium just prior to cleavage into incipient cystosori.
Fig. 17. Cleavage of plasmodium into cystosori.
Fig. 18–21. Variations of cystosori. (Fig. 20 and 21 drawn from material presented by Ledingham.)
Fig. 22. Zoospore from resting spore stained *in toto.*

Rhizomyxa hypogeae

(All figures after Borzi)

Fig. 23. Zoospore.
Fig. 24, 25. Germination and infection stages.
Fig. 26. Plasmodia within host cells.
Fig. 27, 28. Sporangiosori and sporangia.
Fig. 29. Emergence of zoospores.
Fig. 30. Zoospores from sporangia.

PLATE 12



LIBRARY

Polymyxa, Rhizomyxa

ear rows of sporangia. Large single zoosporangia spherical, oval and elongate, producing up to 24 zoospores; zoosporangia in sporangiosorus usually small, spherical and ovoid, 5–6 μ in diameter with thin, hyaline, smooth walls and a short exit pupilla; forming usually 1–2 zoospores which emerge fully formed and swim directly away. Zoospores pyriform, egg-shaped and small; flagellum 10–15 μ. Cystosori (?) of indefinite size and shape, 20–60 μ in diameter. Resting spores oval and spherical, 8 μ; germination unknown.

Parasitic in the cortical cells of young roots and in root hairs of *Agrostis alba, Aira Cupaniana, Briza maxima. Poa annua, Setaria glomerata, Stellaria media, Silene colorata, Capsella bursa pastoris, Biscutella lyrata, Delphinium longipes, Lotus ornithopodioides, Medicago tribuloides, Trifolium resupinatum, Anagallis arvensis, Borrago officinalis, Dinaria reflexa, Bartsia Trixago, Lamium amplexicaule, Fedia cornucopiae, Campanula dichotoma, Calendula arvensis,* and *Erigeron canadensis* in Italy (Borzi, l.c.); *Triglochin palustre, Juncus Gerardi* and *Ranunculus sceleratus* in Germany (Fischer, '92); in numerous species of grasses in Belgium (De Wildeman, '93); and *Stellaria media* in the U.S.A. (Barrett, '35), without causing hypertrophy of the host cells.

The above diagnoses differ somewhat from those given by Borzi, since it is now generally agreed that the antheridia and oogonia which he described relate to another organism. The plasmodia (fig. 26), sporangiosori, (fig. 27, 28) and zoospores (fig. 23, 36), however, doubtless relate to a species of the Plasmodiophoraceae. The identity and relationship of *R. hypogeae* have been the subject of lengthy discussion and speculation since the time of its discovery in 1884. Borzi was uncertain of its taxonomic position, but in 1892 Fischer pointed out that it is probably a combination of two or more fungi, *Olpidium*- and *Olpidiopsis*-like species and a *Woronina*-like fungus. Because of the presence of sporangiosori and cystosori, he placed it next to *Woronina* in the Synchytriaceae. A year later de Wildeman found it in the roots of various grasses in Belgium and from a study of the plasmodia and sporangiosori came to the same conclusions as Fischer concerning its identity and relationship. Schroeter ('97), however, emphasized the heterogamous type of sexual reproduction described by Borzi and included *R. hypogeae* in the Lagenidiaceae. In 1911 Maire and Tison pointed out the similarities between certain of its stages and those of their new genus *Ligniera,* and suggested that *R. hypogeae* is probably a combination of *L. verrucosa* and another fungus. This viewpoint was subsequently supported by Guyot ('27), and Cook ('33). Minden ('11) excluded the sexual phase as relating probably to a species of *Myzocytium,* included the remaining stages of Borzi's fungus in the Synchytriaceae, and pointed out that it is very similar to *Woronina* except for its anteriorly uniflagellate zoospores. Fitzpatrick ('30) believed that the large zoosporangia relate to *Olpidium.* In more recent years Barrett has thrown further light on the identity of Borzi's fungus. He found a species of *Ligniera* in roots of *Stellaria media* which was frequently associated with antheridia, oogonia and oospores of the type described by Borzi. The zoosporangia of what he called *Ligniera* sp., are comparatively large and isolated with fairly broad exit tubes and form anteriorly biflagellate zoospores as in *Plasmodiophora, Polymyxa, Octomyxa,* etc. Antheridia and oogonia may occur in association with *Ligniera* or are isolated in separate roots, and Barrett thus concluded that the two are unrelated. In his opinion Borzi's fungus is a combination of *Ligniera* and a species of the Lagenidiales (Ancylistales). Barrett's observation is particularly noteworthy in that it is the first record of biflagellate zoospores in the genus *Ligniera.* The early suggestion of Maire and Tison that *R. hypogeae* is in part a species of *Ligniera* thus appears to be confirmed by the observations of Barrett. It is further supported by the fact that this fungus does not cause hypertrophy of its host cells and occurs in some of the hosts which harbor other species of this genus. Whether or not it is identical to *L. verrucosa* as Maire and Tison, and Cook suggest, however, is not certain at present, since well-defined cystosori and resting spores have not yet been described.

SOROLPIDIUM

Nemec, 1911. Ber. Deut. Bot. Ges. 29: 48. 1911b, Bull. Int. Emp. Fran. Joseph Acad. Sci. 16: 69.

(PLATE 13, FIG. 1–25)

Cystosori one or more in a cell, indefinite in size and shape; flat and almost round, oval, elongate, angular and lamellate; consisting of few to many resting spores arranged in linear series, in single or double, flattened layers, or irregular masses. Resting spores variable in size and shape, usually polygonal or hexagonal at first but becoming knobby and somewhat stellate at maturity; usually producing several zoospores in germination. Plasmodia one to several in a cell, variable in size and shape, often lying in the central vacuole or surrounding the latter as a broad band or plate; producing either zoosporangia or cystosori; schizogony unknown. Zoosporangia one or more in a cell, spherical, oval, and elongate; producing few to many zoospores which emerge through an irregular opening in the sporangium wall. Zoospores oval, obpyriform, uniflagellate (?), size unknown.

S. BETAE Nemec, l.c., pl. 1, 2, text-figures 1–6. *Ibid.* 18: 24.

Resting spores 4.2×5 μ—4.6×5.2 μ. For further details see the generic diagnosis above.

Parasitic in the roots of *Beta vulgaris*[1] in Czecho-

[1] Cook ('26) reported that Nemec found the parasite in *B. maritima,* which is obviously a mistake.

slovakia (Nemec, l.c.), the U. S. A. (Rawlings, '25), and France (?) (Guyot, '27) without causing hypertrophy of the invaded tissues.

Sorolpidium Betae has been the subject of considerable discussion since the time of its discovery by Nemec. He described it as a species of the Chytridiaceae with close affinities with the Plasmodiophoraceae, but because of the presence of large, thin-walled zoosporangia he did not believe it should be included in this family. Since similar zoosporangia have subsequently been found in several genera of the Plasmodiophoraceae, this objection is no longer significant. The large, thick-walled, stellate resting cysts surrounded by a thin envelope which Nemec figured are now generally recognized as relating to *Olpidium*, and outside of these cysts there is nothing in the life cycle of *Sorolpidium*, as described by Nemec, which excludes it from the Plasmodiophoraceae. The presence of large holoearpic zoosporangia and multinucleate resting spores which produce several zoospores is in line with more recent discoveries in other genera of this family. Saccardo ('26) likewise included *S. Betae* among the Chytridiales. Winge ('13), however, asserted that it is closely related to *Pyrrhosorus* and the Plasmodiophoraceae. Subsequent workers, on the other hand, have questioned the identity of *Sorolpidium* as a distinct genus of this family and contended that it relates to *Ligniera*. Cook ('26) regarded it as a combination of *Ligniera* and *Asterocystis*, a view which Guyot supported in 1927. The latter worker succeeded in inoculating roots of *Beta vulgaris* with *L. verrucosa* and *Asterocystis radicis*, and concluded that Nemec's fungus is merely an accidental association of these two species in the same host tissues. Cook ('32, '33) later incorporated *Sorolpidium* in *Ligniera* and classed *S. Betae* (pro parte) as a synonym of *L. Junci*. In the shape of its cystosori and the fact that it does not cause hypertrophy of the host tissues *S. Betae* is very similar to species of *Ligniera*. Should it prove to be a species of this genus its identity to *L. Junci* and *L. verrucosa* will nonetheless remain somewhat questionable, because Nemec unfortunately did not give any measurements of the sporangia and zoospores.

The life cycle of *S. Betae* is similar to that of other plasmodiophoraceous species. The earliest recognizable stage consists of a uninucleate oval or spherical, highly vacuolate thallus (fig. 1) which usually lies in the primordial utricle of the host cell. This thallus is probably the result of zoospore infection, although Nemec was uncertain whether the zoospore enter directly or first become amoeboid. Within the host the thallus grows in size (fig. 2, 3, 4, and 5), becomes multinucleate and plasmodium-like. The division of the nuclei (fig. 3) during this developmental phase appears to be "promitotic,"[1] according to Nemec's figures, and no sharply-defined chromosomes are formed. One or more plasmodia (fig. 4, 5) may occur within a single host cell and are usually embedded in the host protoplasm or occupy the central vacuole. They may be spherical, oval, elongate, or take the shape of the cell which they occupy. Sometimes, the plasmodium may form a broad band or plate around the vacuole (fig. 5).

At maturity the plasmodium develops a relatively thin, enveloping membrane and may be transformed directly into a zoosporangium. In this respect *Sorolpidium* differs from *Plasmodiophora*, *Ligniera*, and *Octomyxa*, where the plasmodium is reported to cleave into a number of uninucleate segments which develop into zoosporangia. This difference suggests perhaps that the zoosporangia (fig. 6, 7) which Nemec described may relate to a species of *Olpidium* (*Asterocystis*) with large stellate resting spores. It is to be noted in this connection, however, that the sporangia of *Olpidium* usually form more or less elongate exit tubes, which are lacking in Nemec's *S. Betae*. On the other hand, Nemec may have overlooked the cleavage stage of the plasmodium which results in the formation of several zoosporangia. His text-figure 5 suggests this possibility. At any rate, the protoplasm of the zoosporangium cleaves into uninucleate segments (fig. 6, 7) which become zoospores and emerge through an irregular opening in the sporangium wall. The zoospores from such sporangia are usually uninucleate, oval or pyriform (fig. 8) and uniflagellate (?). Unfortunately Nemec did not say whether they were anteriorly or posteriorly flagellate, which would have settled conclusively their identity as well as that of the large zoosporangia shown in figures 6 and 7. If these zoospores relate to a plasmodiophoraceous species they will doubtless prove to be anteriorly biflagellate and heterocont.

In other mature plasmodia which occur in almost empty host cells, Nemec found that the nuclei lacked nucleoli and were comparatively poor in chromatic material (fig. 9). Peripheral chromosomes later appeared (fig. 10), and the nuclei divided in regular mitotic fashion (fig. 11–15). The appearance of these nuclei and their manner of division are very similar to what has been described in most of the other genera, and suggests that figures 9 to 15 relate to the so-called "akaryote" stage and prophases of meiosis preceding sporogenesis. Some of the nuclei in figure 10 have six chromosomes. The same numbers are present in figures 14 and 15, but whether or not this is the basic number in *Sorolpidium* is uncertain. Nemec described a second mitosis in such plasmodia in which the chromosomes are larger, elongate, and rod-shaped, but it is difficult to reconcile his conclusions about this division with previous and subsequent descriptions of the homeotypic mitosis in other genera.

These plasmodia, nonetheless, develop a thin enveloping membrane and cleave into uninucleate segments (fig. 16), which form fairly thick walls and become resting spores. The enveloping membrane soon disappears, but the resting spores remain attached and thus form cystosori of various sizes and shapes (fig. 17–24). They may consist of a linear

[1] Cook ('26) stated that Nemec did not figure "promitosis," but he obviously overlooked figure 3.

row of resting spores (fig. 19), double rows (fig. 18, 24), or flat, rounded or irregular masses (fig. 17, 20, 21). When first formed the resting spores are usually polygonal (fig. 17), but later they become more globose. As they mature they become knobby and somewhat stellate (fig. 18, 19, 20, 22, 23) with intercellular spaces between them. Single isolated resting spores may be formed occasionally (fig. 23), and among the normal-sized spores in a cystosorus unusually large ones may sometimes occur as is shown in figure 17. In these respects *S. Betae* shows the same variations as other genera.

Since the resting spores function as sporangia in germination, Nemec called these aggregates sporangesori. In germination the resting spores increase in volume and become more rounded in outline (fig. 17), their nuclei divide, and the protoplasm cleaves into uninucleate segments which round up (fig. 22) and become zoospores. An opening in the spore wall is soon formed through which the zoospores emerge (fig. 24). The number of zoospores formed varies with the size of the resting spores. Nothing is known about the size of these zoospores, but they are probably similar to those formed in the large zoosporangia. Nemec found no evidence of gametes and sexual fusion in *S. Betae*.

Like species of *Ligniera, S. Betae* causes no hypertrophy or other malformations of the invaded tissues. In fact, parasitized rhizodermal cells may remain alive longer than non-infected cells, according to Nemec. The presence of the parasite, however, causes an accumulation of cytoplasm in infected cells and enlargement of the host nucleus (fig. 4, 5). The latter may often become irregular and develop an unusually large nucleole. As the plasmodia mature the host protoplasm is reduced to a thin parietal layer and eventually disintegrates. The entrance of the zoospore through the cell wall often leads to a marked reaction. As is shown in figure 25 the entrance hole becomes plugged up and a conspicuous thickening around this plug is formed on the inner periphery of the wall.

ANISOMYXA

Nemec, 1913. Bull. Int. Empr. Fran. Joseph Acad. Sci. 18: 18.

(PLATE 13, FIG. 26–45)

Plasmodia usually solitary, partly or almost completely filling host cell and conforming with the latter's size and shape; schizogony unknown; cleaving into groups (sporangiosori) of small and large zoosporangia. Sporangiosori usually solitary, rarely more than one in a cell; indefinite in size and shape; spring and winter sporangiosori composed of small and large zoosporangia respectively. Zoosporangia variable in size, exit papillae or tubes lacking; producing four or more uniflagellate (?) zoospores. Cystosori made up of relatively thick-walled resting-spores; germination unknown.

It is not certain from Nemec's account whether or not cystosori composed of thick-walled resting spores are formed in this genus. He reported that the plasmodium divides into aggregates or sori of polygonal, hexagonal and oval cells which are quite variable in size. In spring and summer, sori of small and uniform cells are formed (fig. 41), while those produced in the winter are made up of much larger cells (fig. 43, 44). In both types of sori, however, the cells are uninucleate at first but later become multinucleate. Because they have thin walls and produce several

PLATE 13

Sorolpidium Betae

(All figures after Nemec)

Fig. 1. Cell of *Beta vulgaris* with two uninucleate parasites.

Fig. 2. Binucleate stage of *S. Betae*.

Fig. 3. Four-nucleate stage; nuclei dividing "promitotically" (?).

Fig. 4. Host cell with four plasmodia.

Fig. 5. A large band-shaped plasmodium surrounding the central vacuole.

Fig. 6, 7. Large and small zoosporangia with zoospores.

Fig. 8. Zoospores from sporangia.

Fig. 9. Plasmodium in which the nuclei lack large nucleoli (achromatic stage?).

Fig. 10. Later stage; nuclei with parietal chromosomes.

Fig. 11–15. Division stages of such nuclei with six well-defined chromosomes.

Fig. 16. Plasmodium cleaving into resting spores.

Fig. 17. Young cystosorus (?) with polygonal resting spores.

Fig. 18–22. Cystosori of various sizes and shapes.

Fig. 19, 20, 22. Cystosori of mature knobby, stellate resting spores.

Fig. 22. Resting spores with zoospores.

Fig. 23. Single, isolated stellate resting spore.

Fig. 24. Cystosorus of empty germinated resting spores.

Fig. 25. Swollen cell wall at point of entry of zoospore.

Anisomyxa Plantaginis

(All figures after Nemec)

Fig. 26. Uniflagellate zoospore.

Fig. 27. Biflagellate (?) zoospore.

Fig. 28. Small uninucleate thallus.

Fig. 29. Binucleate thallus with resting nuclei.

Fig. 30. Same with both nuclei dividing "promitotically" (?).

Fig. 31. Tetranucleate thallus with centrosomes and astral rays.

Fig. 32. Equatorial plate stage of "promitosis" with cap-like centrosomes at poles.

Fig. 33. Achromatic or "akaryote" (?) stage of nuclei.

Fig. 34–36. Prophase of meiosis (?).

Fig. 39. Mature multinucleate plasmodium with some of the nuclei associated in pairs.

Fig. 40. Zoosporangia cleaving into zoospores.

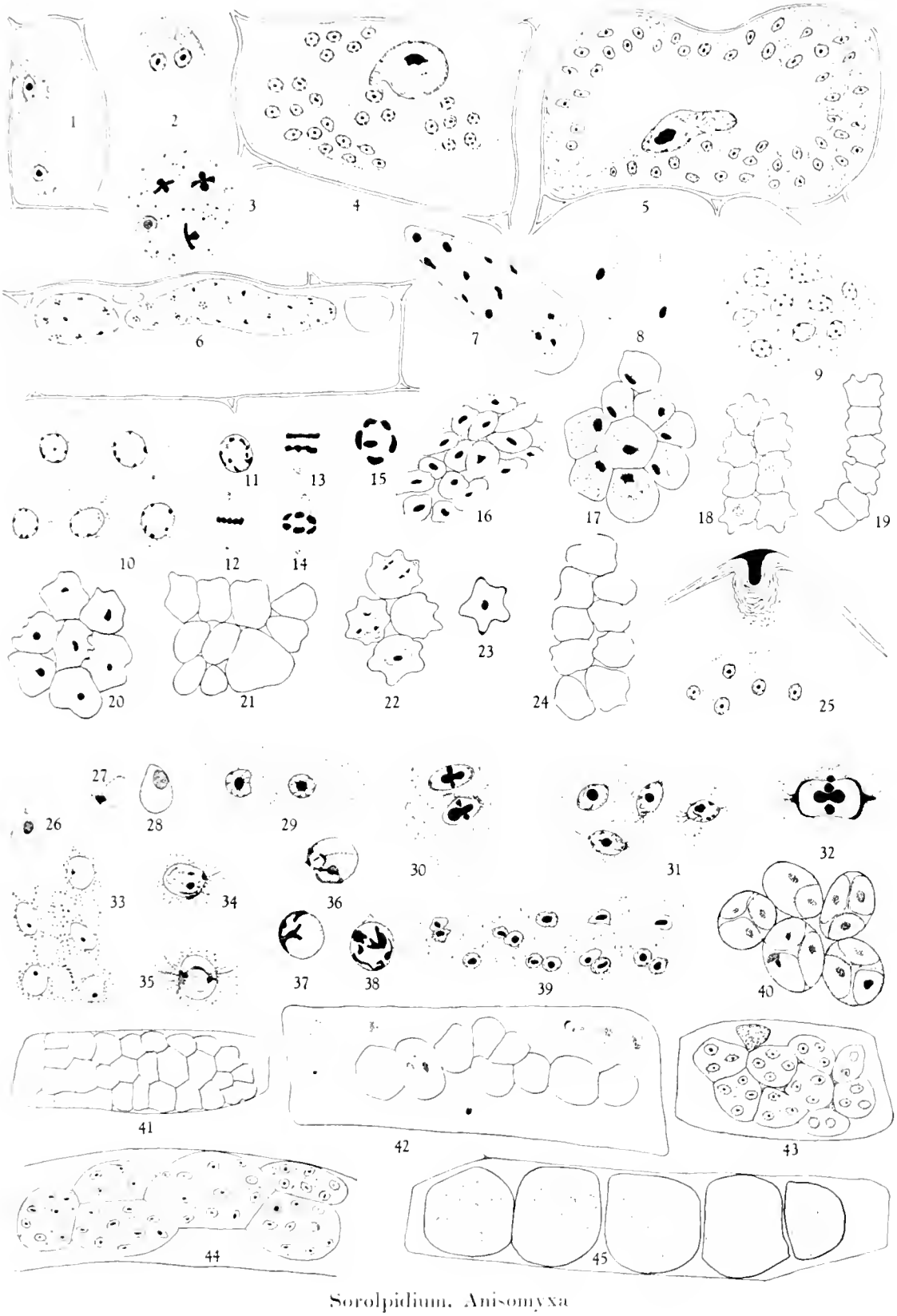
Fig. 41. Spring sporangiosorus composed of small zoosporangia.

Fig. 42. Sporangiosorus composed of sporangia arranged in a linear series.

Fig. 43, 44. Sporangiosori of large multinucleate sporangia.

Fig. 45. Cell with cyst-like sporangia.

PLATE 13



Sorolpidium. Anisomyxa

zoospores Nemec regarded them as zoosporangia and like in *Sorolpidium* named the aggregates sporangiosori. It is not improbable, however, that some of these sori may be cystosori of relatively thin-walled resting spores, since in describing the cytology of *Anisomyxa* Nemec reported several nuclear changes and appearances (fig. 11–15) which suggest the meiotic prophases which precede sporogenesis.

Although his account of *Anisomyxa* is fragmentary and not altogether clear, it is evident that Nemec was dealing with a species of the Plasmodiophoraceae. Whether or not it represents a new and distinct genus, however, remains to be seen from future studies. Nemec regarded *Anisomyxa* as closely related to *Rhizomyxa* and possibly intermediate between the Plasmodiophoraceae and Synchytriaceae. Fitzpatrick ('30) discussed it as a doubtful genus, while Cook ('32, '33) merged it with *Ligniera* and listed *A. Plantaginis* (*pro parte*) as synonymous with *L. Junci*. The latter worker had previously ('26, '27) found *L. Junci* in roots of *Plantago major*, which doubtless influenced his belief that *A. Plantaginis* is a combination of *L. Junci* and a chytrid.

A. PLANTAGINIS Nemec, l.c., p. 21, pl. 1, 2. Text-figures 1–5.

Spring and winter sporangiosori variable in size and shape; irregular, elongate, and oval; consisting of a few to numerous sporangia. Zoosporangia usually remaining attached together in a sorus; polygonal, hexagonal, oval or almost spherical with thin, smooth walls; spring zoosporangia approximately $4.5\times6\ \mu$, producing 4 zoospores; winter sporangia $10.5\times15\ \mu$, forming numerous zoospores. Zoospores oval, $1.5\times1.8\ \mu$, spherical, $1.5\ \mu$ in diameter.

Parasitic in the roots of *Plantago lanceolata* in Czechoslovakia, without causing hypertrophy of the invaded tissues.

The zoospores of *A. Plantaginis* are very small and oval to spherical in shape (fig. 26). Nemec reported them as uniflagellate, but he did not state if the flagellum is anteriorly or posteriorly inserted. It is to be noted here that he figured one zoospore (fig. 27) which appears to be biflagellate. It is accordingly quite probable that when this species is studied more intensively the zoospores will prove to be anteriorly biflagellate and heterocont. Nemec postulated that zoospores of two sizes might be produced, because he found cleavage segments of unequal sizes in several zoosporangia.

Penetration of the parasite into the host cell has not been observed. Nemec found small oval uninucleate thalli in several host cells (fig. 28, 42) which appear to have come from zoosporangia. Such thalli apparently grow in size as their nuclei divide and eventually become multinucleate plasmodia (fig. 31, 39). The nuclear divisions (fig. 30, 32) in the developing plasmodium resemble the so-called "promitosis" type and are described by Nemec as vegetative mitoses in which centrosomes and astral rays are usually quite conspicuous (fig. 31, 32). Following completion of the vegetative divisions the nuclei lose their chromatin, and the nucleole is reduced to a small globule (fig. 33). The cytoplasm, on the other hand, becomes filled with small deeply stainable granules. This stage is followed shortly by another in which dense chromatic granules, rods, and bands appear at one side of the nuclei (fig. 36, 37) and suggest synaptic phases of meiosis. These stages initiate the reproductive divisions, according to Nemec. However, figures 33 to 38 are strikingly like the "akaryote" phase and prophase stages of meiosis which in other genera have been interpreted as initiating sporogenesis. It is not clear from Nemec's account whether these stages precede the formation of spring or winter sori.

The mature plasmodium does not become enveloped by a wall like in *Sorolpidium* but cleaves directly into sporangia which remain aggregated and form sori. The zoosporangia are polygonal (fig. 41) at first but later become oval and spherical (fig. 40). In the small spring sporangia, two nuclear divisions of the mitotic type occur, and the protoplasm cleaves into four segments which become zoospores (fig. 40). In the larger winter sporangia numerous mitoses occur, producing multinucleate zoosporangia (fig. 42–44) which give rise to numerous zoospores (fig. 21, 22). No exit papillae or tubes were observed by Nemec and nothing is known about the emergence of the zoospores from the sporangia.

Nemec found no evidence of sexual fusions in *Anisomyxa*, but he pointed out that the nuclei in the mature plasmodium (fig. 39) are often associated in pairs, implying perhaps that karyogamy may take place. This suggestion is further implied by his figures of synaptic (fig. 36, 37) and diakinetic (fig. 38) division stages. In addition to the two types of sporangiosori Nemec also found several large, sporangium-like oval cysts (fig. 45), $14.5–20\ \mu\times20–26\ \mu$, which he believed might possibly be cystosori. Whether or not these are large isolated resting spores of *A. Plantaginis* is not certain.

TREMATOPHLYCTIS

Patouillard, 1918. Bull. Soc. Mycol. France 34: 86, fig. A–G.

(PLATE 14, FIG. 1–6)

Patouillard established this genus to include a species, *T. Leptodesmiae*, which parasitizes petioles and leaves of *Leptodesmia congesta* in Madagascar. His diagnosis was based on dried material collected by Viguier in 1912, and there is very little evidence in his brief description to warrant inclusion of this species in the Plasmodiophoraceae at the present time. The infected leaves and petioles become thick, fleshy (fig. 1, 2), and reddish in color, and later numerous round or irregular, 0.5 to 3 mm. high, solitary or aggregated, open, aecium-like pustules filled with yellow spores appear in the infected areas.

The earliest known developmental stages of *T. Leptodesmiae* consists of an elliptical, round, or irregular plasmodium (?) which fills the hypertrophied host cell (fig. 4). Its protoplasm is homogeneous, brownish, and slightly granular and not enveloped by a distinct membrane. With maturity the protoplasm becomes more granular, and the entire thallus segments into spores, which are at first polygonal but later become oval and spherical, 12–16 μ, and develop smooth hyaline walls (fig. 5). When mature they have a yellowish tint, and as the sorus breaks open to the outside of the host it assumes the structure and appearance of a cup-like pustule filled with pulverescent spores (fig. 2). Germination of these spores has not been observed.

Patouillard's figures and description of the sorus, spore formation, and the appearance of the pustules suggest that *T. Leptodesmiae* may possibly be a species of *Synchytrium* of the *S. decipiens* type which forms open pustules. His figures of a naked plasmodial stage and comparatively thick-walled spores, however, militates against this view, but in dried herbarium material it is obviously difficult to determine the presence or absence of an enveloping membrane. Saccardo ('31) listed *T. Leptodesmiae* among the Plasmodiophoraceae, but Cook ('33) excluded it. Palm (see Palm and Burke, '33) collected material of a species closely related, if not identical, to *Trematophlyctis* on an unnamed host in southern Madagascar, and his statement that it is an "undoubted member of this family" carries the implication that he believed Patouillard's genus might be valid. Unfortunately Palm has published nothing additional on this fungus, and the status of *Trematophlyctis* will remain doubtful until more is known about its life cycle.

In relation to these doubtful genera a discussion of *Pyrrhosorus* Juel may be logically presented at this point, although in so doing the author does not imply that it should be included in the Plasmodiophoraceae as this family is now recognized. This genus was created by Juel ('01) for an orange-colored species, *P. marinus*, which he discovered in a red alga, *Cystoclonium purpurasceus*, in Sweden. Since he found it only in dead branches Juel concluded that it is a saprophyte, but Winge ('13) believed that during some of the developmental stages reported by Juel the organism may be parasitic. *Pyrrhosorus marinus* has never since been observed, but because it includes several plasmodiophoraceous-like stages in its life cycle it merits consideration in any discussion of the Plasmodiophorales. Juel was uncertain about its taxonomic position, relationship, and phylogeny but pointed out and discussed the characters it has in common with *Woronina*, *Rhizomyxa*, *Tetramyxa*, *Protomyxa*, and other genera of lower organisms. He particularly stressed the similarity of its type of sporogenesis to that of *Tetramyxa*.

The life cycle of *P. marinus* is as follows: In the early developmental stages it consists of small globular thallus lying within the host cell (Pl. 11, fig. 8). Such thalli may often be associated in pairs (fig. 9) or groups, and Juel accordingly considered it possible that they may later coalesce and form a large plasmodium. The uninucleate thallus grows in size as its nucleus enlarges (fig. 10) and apparently divides. Mitoses in the plasmodium have not been observed, and Juel was uncertain as to the manner of origin of the multinucleate stages. A later stage is shown in figure 11 of a plasmodium with four large nuclei. The developing plasmodia apparently have the ability to dissolve intervening cell walls (fig. 11) and may eventually occupy several cells. Although they may be distinctly amoeboid in shape with numerous blunt, pseudopod-like extensions and vacuoles (fig. 12, 13) it is not certain from Juel's account that they move about and migrate from cell to cell as in *Plasmodiophora*, etc. No evidence of schizogony was observed by Juel, but Winge interpreted some of the uninucleate stages as probable meronts.

The mature plasmodium is multinucleate, vacuolate, and usually irregular in shape (fig. 12–14), and just before sporulating forms an enveloping membrane like *Sorolpidium*. Plasmodia which are extensively drawn out and occupy several host cells may accordingly appear lobed, irregular and tubular (fig. 18) after the wall has formed. Following this stage the protoplasm divides into uninucleate segments. In this process no distinct cleavage furrows have been observed. The plasmodium appears to become highly vacuolate (fig. 14) during this process, and the cytoplasm accumulates around the nuclei and forms stellate protoplasmic islands which resemble somewhat the sporonts of *Tetramyxa*. These segments soon become almost spherical or spindle-shaped (fig. 14), and Juel thought that the latter type of cells are formed in plasmodia which are highly vacuolate and scarce in cytoplasm. In addition to these two kinds of segments, irregular elongate, oval and smaller ones may be formed, apparently as the result of unequal cleavage, which finally degenerate.

The spherical, 8 μ in diameter, and spindle-shaped segments are uninucleate, naked, and never develop a distinct wall. They aggregate to form a definite sorus (fig. 15) and each cell soon divides into octads of spores as in *Octomyxa*, which led Juel to call them spore-mother cells. In this process of spore formation the nuclei divide mitotically (fig. 21–24) and each mitosis is followed by cell division. Definite chromosomes (2 to 5) are formed on a sharply-defined spindle during mitosis, and there is no evidence of "promitosis," according to Juel's figures. Each of the eight naked spores soon becomes transformed directly into a zoospore without developing a thick wall and becoming dormant. The mature zoospores are small, pyriform, 4.5 × 2.5 μ, with a tapering end, laterally biflagellate and isocont (fig. 7). In addition they possess a brilliant orange-colored spot or globule which resembles the eye spot of algae and lies at the point of insertion of the flagella. The zoospores apparently infect the host cells and develop into the small thalli shown in figures 8 and 9. Cysto-

sori or resting spores have not been observed in *P. marinus*.

It is apparent from this description that Juel's fungus differs primarily from the valid species of the Plasmodiophoraceae by its laterally biflagellate, isocont zoospores, naked spore-mother cells and spores, lack of zoosporangia, resting spores, and by its saprophytic nature. As Juel emphasized, the formation of uninucleate spore-mother cells or sporonts by fragmentation of the plasmodium and their subsequent division into 4 and 8 cells is strikingly similar to spore development in *Tetramyxa*. Had *Octomyxa* been known at that time Juel would doubtless have emphasized the relationship of his species with the Plasmodiophorales even further. It is to be noted, however, that in these two genera each mitosis in the sporonts is not immediately followed by cell division as in *Pyrrhosorus*, and that the spores which are formed encyst and pass through a dormant period before giving rise to zoospores. It is possible that under the conditions of Juel's study the spores of *P. marinus* failed to encyst and become dormant. It is also possible that zoosporangia occur in this species but were not present in Juel's material. In that event *P. marinus* would be very similar to *Octomyxa*. However, its laterally biflagellate isocont zoospores with an orange-colored eye-spot constitute a serious obstacle to including it in the Plasmodiophorales at present, unless, of course, Juel was mistaken about the relative lengths and insertion of the flagella. These possibilities, however, are purely speculative. On the other hand, the zoospores are similar to those figured for species of the lower biflagellate Oomycete-like fungi, but until more is known about *P. marinus* its relationship will remain obscure. Winge, nonetheless, considered it closely related to the Plasmodiophoraceae and made extensive comparisons between its life cycle and that of *Sorolpidium*. He regarded the sporangiosori of the latter genus as homologous with the aggregates or sori of spore-mother cells of *Pyrrhosorus*, and believed that the absence of wall around the sporonts in the latter is of minor importance. Cook ('33), on the other hand, regarded the relationship of *Pyrrhosorus* with the Plasmodiophorales as highly questionable.

BIBLIOGRAPHY: DOUBTFUL GENERA

Barrett, J. T. 1935. Phytopath. 25: 898.

Cook, W. R. I. 1926. Trans. Brit. Mycol. Soc. 11: 210. 1927. *Ibid.* 12: 282.

———. 1932. Hong Kong Nat. Suppl. 1: 38.

———. 1933. Arch. Protistk. 80: 223.

Fischer, A. 1892. Rabenhorst's Krypt. 1, abt. 4: 67.

Fitzpatrick, H. M. 1930. The lower fungi—Phycomycetes. New York: p. 63.

Guyot, A. L. 1927. Bull. Soc. Path. Veg. l'Ent. Agr. France 14: 181.

Juel, H. O. 1901a. Bih. K. Svensk. Vet.-Akad. Hand. 26 afd. III, No. 14: 1.

———. 1901b. Rev. Mycol. 24: 111.

Maire, R., and A. Tison. 1911. C. R. Acad. Sci. Paris, 152: 206.

Minden, M. 1915. Krypt. Fl. Mark. Brandenburg 5: 278.

Nemec, B. 1911. Österr. Ungar. Zeitschr. f. Zucker u. Landw. 40.

Palm, B. T., and M. Burk. 1933. Arch. Protistk. 79: 263.

Rawlins, T. E. 1925. Phytopath. 15: 727.

Saccardo, P. A. 1926. Sylloge fungorum 24, sect. 1: 17. 1931, *Ibid.* 25: 12.

Schroeter, J. 1897. Engler and Prantl, die Nat. Pflanzf. I, 1: 5.

Wildeman, E. 1893. Ann. Soc. Micro. Belge. 17: 25.

Winge, O. 1913. Ark. f. Bot. 12, No. 9: 29.

EXCLUDED GENERA

Herewith are presented descriptions and illustrations of three genera which have been included in the Plasmodiophorales by various workers, primarily for want of a better group in which to place them. Uniflagellate zoospores are reported to occur in *Cystospora* but are apparently lacking in *Sporomyxa* and *Peltomyces*. Except for a multinucleate plasmodial stage, resting spores, and the occurrence of intranuclear mitosis and schizogony these genera have little in common with the Plasmodiophorales as this order is now generally recognized. They are, nevertheless, described here so that their validity as members of this order may be judged independently.

PLATE 14

Trematophlyctis Leptodesmiae

(All figures after Patouillard)

Fig. 1. Leaves of *L. congesta* with galls.

Fig. 2. Portion of a branch with a large gall and three open pustules.

Fig. 3. Section of a gall showing several sori.

Fig. 4. Naked plasmodium (?) filling greatly enlarged cell.

Fig. 5. Group of resting spores formed by segmentation of plasmodium.

Fig. 6. Individual resting spores.

Pyrrhosorus marinus

(All figures after Juel)

Fig. 7. Laterally biflagellate isocont zoospores.

Fig. 8. Uninucleate thallus.

Fig. 9. Two paired young thalli.

Fig. 10. Uninucleate thallus with enlarged primary nucleus.

Fig. 11. Four-nucleate thallus passing through cell wall.

Fig. 12. Multinucleate thallus.

Fig. 13. Multinucleate amoeboid thallus.

Fig. 14. Cleavage of thallus.

Fig. 15. A sorus of spore mother cells.

Fig. 16. Isolated spore mother cell.

Fig. 17. A sorus, the spore mother cells of which have divided into groups of four daughter cells.

Fig. 18. Spindle-shaped spore mother cells (?) in a branched thallus.

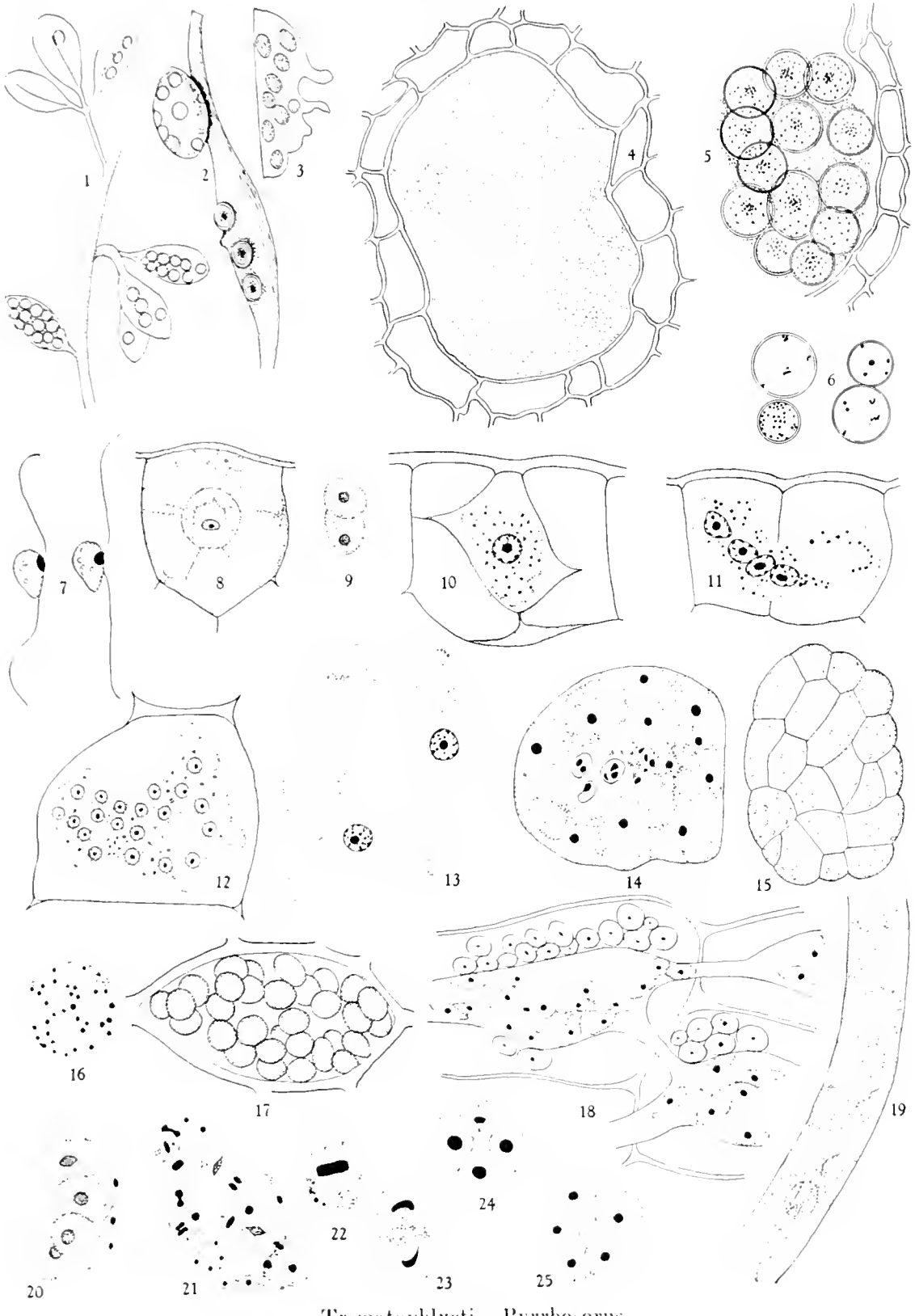
Fig. 19. Spindle-shaped spore mother cells and accessory sterile cells in an elongate host cell.

Fig. 20. Sorus with spore mother and sterile cells.

Fig. 21. Sorus with spore mother cells undergoing mitosis.

Fig. 22–24. Mitosis and cytokinesis of spore mother cells.

PLATE 14



Trematophlyctis, Pyrrhosorus

by researeh workers. Doubtless, there are numerous other plasmodiaeeous organisms whieh resemble the true Plasmodiophoraceae and simple fungi which must eventually be given serious consideration by myeologists and protozoologists, and it is hoped that by presenting the available data here greater interest and research may be stimulated in these borderline organisms.

SPOROMYXA

Leger, 1908. Arch. Protistk. 12: 111.

(PLATE 15, FIG. 1–25)

Sporomyxa was ereated by Leger for a virulent parasite, *S. Scauri,* which he found in the coelome of the imago of *Scaurus tristis* in Algeria. The parasite has a predilection for the adipose tissue and may be found in enormous numbers there. Unlike most plasmodiophoraceous fungi, it destroys infeeted eells completely without stimulating them to divide or enlarge. The earliest known stage consists of a small, naked, spherieal, ovoid, 6–8 μ, or spindle-shaped body with an unusually large, 5 μ, nueleus and finely granular eytoplasm (fig. 1). It does not appear to have a sharply defined membrane and lies embedded in the host eytoplasm. As it inereases in size the nucleus divides mitotieally with an intranuclear spindle (fig. 2), and the thallus beeomes binueleate. In this stage it may divide by binary fission (fig. 3). Additional nuelear divisions oeeur (fig. 4), and larger, naked, multinueleate plasmodium-like thalli are eventually formed (fig. 6). Leger found no thalli with more than 8 nuclei, and he believed that from this stage on the parasite undergoes sehizogony into uninueleate meronts or sporulates, so that thalli with a large number of nuclei are never formed.

The mature thallus may be spherical, elliptieal, and sometimes amoebiform, aeeording to the position its oeeupies in the host tissue, and although it may have the shape and appearanee of an aetive amoeba, it does not move or undergo ehanges in form. Its eytoplasm is denser toward the eenter, but no distinet endo- and ectoplasmie layers are distinguishable. No wall or membrane is present, and the whole thallus may be enveloped by the host protoplasm (fig. 6). In addition to these thalli, Leger found other smaller ones with numerous fat globules and ehromatie granules in the eytoplasm and small nuclei whieh appeared to be laeking in ehromatin (fig. 7). He believed sueh thalli oeeur at the close of the vegetative phase of *S. Scauri* and mark the beginning of sporogenesis.

Unlike the true plasmodiophoraceous genera, no segmentation of the multinueleate thallus into numerous separate spores or eystosori has been observed in *S. Scauri.* Resting spores, however, oeeur very abundantly in the adipose tissue, but Leger was not certain whether they are formed by eneystment of vegetative uninueleate thalli or are the produets of more or less simultaneous sehizogony of a multinueleate body. He admitted the possibility of both methods, but did not show any figures of the latter proeess. The spores may sometimes oeeur in groups, but it is not evident that these aggregates have been formed by segmentation of a multinueleate plasmodium as in *Plasmodiophora.* The only developmental stages of resting spores deseribed by Leger relate to small, isolated spores. These are apparently formed by the eneystment of uninueleate thalli during whieh proeess the nueleus shrinks in size as ehromatie material is extruded from the nueleole into the eytoplasm (fig. 9–13). As this goes on, the wall thickens and differentiates into a thiek outer and a thin inner layer. In bi- and multinucleate thalli, spore formation may be aeeompanied by nuclear fusions (fig. 12, 13) of the type deseribed by Prowazek ('05) for *P. Brassicae.* Leger interpreted these fusions as representing rudimentary sexuality. The majority of spores are ovoid, $8\times10\ \mu$, but they may often be more elongate, $4\times8\ \mu$, spherical, obpyriform, eonstrieted in the middle, and unusually large, 30–40 μ (fig. 15–17). The small spores are usually

PLATE 15

Sporomyxa Scauri

(All figures after Leger)

Fig. 1. Uninueleate thallus.

Fig. 2. Mitosis with intranuclear spindle and minute chromosomes.

Fig. 3. Binucleate thallus undergoing binary fission.

Fig. 4. Mitosis in a binucleate thallus.

Fig. 5. Tetranucleate thallus.

Fig. 6. Large, amoebiform, eight-nucleate thallus within host cell.

Fig. 7. Thallus with ehromatie granules in eytoplasm; nuclei without (?) chromatin.

Fig. 8–12. Sueeessive stages in resting spore formation.

Fig. 13, 14. Nuclear fusion (?) in resting spore.

Fig. 15–17. Large, abnormal resting spores.

S. Tenebronis

(All figures after Reitschel)

Fig. 19–20. Developmental stages of thallus.

Fig. 21. Synchronous nuelear division; polar and profile views.

Fig. 22. Completion of cleavage into spore rudiments.

Fig. 23. Later stage of same.

Fig. 24, 25. Uni- and binucleate spores.

Cystospora batata

(All figures after Elliott)

Fig. 26. Resting state.

Fig. 27. Amoebae.

Fig. 28–32. Nuclear division and multiplieation.

Fig. 33. Sixteen-nueleate stage of thallus; nuclei of unequal size.

Fig. 34. Migration of plasmodium through rootlet.

Fig. 35. Cells of host with amoebae and plasmodia.

Fig. 36. Root tip eells with plasmodium and amoebae; nuclei of unequal size in plasmodium.

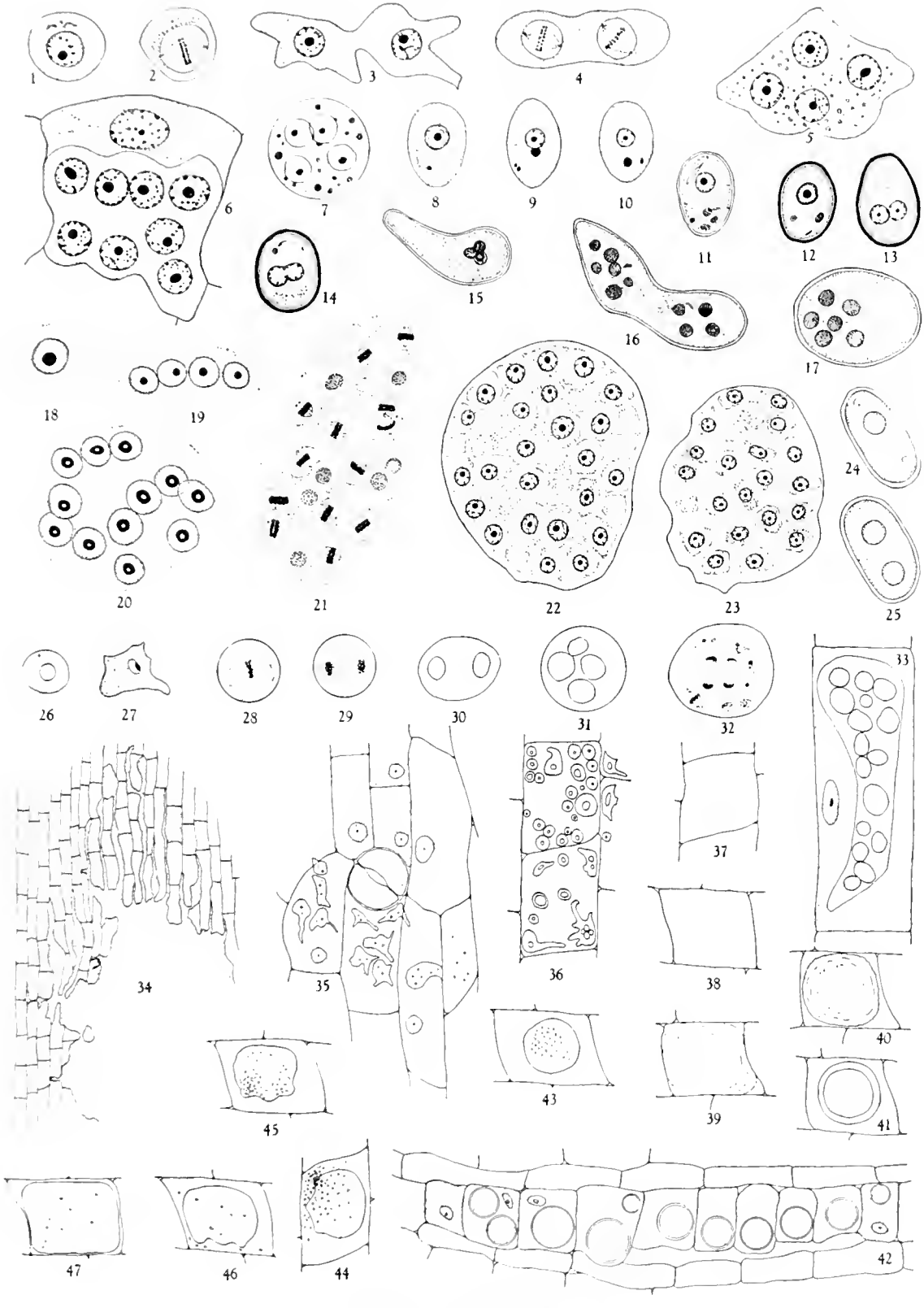
Fig. 37–41. Stages in eyst formation from a plasmodium.

Fig. 42. Row of cysts.

Fig. 43, 44. Formation in and liberation of zoospores from cysts.

Fig. 45–47. Degeneration of cysts.

PLATE 15



Sporomyxa. Cystospora

uninucleate, but the abnormal ones may possess 2 to 30 nuclei scattered about or aggregated in groups. The wall of the spore is hyaline, streaked, and thick, and by treatment with iodine and sulphuric acid it assumes a bluish tint, indicating the presence of cellulose.

A second species, *S. Tenebriones,* was found by Reitschel ('36) in the fat bodies, ovaries, and connective tissues of the larvae and imago of *Tenebrio molitor.* The life history and development of this species (fig. 18–25) are similar to those of *S. Scauri* with the exception that the thalli become larger and undergo cleavage at maturity. At the time of sporulation they may contain considerably more than a hundred nuclei and are enveloped by a thin membrane. The protoplasm cleaves into uninucleate segments (fig. 22, 23) which later round up and become the resting spores as in *Plasmodiophora.* The soral membrane disintegrates shortly thereafter and frees the spores. These are usually uninucleate (fig. 24), rarely binucleate (fig. 25), hyaline, smooth, and measure 9–13 μ by 4.5–7 μ. In neither of these species have spore germination, zoosporangia, and zoospores been observed.

Leger believed that *Sporomyxa* may be closely related to *Sappinia* because of its method of sporulation. Maire and Tison ('09) regarded it as of doubtful affinity with the Plasmodiophorales and stressed lack of promitosis in nuclear division as a distinctive character. Fitzpatrick ('30) and Cook ('33) excluded it on the grounds of its habitat and ellipsoidal isolated resting spores, but as Palm and Burk ('33) have pointed out, "the circumstance that it attacks an animal host could hardly be taken as a serious objection." However, our knowledge of its life cycle and cytology seems hardly sufficient to justify its inclusion in the Plasmodiophorales at the present time.

PELTOMYCES

Leger, 1909. C. R. Acad. Sci. Paris 149: 239.

Leger founded this genus to include three parasites, *P. hyalinus, P. Blatella,* and *P. Forficulae,* which occur in the malpighian tubes of *Oloerates, Blatella,* and *Forficula* species. His description of the genus was based primarily on the development and life cycle of *P. hyalinus,* apparently the only species which he studied in detail. This species makes its appearance in the epithelium as a small, 2 μ, uninucleate globular body. Its nucleus multiplies mitotically, and the parasite soon grows into a multinucleate disc-shaped plasmodium which subsequently undergoes schizogony and forms a large number of small, 2–3 μ, uninucleate sporonts.

At the conclusion of schizogony the sporogonic phase begins. Each sporont increases in size while its nucleus divides mitotically several times. Two types of nuclei are thus formed: small, densely-stainable somatic nuclei without membranes, and larger, normal-looking gametic nuclei with well-defined membranes. The former nuclei disintegrate, while the latter become enveloped in a small spherical mass of cytoplasm and are soon transformed into bowl-shaped, 2 μ, gametes. These fuse in pairs after their nuclei have undergone a chromatic reduction, and this is soon followed by karyogamy. The zygotes or incipient diploid resting spores formed in this manner develop a wall and assume a cylindrical, 3×9 μ, shape. Each mature sporont thus encloses within its thin wall 4 to 8 spores arranged side by side and looks like a sporangium. The gametes in the sporonts which fail to fuse develop into parthenogenetic spores of about half the size of the diploid spores. In some cases prematurely formed sporonts, instead of producing gametes, form small endogenous cells which escape from the sporonts and behave as schizozoites in the host. Leger did not illustrate any of these species, and his account of their development is brief and fragmentary. Zoospores, sporangia, and cystosori are unknown in *Peltomyces.*

CYSTOSPORA

Elliott, 1916. Delaware Agr. Exp. Sta. Bull. 114: 15.

(PLATE 15, FIG. 26–47)

This genus was created by Elliott for a myxomycete-like organism, *C. batata,* which is reported to cause "soil rot," "pit" or "pox" of sweet potatoes in the United States. Elliott placed it in the Plasmodiophorales, but its inclusion here is very doubtful, if at all warranted. In fact, some workers (Manns and Adams, '25) have expressed doubt about the existence of an organism of this type and asserted that some of the stages figured by Elliott may be nothing more than products of disturbed metabolism of the sweet potato. Tabenhaus ('18), however, reported that he was able to grow this organism in pure culture on sweet potato agar made up according to Elliott's formula. He further confirmed Elliott's account of the life cycle of *C. batata.*

According to these workers, the zoospores are small, 1–2 $\mu\times1.5$–3 μ, globose with tapering ends and possess a short flagellum, but it is not evident from their descriptions whether the flagellum is anterior or posterior. The zoospores are nonetheless produced in great numbers (fig. 43, 44) and may remain active from 1 to 7 days in rare instances, according to Tabenhaus. The period of activity, however, is usually short, often less than half an hour. The zoospores may sometimes fuse in pairs and form round zygotes which later become amoeboid (fig. 26, 27). According to Elliott, they bore through the cell wall and infect the host as amoebae, but Tabenhaus reported that infection may also take place by means of a plasmodium. The nuclei of the young parasite divide mitotically and simultaneously (fig. 28, 29, 32), but unfortunately Elliott's figures are so small and poorly drawn that it is impossible to deter-

mine whether or not the divisions resemble the promitosis described for other genera.

Several amoebae and small plasmodia may coalesce and form larger plasmodia, according to Elliott, which migrate deeper into the infected tissue (fig. 34) in much the same manner described by Kunkel for *Sorosphaera*. Large plasmodia may contain from 200 to 300 nuclei, and at maturity form large multinucleate cysts (fig. 37–42). Elliott reported that each plasmodium forms a single cyst, but his figures suggest that more than one may be produced. The plasmodium fills the host cell at maturity (fig. 36–38), becomes more dense in the center, condenses, and eventually forms a thick, smooth wall (fig. 39–41). After a short rest period the cyst germinates, and in this process the wall becomes very thin (fig. 43, 44), and the protoplasm cleaves into numerous zoospores. In this manner several generations of zoospores per season are formed in infected roots and pox lesions, each generation of which migrates deeper into the tissues. Eventually "all plasmodia seem to collect, cease advancing, turn backwards, and leave the pit for the soil," according to Tabenhaus. These plasmodia are believed to encyst in the soil and live through the winter in this stage.

Soil rot, pit, or pox is a widely distributed and common disease and has been reported from a number of states (Halsted, '90, '92, '96; Price, '95; Duggar, '97; Townsend, '99; Wilcox, '06; Barre, '10; Tabenhaus, '14, '16; Harter, '16; Poole, '22, '24, '25; Anonymous, '24, 26; Harter and Weimer, '29, and others). Pox may also occur on the white potato, turnips, and possibly beets and tomatoes (Tabenhaus, '18). The cause of pox, however, has been the subject of much controversy. Halsted attributed it to a filamentous fungus which he named *Acrocystis batatas*, but from extensive study of the disease, Tabenhaus ('14) and Elliott concluded that *A. batatas* is non-existent and had previously been mistaken for another organism. The latter worker claimed that pox is caused by a myxomycetous fungus which he named *C. batata*. Elliott further asserted that Halsted had figured several stages of this slime mold, and he accordingly listed *Acrocystis batatas* as synonymous with *C. batata*. Tabenhaus ('18) confirmed Elliott's observations in Texas, and found that another fungus, *Actinomyces poolensis*, may also occur as a superficial wound parasite in pox spots produced by *C. batata*.

Since that time the existence of cysts and other stages of *C. batata* has been seriously questioned and denied by Manns and Adam. In mature pox lesions no evidence of an organism resembling a slime mold was found by these workers, and they ('21) interpreted the so-called cysts of Elliott as "products of metabolism in the form of reserve substances." Later Manns ('24) demounted some of the pox material which Elliott had stained with Flemming's triple dye and restained it with Ziehl's carbol fuchsin, and in each instance he found an *Actinomyces* species present. He ('25, '26) and Adams ('29) later questioned the existence of *C. batata* and maintained that pox of sweet potato is caused by a species of *Actinomyces*. Harter and Weimer ('29) were also unable to isolate *C. batata* from pox lesions or find any evidence of zoospores, plasmodia and cysts in fixed and stained preparations.

This is the present status of *C. batata* in relation to pox. Elliott and Tabenhaus doubtless had some saprophytic plasmodial organism at hand, but whether or not it is a species of the Plasmodiophorales is obviously questionable. Fitzpatrick and Cook excluded it from this order, but Saccardo ('31) listed it among the valid species. Palm and Burk, however, implied that it is valid but stands distinctly apart from the other genera because of its method of cyst formation. Except for the presence of zoospores, *C. batata* is somewhat similar to *Leptomyxa reticelata* var. *humili*, a saprophytic proteomyxean organism which Miss McLennan ('31) found in hops. There are a large number of saprophytic, soil inhabiting organisms of this type which may become secondary invaders of roots, and unless they are carefully studied and cultured they may be readily mistaken for stages in the life cycle of plasmodiophoraceous species.

BIBLIOGRAPHY: EXCLUDED GENERA

Adams, J. F. 1929. Phytopath. 19: 179.

Anony. 1924. U. S. Dept. Agr. Pl. Dis. Rept. Suppl. 34. 1926. *Ibid.* 45.

Barre, W. H. 1910. South Carolina Exp. Sta. 23 Ann. Rept., p. 23.

Cook, W. R. I. 1933. Arch. Protistk. 80: 179.

Duggar, J. F. 1897. U. S. Dept. Agr. Farm. Bull. 26.

Elliott, J. A. 1916. Sci. n. s. 44: 709.

Fitzpatrick, H. M. 1930. The lower fungi—Phycomycetes. New York.

Halsted, B. D. 1890. New Jersey Agr. Exp. Sta. Bull. 76.

———. 1892. New Jersey Agr. Exp. Sta. 12th Ann. Rept. p. 260. 1896, *Ibid.* 17th Ann. Rept. p. 39.

Harter, L. L. 1916. U. S. Dept. Agr. Farm. Bull. 711.

———, and J. L. Weimer. 1929. U. S. Dept. Agr. Tech. Bull. 99.

Maire, R., and A. Tison. 1909. Ann. Mycol. 7: 226.

Manns, T. F., and J. F. Adams. 1921. Delaware Agr. Exp. Sta. Bull. 129: 18. 1922, *Ibid.* 133: 26. 1924, *Ibid.* 135: 25. 1925a, *Ibid.* 139: 24. 1925b, *Ibid.* 141: 24. 1926, *Ibid.* 147: 29.

McLennan, E. I. 1931. Australian Journ. Exp. Biol. 8: 9.

Palm, B. T., and M. Burk. 1933. Arch. Protistk. 79: 271.

Poole, R. F. 1922. New Jersey Agr. Exp. Sta. Bull. 356.

———. 1924. New Jersey Agr. 6: 16.

———. 1925. Phytopath. 15: 287.

Price, R. H. 1895. Texas Agr. Exp. Sta. Bull. 36: 309.

Prowazek, S. 1905. Arb. Kais. Gesundheit 22: 396.

Rietschel, P. 1936. Arch. Protistk. 86: 319.

Saccardo, P. A. 1931. Sylloge fungorum 25: 16.

Tabenhaus, J. J. 1918. Jour. Agr. Res. 13: 137.

Townsend, C. O. 1899. Maryland Agr. Exp. Sta. Bull. 60.

Wilcox, E. M. 1906. Alabama Agr. Exp. Sta. Bull. 135.

Chapter V

Phylogeny and Relationships of the Plasmodiophorales

Historical

The phylogeny and relationships of the Plasmodiophorales have been the subject of great interest and discussion among mycologists and protozoologists during the past half century because species of this order possess certain developmental stages which are similar to those of the Myxomycetes, Proteomyxa, and other Protozoa, and the simple fungi. Because of inadequate data relative to the order itself as well as to the groups with which it appears to be related, these discussions have been largely speculative, and a review of the literature shows that but few of the workers have agreed on the systematic position of the Plasmodiophorales.

Woronin ('78) stated that *Plasmodiophora* stands closest to the Myxomycetes but differs by the lack of a true sporangium and by its parasitic mode of life. In every other way, in his opinion, it resembles most closely the myxochytridiales. De Bary ('84) described *P. Brassicae* as a doubtful member of the Myxomycetes, but Zopf ('84) established a separate family, Plasmodiophoraceae, for *Plasmodiophora* and *Tetramyxa* under the zoosporic group of the Monadineae next to the Gymnococcaceae. He nonetheless included the Monadineae in the Myxomycetes, and his exclusion of the latter group from the fungi in 1890 suggests that he did not regard the Plasmodiophoraceae as true fungi. A year later Lankester incorporated the Monadineae, *Plasmodiophora* and *Tetramyxa* in a new class, Proteomyxa, of the protozoa. As noted elsewhere, Schroeter ('86) ignored Zopf's family and created a new order, Phytomyxini, with one family, Phytomyxaceae, to include *Plasmodiophora, Phytomyxa,* and *Sorosphaera* and placed it next to the Myxogastres. In 1897 he placed the Phytomyxinae between the Acrasiae and Myxogastres and pointed out that because of its free spores *P. Brassicae* stands close to the Acrasiae but differs principally from this group by its true plasmodium, zoospore stage, and intramatrical habit of life. Tubeuf and Smith ('97), however, excluded *Phytomyxa* from the Phytomyxinae and described *Plasmodiophora, Tetramyxa,* and *Sorosphaera* as pathogenic slime-fungi. Schroeter's disposition and viewpoint was supported by Lotsy ('07) and Pavillard ('10) who regarded *Plasmodiophora* as a myxomycete which has retrogressed because of its parasitic mode of life. Pavillard in particular stressed the presence of an initial flagellate stage as the chief indication of relationship between the two groups.

This viewpoint was severely criticized by Maire and Tison ('09). After a careful cytological study of *Sorosphaera,* they refuted Pavillard's claim and expressed the opinion that the Plasmodiophoraceae constitute an entirely distinct group, intermediate between the Sporozoa and Myxomycetes and derived more or less directly from the Flagellata. They further pointed out that although the type of nutrition of the Plasmodiophoraceae is plant-like, while the absence of cellulose and the presence of chitin in the spore membrane are animal characteristics. Later ('11), however, they emphasized the close resemblance of *Ligniera* to *Woronina polycystis* and postulated that this genus may have been derived from *Woronina*-like ancestors through the disappearance of sporangiosori. Maire and Tison thus concluded that the origin of the Plasmodiophorales should be sought in the neighborhood of the Chytridiales, as this order was interpreted at that time. Winge ('13) likewise maintained that "the relationship of the Plasmodiophoraceae with the holocarpic Chytridiaceae is beyond doubt," and pointed out that certain species of *Synchytrium, Asterocystis, Rhizomyxa, Sorolpidium, Woronina,* and *Pyrrhosorus* occupy intermediate positions and represent transition forms between the two groups. Stevens ('13, '25) included the Plasmodiophorales as the first order under the Myxomycetes. Maire and Tison were supported by Schwartz ('14) who stated that the differences between the Plasmodiophoraceae and Myxomycetes are too great to be accounted for by the former's parasitic mode of life. Although he regarded the two groups as related, Schwartz, nonetheless, believed that the Plasmodiophoraceae should form a separate order intermediate between the Myxomycetes and Chytridiales. Jahn ('14), Cavers ('15), and Pascher ('18) concurred in general with the views of Winge and Schwartz. In reviewing Schwartz's paper, Jahn stated that the Plasmodiophorales have little in common with the Myxomycetes and are closely related cytologically with the Chytridiales. He excluded the order entirely from the Myxomycetes in 1928. Nawaschin ('24) asserted that *P. Brassicae* has nothing in common with the Myxomycetes as far as nuclear structure is concerned and advocated its inclusion among the non-amoeboid type of Protista. Cavers ('15) stressed the relationship of the Plasmodiophoraceae and chytrids and believed that *Sorolpidium* may possibly be a connecting link between this family and the *Synchytriaceae.*

The view that the Plasmodiophoraceae are closely related to the Chytridiales has been rather widely accepted. Gaumann ('26) and Gaumann and Dodge ('28) included the Plasmodiophoraceae with the Woroninaceae, Olpidiaceae, and Synchytriaceae in a special group, the Archimycetes, apart from the Phycomycetes. They accordingly linked the Plasmodiophorales with Fischer's earlier-named Myxochytridiales. Kniep ('28) regarded them as fungi, and while admitting that they may perhaps be included in the Chytridiales, he said that the last word on their exclusion from the Myxomycetes had not been

spoken. Fitzpatrick ('30) was the first to definitely include this family in the Chytridiales next to the Woroninaceae and Synchytriaceae, and maintained that they have more in common with these two chytridiaceous families than with the Myxomycetes. In so doing, however, he did not imply a close relationship. Fitzpatrick expressed the opinion that the Synchytriaceae, Woroninaceae, and Plasmodiophoraceae "have arisen more or less in parallel from yet more primitive protozoa and wholly independent of the Myxogastres." Wettstein ('35) also included the Plasmodiophorales among the chytrids next to the Synchytriaceae and stated that their cytology as well as the presence of chitin in the walls indicates a close relationship. Cadman ('31) and Bessey ('35), however, believed that the Plasmodiophorales show a closer affinity to the Myxomycetes, and the former worker listed them as a sub-group of the Myxomycetes. Martin ('36) listed the Plasmodiophorales as the lowest order of the Phycomycetes but distinct from the so-called myxochytridiales.

On the other hand, Gwynne-Vaughan and Barnes ('26, '37) maintained that the Plasmodiophorales and Myxomycetes are not fungi and have doubtless arisen from lower forms along independent lines. Cook ('26) agreed in general with these mycologists and expressed the view that "it seems very desirable to keep the Plasmodiophorales quite separate from the Chytridiales and other fungi. If there is any relationship, it is most likely through the Mycetozoa." In 1928, however, he held that the Plasmodiophoraceae and Myxomycetes originated from a proteomyxean complex through the Lobosa and more specifically *Arcella vulgaris* and *Amoeba muscicola* and diverged at slightly different points. On the basis of the type of nuclear division in the vegetative phase, Cook believed that the Plasmodiophorales diverge from the *Amoeba* series at a more distant point than the Mycetozoa. Later ('33) he asserted that "no close relationship with either the fungi or protozoa is probable," and that the Plasmodiophorales "represent an independent group having their origin in the Proteomyxa." Cook thus revived and supported the earlier views of Zopf, Delage and Herouard, Lankester, and others on the relationship of the Plasmodiophorales to the Proteomyxa.

Zoologists also have asserted their claims to the Plasmodiophorales and included this order as a subclass of the Myxomycetes among the Protozoa, particularly the Rhizopoda. Most protozoologists, however, have continued to use Schroeter's term, Phytomyxinae, for the group, although it has been evident since the beginning of the present century that *Phytomyxa*, the genus after which Schroeter named the order and family, is no longer tenable and relates to what are now known as bacteria and mycorrhizal fungi. Protozoologists, furthermore, have ignored the discovery and presence of zoosporangia and biflagellate, heterocont zoospores in six genera of the Plasmodiophorales and have adhered to the older, outworn conceptions regarding these organisms. Delage and Herouard ('96) followed Zopf's disposition of *Plasmodiophora* and *Tetramyxa* by including them with *Gymnococcus*, *Pseudospora*, etc., in the zoosporic Proteomyxa or Monadiniae—their first class under the Rhizopoda. Doflein ('01) listed the Mycetozoa as the fifth and last class of the Rhizopoda next to the Foraminifera and divided it into two subclasses of equal rank, the Protomyxidea and Mycetozoidea. *Plasmodiophora* and *Tetramyxa* were placed in the zoosporic group of the Protomyxidea, a classification which corresponds essentially with that of previous protozoologists. In subsequent editions of his text-book ('09, '11, '16) Doflein placed the Mycetozoa next to the Radiolaria and limited the sub-class Protomyxidea to what are now generally known as the azoosporic and zoosporic Proteomyxa. For the plasmodiophoraceous genera he adopted Schroeter's Phytomyxinae, made it a sub-class of the Myxomycetes, and placed it between the Acrasiae and Myxogastres. In the sixth edition, however, which was rewritten by E. Reichenow, the Phytomyxine and Acrasiae were excluded from the Mycetozoa proper and were discussed merely as an "anhang" to this class.

Lankester ('85, '09) and Hartog ('06, '09, '22, '36) also included *Plasmodiophora* and *Tetramyxa* in the Proteomyxa along with *Vampyrella, Gymnococcus, Pseudospora*, etc. Hartog, however, assigned *Plasmodiophora* together with *Protomyxa, Vampyrella*, and *Serumsporidium* to the non-flagellate or azoosporic Myxoidea. In 1909 Calkins referred to *P. Brassicae* as a mycetozoan and later ('33) included the Phytomyxida (Phytomyxinae) as an order in the Myxomycetes. He believed that the lack of peridia and capillitia in the plasmodiophoraceous species is due to their parasitic mode of life. Minchen ('12, '21) was uncertain of the relationships of the Phytomyxinae and merely discussed them as border-line organisms in relation to the Sarcodina. Hertwig ('19) listed the Mycetozoa as the fifth class of Protozoa and of equal rank with the Rhizopoda and included *Plasmodiophora* among the Myxomycetes. Galiano (1921) also grouped the Phytomyxinae as a suborder of the Myxomycetes, while Rumbler ('23–'25) reverted to Doflein's ('09, '11) classification. Kudo ('31, '39), on the other hand, included the Phytomyxinae directly in the Myxomycetes.

It is evident from this review that several points of origin and lines of development and relationship have been emphasized for the Plasmodiophorales. These suggested relationships involve the Myxomycetes, Chytridiales, Protozoa, Sporozoa, and Proteomyxa. The evidence in support and against these relationships will now be considered in greater detail.

Plasmodiophorales and Myxomycetes

Inasmuch as the belief that the Myxomycetes and Plasmodiophorales are closely related is rather widely held, this view will be presented in considerable detail. Proponents of this view have stressed the presence of a large multinucleate plasmodium and

anteriorly uniflagellate zoospore in both groups as evidence that they have originated from a common ancestor. Considerable significance has also been attached to the reports that the plasmodia of *Spongospora* (Kunkel, '15) and *Plasmodiophora* (P. M. Jones, '28) can live outside of the host and may be cultivated on synthetic media like those of the Myxomycetes. Careful analysis of Kunkel's paper, however, shows that the saprophytic plasmodia which he described apparently do not relate to *Spongospora* at all, because at maturity they form stalked *Dictyostelium*-like sorocarps instead of spongy cystosori. Likewise, the peculiar and abnormal life cycle described by P. M. Jones for *P. Brassicae* suggests that he may have been studying some other plasmodial organism instead of *Plasmodiophora*. It thus remains to be seen whether or not the plasmodium of the Plasmodiophorales can be cultivated saprophytically outside of the host.

As to the mode of nutrition, data are accumulating which suggest that it possibly may be very similar in both groups. The zoospores, amoebae, the plasmodia of the Myxomycetes are capable of engulfing food particles, digesting them, and discarding the extraneous waste material. While this type of nutrition is not particularly evident in the Plasmodiphorales, claims have nonetheless been made that the plasmodium at least engulfs starch grains and masses of host protoplasm. According to Woronin, Nawaschin, Prowazek, and Lutman, starch grains may often be found in the folds and vacuoles of the plasmodium of *P. Brassicae*. Nawaschin ('99), Favorsky, and Henekel did not believe these had been engulfed, but Woronin, Eyeleshymer, and Lutman nevertheless inferred that the plasmodium feeds on these grains. Maire and Tison ('11) likewise reported that the small plasmodia of *Ligniera Junci* may engulf algal cells. The zoospores of some species also appear to be capable of taking in solid bodies, but how generally it occurs is not known. In *Polymyxa graminis* Ledingham reported that the pseudopods of amoeboid zoospores may flow around and engulf small objects.

The evidence of relationship on the basis of similarity in zoospore structure is not particularly convincing in light of recent discoveries. Until 1934 it was believed that the zoospores of the Plasmodiophorales were like those of the Myxomycetes in having one anterior flagellum, but since that time it has been clearly shown that the zoospores of six genera of the former group are anteriorly biflagellate and heterocont. Further study will doubtless show this to be true in the remaining genera of the Plasmodiophorales also. The structure of the zoospores and the number, position, and relative lengths of the flagella are very significant phylogenetically, and it would seem offhand that the presence of biflagellate, heterocont zoospores in the Plasmodiophorales separates this order very sharply from the Myxomycetes. It must be remembered, however, that although the majority are uniflagellate, zoospores with two flagella are not uncommon in the Myxomycetes also. De Bary ('84) and Vouk ('11) early noted zoospores with two flagella, and since that time numerous reports of similar zoospores have appeared. Gilbert ('27) found that 25 per cent of the zoospores of *Stemonitis fusca* are biflagellate, and his figures *1e* and *1f* show that one of the flagella is considerably shorter. Similar zoospores have been subsequently described and figured by Smith ('29) for *Dictyae thalium plumbeum*, by Howard ('31) for *Physarum polycephalum*, and by Sinoto and Yuasa ('34) and Yuasa ('35) for *Physarella oblonga*, *Fuligo septica*, and *Comatrichia longa* var. *flaccida*. In the latter species 13 per cent of the zoospores were biflagellate, and in rare cases triflagellate. As is shown in figures 2 to 5, Plate 17, the flagella are of equal as well as of unequal length. Stosch ('35) also found biflagellate zoospores in *Didymium eunigripes*, *D. xanthopus*, *D. squamosum*, *D. difforme*, *Physarum cinereum*, *P. nutans*, *Trichia favoginea*, *Comatrichia nigra*, and *Lycogola epidendrum*.

In most species which normally have uniflagellate zoospores, bi- and multiflagellate cells are usually the result of unequal or incomplete cleavage, and are consequently large and bi- or multinucleate. Such does not appear to be true of the zoospores shown in figures 2 to 6, Plate 17, since there is but one nucleus present regardless of the number of flagella and the size of the zoospore. A more fundamental cause may perhaps be operating in these cases. Of particular interest in these figures are the basal bodies upon which the flagella are oriented. In *Ceratiomyxa fruticulosa* var. *flaccida*, *Physarella oblonga*, and *Fuligo septica*, they are double regardless of whether one or more flagella are present. E. A. Bessey, Professor of Botany, Michigan State College, believes that this double condition may perhaps be significant phylogenetically. In correspondence with the author concerning these zoospores, he asks: "Are these two granules homologous to the basal granules found in algae and . . . . sperm cells of mosses or ferns, where each flagellum arises from such a granule? Then do the planocytes with but one flagellum represent cases where there has been a loss of one flagellum in progressive evolution from a normally biflagellate condition, and do the biflagellate cells of these slime molds represent the ancestral condition which has not been completely lost in this group? In the Plasmodiophorales, which are probably closely related to the slime molds, the biflagellate condition has not yet been lost, though one flagellum is smaller than the other." Bessey thus suggests that the presence of a second basal granule in uniflagellate zoospores may possibly be a relic of the biflagellate condition and that the Plasmodiophorales are more primitive than the slime molds. However, it remains to be seen how general the double condition is in uniflagellate zoospores. Jahn ('04), Wilson and Cadman ('28), and Cadman ('31) figured and described only one basal granule, while Cotner ('30) and Stoseh reported the presence of several bodies at the base of the flagellum. Sinoto and Yuasa's accounts of the presence of two basal

bodies in the Mycetozoa have accordingly not been universally confirmed. Jahn ('36) severely criticized the belief that the presence of two flagella are of much significance, questioned the presence of more than one basal granule, and regarded all biflagellate zoospores as abnormal.

In the Plasmodiophorales little is known about the blepharoplast and its composition. Terby ('24a) and Cook and Schwartz ('30) figured only one blepharoplast in the uniflagellate zoospores of *P. Brassicae*, but later Terby ('24b) reported that the blepharoplast may divide and form two bodies in the incipient spore. Neither Ledingham ('34, '35) nor Couch, *et al.* ('39), showed basal granules in their figures of the biflagellate zoospores of *Plasmodiophora*, *Spongospora*, and *Octomyxa*. In *Polymyxa*, on the other hand, Ledingham ('39, p. 42) figured the two flagella attached directly to the nuclear membrane without the presence of blepharoplasts or basal granules. It is thus obvious that but little is known about the number of basal granules in the zoospores of this order and their relations to the flagella. Nevertheless, Bessey's suggestion concerning the significance of basal granules and the occasional occurrence of biflagellate zoospores in the slime molds is very stimulating and merits further investigation.

Turning now to other differences within the two groups, it may be noted that sporangia and capillitia of the type found in the slime molds are lacking in the Plasmodiophorales. As has been noted before, mycologists and protozoologists have regarded this reduction as due to the parasitic mode of life adopted by the Plasmodiophorales. Cook ('33) suggested that the membrane around the cystosori in certain plasmodiophoraceous genera, *Sorodiscus*, *Sorosphaera*, etc., may be looked upon as equivalent to the sporangium wall of the Myxomycetes. However, there is considerable doubt about the presence of a soral membrane in these genera. The Myxomycetes, on the other hand, lack sporangiosori and thin-walled evanescent zoosporangia, which have recently been shown to occur in most genera of the Plasmodiophorales. These zoosporangia may arise directly from zoospores which have entered the host or later from small or large, segmented, vegetative plasmodia. These differences—lack of thin-walled, intramatrical zoosporangia in one group and specialized sporangia and capillitia in the other—are of fundamental significance, in the author's opinion, and are difficult to explain wholly by differences in mode of life.

Other developmental phases and cytological differences between the two groups are to be noted here. Schizogony of the young plasmodium has been described in most genera of the Plasmodiophorales, but it appears to be lacking in the Myxomycetes. At least, no conclusive evidence of its occurrence has yet been presented. Furthermore, neither the so-called "promitotic" nuclear divisions nor a marked "akaryote" stage, which are reported to be characteristic developmental phases of the Plasmodiophorales, have been found in the Myxomycetes. Whether or not these differences alone are of much phylogenetic significance, however, is questionable.

Comparisons of the two groups on the basis of sexuality, alternation of generations, time and place of meiosis, etc., are difficult to make at present, because so little is known about these processes in the Plasmodiophorales. In the Myxomycetes also there is considerable disagreement among workers about these developmental phases. As far as is now known the resting spores of the slime molds usually form more than one zoospore in germination, and these in turn divide once to several times before becoming gametes. In the Plasmodiophorales, on the other hand, it is claimed that only one zoospore is formed, which functions directly as a gamete without dividing. Cook ('33) emphasized this distinction and stated that it is "the chief difference between the two groups." In light of data in the literature, this statement is obviously open to criticism. Maire and Tison, and Horne found an additional or third mitosis after the two meiotic divisions in *Sorosphaera* and *Spongospora*, respectively, whereby binucleate spores were occasionally produced. Lutman and Terby also figured binucleate spores in *P. Brassicae* and believed that these arise as the result of division of the spore nucleus. It is not improbable that such spores form more than one zoospore or gamete. In addition to such spores, unusually large multinucleate ones have been found in several genera, and it is not unlikely that they also give rise to several motile cells in germination. Likewise, Cook's assertion "that division of the swarm cells does not take place in the Plasmodiophorales prior to fusion" is rather dogmatic and premature in light of our meager present-day knowledge of the behavior of the zoospores in this order. They have never been cultured with certainty outside of the host, and very little is known about their behavior within the host cells. Cook's assertion is furthermore contradicted by Massee's (Pl. 10, fig. 10), Osborn's, Horne's and Fedorintschik's accounts of the multiplication of amoebae or gametes in *Spongospora* and *Plasmodiophora* by equal division and budding. In maintaining that the gametes are the direct products of the resting spores, Cook further contradicted his own and Schwartz's ('30) earlier assertion that the gametes of *P. Brassicae* are produced in thin-walled zoosporangia or gametangia. The origin and method of formation of gametes in the Plasmodiophorales are thus somewhat doubtful at present, and it seems premature to make definite comparison between the two groups on this basis.

Fusion in pairs of isomorphic amoeboid and flagellate gametes has been reported to be characteristic of both groups, but as noted elsewhere actual fusion has so far been seen very seldom in the Plasmodiophorales. The respective gametes are alike in size and structure in both groups, but in the Myxomycetes certain other differences between gametes of the opposite sex have been reported. According to Abe ('34) the male gamete loses its flagellum as it

flows into the female, and its nucleus migrates towards that of the female gamete. Furthermore, the latter gamete carries a positive charge and has a low oxidation-reduction potential, while the male gamete is the opposite in these respects. Kambly ('39), however, was unable to confirm these results of Abe, and found no marked physiological differences between swarm cells of various species. Gilbert ('35) and Stosch ('37) likewise reported that the male gamete may be distinguished during fusion by the migration of its nucleus toward that of the female. Such differences have not been reported for the Plasmodiophorales as far as the author is aware. In the Myxomycetes the gametes fuse by their posterior ends, while in the Plasmodiophorales, according to Cook ('33), they fuse at the anterior ends. However, so little is known about gametic union in this order that it is premature to regard the latter type of fusion as characteristic of the Plasmodiophorales.

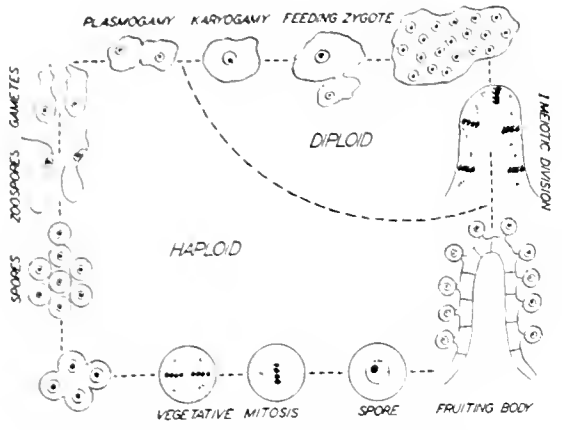
Comparison of the two groups on the basis of time and place of sex segregation is also impossible at present, because little is known about sexuality in the Plasmodiophorales. No monospore cultures or infections have yet been made to determine whether the species are homo- or heterothallic. If, as Cook ('33) maintained, the gametes are the direct product of uninucleate spores and no division occurs in amoebae and zoospores, sex segregation obviously takes place during one of the meiotic divisions before or during sporogenesis. Otherwise, it is phenotypically determined in the haploid generation, and the species are accordingly haplosynoecious. In the Myxomycetes also, there are but few data relating to sex segregation. Skupienski ('17) believed that in *D. difforme* it occurs during one of the divisions in the zoospores. Miss Clayley reported its occurrence at the second meiotic division in the zoospores of *D. nigripes*. Schünemann confirmed her report of haplophenotypic sex segregation in this species and described *D. nigripes* as haplomonoecious. Miss Cadman, however, noted no differences, morphological or physiological, between the gametes in *Reticularia* and *D. nigripes* and concluded that no sex segregation is necessary or takes place in these species. Stosch, on the other hand, implied by his statement concerning crosses in *D. eunigripes* that sex is genotypically determined.

As to the time and place of karyogamy, meiosis, and alternation of haploid and diploid generations in the Plasmodiophorales, a detailed account of these subjects has been given in Chapter III. As is evident from this description, the majority of workers have assumed that the isomorphic gametes fuse in pairs, after which karyogamy soon occurs. Nuclear fusion in the zygote thus initiates the diploid phase which includes the plasmodial stage up to the last two nuclear divisions preceding or during cleavage where reduction occurs. Plasmogamy and karyogamy are accordingly not followed at once by meiosis. The haploid phase includes the cystosori, spores, zoospores, and gametes, according to this viewpoint. However, as noted before, exceptions to this view have been presented by Prowazek, Osborn, Horne, Webb, and Whiffen.

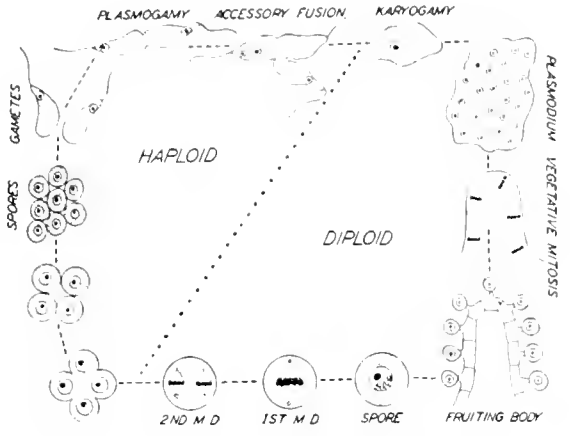
In the Myxomycetes likewise there is considerable disagreement and controversy concerning karyogamy, meiosis, and alternation of generations. Much of the controversy about meiosis hinges upon the question of whether one or two divisions occur prior to spore formation in the fruiting bodies. Strasburger ('84), A. Lister ('93), Rosen ('93), Harper ('00), Jahn ('07–'36), Kränzlin ('07), Gilbert ('35), and Stosch ('35, '37) found only one, while Wilson and Cadman ('28), Cadman ('31), and Schünemann ('35) reported two divisions. In contrast it may be noted here that most workers on the Plasmodiophorales are in agreement that two divisions precede spore formation. However, in order to draw comparisons between the two groups with respect to meiosis, it is essential to outline briefly the differences of opinion concerning this question in the Myxomycetes.

In the Exosporae, Olive ('07a) found stages resembling synapsis in the young spores of *Ceratiomyxa* and later ('07b) on observing pairing and fusion of nuclei in the pillars, concluded that the two mitoses in the spores of this genus are meiotic. Olive's conclusions on pairing and fusion of gametic nuclei were confirmed in general by Jahn ('07) who, however, held that these processes occur earlier as the plasmodium creeps out of the wood. On the other hand, he refuted Olive's contention that meiosis occurs in the spore and claimed instead that the two divisions which precede cleavage are reductional. The incipient uninucleate spores are accordingly haploid. Jahn ('08) reasserted his observations on nuclear pairing and fusion, but maintained that only one, instead of two, division occurs prior to cleavage. This division is heterotypic, according to Jahn, and reduction is thus accomplished by one division. The plasmodium is formed by the fusion or coalescence of numerous haploid myxamoebae, the nuclei of which divide mitotically several times in the plasmodium. Karyogamy is accordingly delayed until the plasmodium creeps out to fructify. In 1911, however, Jahn concluded that his previous observations on nuclear pairing and fusion in the mature plasmodium were incorrect and that the appearances of karyogamy were the results of nuclear degeneration. His observations of endosporus species led him to the belief that nuclear fusion follows plasmogamy of amoebae. Jahn ('11, '33, '36) nonetheless persistently adhered to his early view that meiosis occurs during the last division before cleavage, as is shown in text-figure 12. Gilbert ('35), on the other hand, confirmed Olive on meiosis in the spore and in addition showed that the haploid motile gametes fuse posteriorly in pairs to initiate the plasmodium (text-figure 13). He also found that karyogamy follows plasmogamy within 24 hours, thus refuting Olive's observations but confirming Jahn's later view.

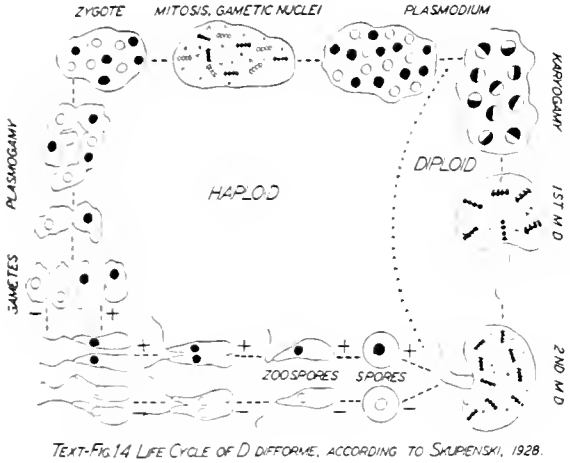
In the Endosporeae, Jahn ('07) reported the same type of nuclear pairing and fusion in the young fruit-



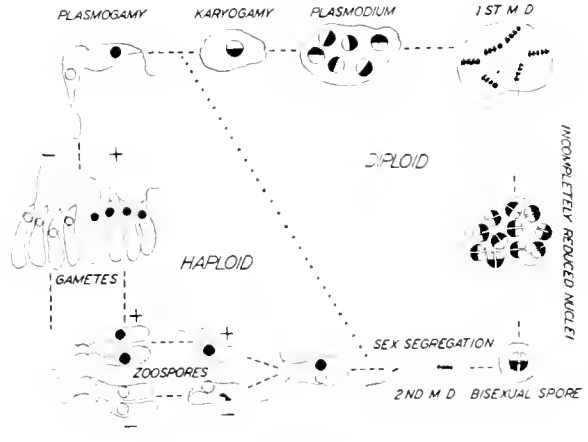
TEXT-FIG 12 LIFE CYCLE OF CERATIOMYXA, ACCORDING TO JAHN, 1911-36.



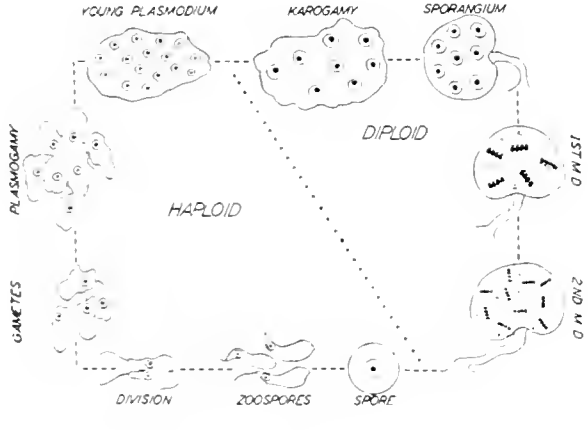
TEXT-FIG 13 LIFE CYCLE OF CERATIOMYXA, ACCORDING TO GILBERT, 1935.



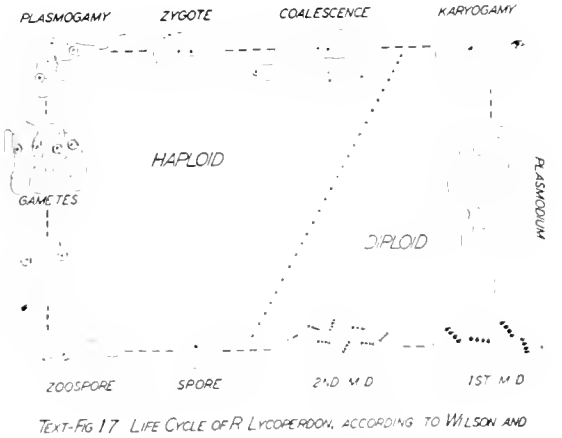
TEXT-FIG 14 LIFE CYCLE OF D DIFFORME, ACCORDING TO SKUPIENSKI, 1928.



TEXT-FIG 15 LIFE CYCLE OF D DIFFORME, ACCORDING TO CLAYLEY, 1929



TEXT-FIG 16 LIFE CYCLE OF D NIGRIPES, ACCORDING TO SCHUNEMANN, 1930.



TEXT-FIG 17 LIFE CYCLE OF R LYCOPERDON, ACCORDING TO WILSON AND CADMAN, 1928

Text-figure 12-17

ing bodies of *Amaurochaete, Reticularia, Trichia, Stemonitis,* and *Didymium.* In these genera karyogamy is followed by synapsis, and as the spores are delimited, one mitosis, the heterotypic division, occurs. This first meiotic division is followed by a long rest period of the spore, and the second or homeotypic division is delayed until the first mitosis in the germinating spore, according to Jahn. Similar observations were reported by Kränzlin ('07) and Vouk ('11) for species of *Trichia* and *Arcyria,* but these were later found to be incorrect by Jahn in 1911. For the first time in the Myxomycetes he found that haploid myxamoebae of *Physarum didermoides* fuse in pairs to form the zygote. Plasmogamy is followed shortly by karyogamy. The diploid zygote may engulf haploid amoebae, with the result that haploid and diploid nuclei may be found in the young plasmodia. Likewise, zygotes may fuse with each other to form larger plasmodia, but fusion of the diploid nuclei does not occur. Meiosis takes place during the last division in the young sporangium and is not followed by a homeotypic division. Jahn ('33) reported the same type of meiosis in *Badhamia utricularis,* and subsequently persisted in this view on the time and nature of reduction division in the Exosporeae and Endosporeae.

Pinoy ('08) concluded from his culture experiments that *Didymium nigripes* is heterothallic and forms + and — myxamoebae which in turn give rise to + and — plasmodia. Sporangia are formed only when both types of plasmodia are mixed. It is not certain that Pinoy used monospore cultures, and because of this his results have been seriously questioned by Kniep ('28) and Schünemann ('30). Skupienski ('17–'28) also reported heterothallism in *D. nigripes* and *D. difforme.* In 1928 he asserted that the spores of *D. difforme* are unisexual and that no sporangia will develop in monospore cultures. According to him the plasmodium arises by the fusion of two myxamoebae of opposite sex (text-figure 14). Other myxamoebae may unite with the zygote, but the gametic nuclei remain separate and divide mitotically in the young plasmodium. The daughter nuclei later unite in pairs and fuse in the older plasmodium, while those which fail to find partners degenerate. Meiosis occurs during the last two divisions in the sporangium, according to Skupienski.

In the same year Wilson and Cadman showed in *Reticularia Lycoperdon* that haploid motile gametes fuse in pairs by their posterior ends to form a zygote (text-figure 17). Other gametes may coalesce with the zygote, but their nuclei divide amitotically, degenerate, and are digested by the zygote. Karyogamy of the gametic nuclei follows shortly after the coalescence with the non-functional gametes, and meiosis occurs during the last two divisions in the sporogenic protoplasm. Miss Clayley ('29) refuted Skupienski's contention of heterothallism in *D. difforme,* showed that the spores are bisexual, and secured sporangia in monospore cultures (text-figure 15). She also found that plasmogamy takes place between motile gametes instead of myxamoebae, as claimed by Skupienski. Schünemann likewise secured plasmodia in monospore cultures of Skupienski's own *D. difforme* and thus refuted the latter's contention of heterothallism. In *D. xanthopus,* however, neither plasmodia nor sporangia were formed in monospore cultures. In *D. nigripes,* Schünemann found that several haploid myxamoebae coalesce to form plasmodia but their nuclei remain separate until the plasmodia become older (text-figure 16). Karyogamy eventually occurs, and reduction is accomplished during the two divisions preceding spore formation. Schünemann thus concluded that a true antithetic alternation of generations occurs in *D. nigripes.* Cadman ('31), however, found that karyogamy occurs shortly after plasmogamy, and that the diploid zygote can ingest zoospores and haploid myxamoebae and coalesce with other zygotes. She nevertheless confirmed Schünemann on meiosis. In the same year Howard reported fusion in pairs of motile gametes in *Physarum polycephalum* and expressed the belief that plasmogamy is followed at once by karyogamy. Abe ('33, '34) likewise found fusion of motile gametes in *Fuligo septica, Erionema aureum, D. nigripes, P. crateriforme,* and *Stemonitis fusca.* The gametes were found to be isomorphic but differ physiologically, as has been noted previously.

In *D. nigripes,* Stosch ('35, '37) reported the discovery of two forms, *D. eunigripes* and *D. xanthopus,* which are hetero- and homothallic, respectively. In *D. eunigripes,* sexuality is well defined, while *D. xanthopus* is apogamous. *Didymium squamulosum* and *Physarum cinereum* were also reported to be apogamous, the first report of which Jahn ('36) characterized as fantastic. Jahn further refuted Stosch's report of heterothallism in *D. eunigripes* and claimed that the failure of the gametes to fuse and form plasmodia and sporangia in Stosch's monospore cultures was due to the fact that they had not gone through the encystment and rest period which are necessary before fusion occurs. For sexual species of the Didymaceae, Stosch reported that motile gametes fuse in pairs to form zygotes, which in turn fuse with other zygotes in the formation of large plasmodia. Plasmogamy of gametes is apparently followed shortly by karyogamy. Only one vegetative mitosis occurs before cleavage in the sporangium, and meiosis takes place in the spore, according to Stosch. Separation of homologous chromosomes may occur in the first and second divisions. Stosch thus supported Olive's and Gilbert's contention that meiosis occurs in the spore instead of before cleavage in the sporangium. In apogamous species, he reported that fusion may occur between amoeboid as well as motile gametes, and that instead of meiotic divisions in the spore, one or perhaps two vegetative divisions occur which are followed by amitosis.

It is apparent from this survey that there are marked differences in observations and interpretations concerning karyogamy, meiosis, alternation of generations, and sex segregation in the Myxomycetes as well as in the Plasmodiophorales. Nonetheless, certain fundamental similarities do exist, and if the

diagrams representing the life cycles of the Plasmodiophorales in Chapter III are compared with those of the Myxomycetes these similarities become more striking. Most recent workers in both groups agree that the diploid phase is initiated by the fusion of amoeboid or motile gametes and karyogamy and extends to the time of the last two nuclear divisions preceding sporogenesis during which reduction occurs, while the haploid phase includes the spores, zoospores, amoebae, and gametes. However, the presence of a zoosporangial stage in the Plasmodiophorales and the possibility that the zoosporangia may be gametangia complicates the situation, and until more is known about this developmental phase it is impossible to say how close the Plasmodiophorales and Mycetozoa are to each other.

Plasmodiophorales and Chytridiales

As has been noted in the historical review, the suggested relationship of the Plasmodiophorales with the Chytridiales involves principally the families Woroninaceae, Synchytriaceae, and certain members of the Olpidiaceae. Reports of relationship with the Synchytriaceae are based primarily on the fact that the thallus in this family functions as a prosorus and segments into a number of zoosporangia as in some genera of the Plasmodiophorales. It must be noted, however, that this thallus is haploid in the Synchytriaceae, according to Curtis, Kusano, Köhler and others, while in the Plasmodiophorales it is believed by numerous workers to be diploid. Outside of the formation of sporangiosori in both families, there is little or no further similarity. The presence of posteriorly uniflagellate zoospores and gametes in the Synchytriaceae precludes, in my opinion, any close affinity. Furthermore, the presence of a membrane around the mature thallus, lack of amoebae and naked plasmodia, and the absence of schizogony, as well as the fact that the zygotic thallus does not segment and form numerous resting spores or cystosori are other outstanding differences which are difficult to reconcile.

In certain members of the Olpidiaceae, particularly species of *Rozella,* the thallus has been described as naked, plasmodium-like, and indistinguishable from the host protoplasm. In the septigenous species of this genus, the thallus is furthermore reported to segment into numerous portions which develop into zoosporangia or resting spores. However, as the author ('42) has pointed out elsewhere, the presence of a plasmodium with this type of development has not yet been conclusively demonstrated for *Rozella.* In *Pringsheimella,* on the other hand, the evidence of segmentation of the thallus and the formation of sporangiosori is more conclusive, according to Couch's ('39) observations. Certain genera of the Olpidiaceae like *Rozella* and *Pringsheimella* have thus been described as resembling species of the Plasmodiophorales in the development of sporangiosori. On the other hand, they differ fundamentally by their posteriorly uniflagellate zoospores. The contention of Winge that *Sorolpidium Betae, Rhizomyxa hypogaea,* and *Anisomyxa Plantaginis* are transition species between the Plasmodiophorales and Chytridiales is no longer tenable, because these species have since been shown to belong to the former order. Therefore, the evidence of relationship between these two groups is very meager and inconclusive at present.

The family Woroninaceae is at present a convenient dumping ground for all holocarpic, Oomycete-like species with biflagellate zoospores, and as such is not a coherent group of closely related genera. In light of present-day knowledge it should be separated from the Chytridiales proper, which have uniflagellate zoospores. Therefore, a discussion of the relationship between the Woroninaceae and Plasmodiophorales under the present heading is in a sense contradictory. Nevertheless, it may be conveniently inserted here without offense to logic. The life cycle of some species of *Woronina,* particularly *W. polycystis,* as far as is now known, is strikingly similar to that of several members of the Plasmodiophorales, as Zopf, Maire and Tison ('11), Winge, and others have already emphasized. In light of the recent discovery of *Polymyxa* and *Octomyxa* by Ledingham, Couch, *et al.,* these similarities have become more significant and need to be emphasized again. With the purpose of so doing I have reproduced in Plate 16 the life cycle of *W. polycystis.* In this species the contents of the zoospore enters the host hypha as a naked protoplasmic mass (fig. 6–10), undergoes amoeboid changes in shape, develops into a plasmodium-like thallus as it feeds on the host protoplasm, and causes local hypertrophy (fig. 11, 12).

At maturity the thallus cleaves into segments (fig. 13, 14) which develop into zoosporangia (fig. 15, 16) and form a typical sporangiosorus. As in *Octomyxa,* the peripheral zoosporangia are usually independent with a single exit papillae, while the deeper lying ones may be confluent with a common papilla for zoospore emission. Each sporangium produces a number of biflagellate zoospores (fig. 18–21) which reinfect the host hyphae. As the culture becomes older, the thalli cleave into small segments which become the resting spores. These remain closely attached and form compact cystosori of various sizes and shapes (fig. 23–25). As in *Ligniera* and *Polymyxa,* the cystosori may be elongate, irregular, flattened, oval and almost spherical, and include a few to numerous polygonal spores, which produce zoospores in germination.

As to the structure of the zoospores of *W. polycystis,* there is, however, considerable disagreement among students of this species. Fischer described and figured them as ellipsoidal (fig. 1) with a slight indentation at one side and two unequal flagella. The shorter flagellum arises from the anterior end and extends forward in swimming, while the longer one is inserted laterally and projects backward. It must be noted, however, that Fischer's description was not applied directly to *W. polycystis* but relates to the zoospores of *Rozella, Olpidiopsis,* and *Woronina* as a group. Cook and Nicholson ('33), on the other hand,

described the zoospores as spherical (fig. 3, 4) with two anterior flagella which lash back and forth in breast-stroke fashion in swimming. These workers were non-committal as to the relative lengths of the flagella, but most of the figures show them to be equal in length. One of their figures (fig. 3), however, shows flagella of unequal length. If the zoospores are anteriorly biflagellate, as Cook and Nicholson contended, and heterocont as Fischer reported, they do not differ fundamentally from those of the Plasmodiophorales. In view of the wide differences in observations it is not altogether improbable that what is now called *W. polycystis* may relate to more than one organism or species. Further critical studies on this species are therefore highly essential.

So far schizogony has not been reported in *W. polycystis,* and nothing is known about the type of nuclear divisions in the vegetative thallus. This parasite has never been studied critically from fixed and stained material, and it is not improbable that future investigations may reveal the occurrence of schizogony and "promitotic" divisions. It should be noted in this connection, however, that the sporangia and resting spores of *W. polycystis* give a definite cellulose reaction, while those of the Plasmodiophorales do not. Furthermore, in germination the content of the zoospore enters the host through a penetration tube, leaving the empty case on the outside of the host cell as in *Olpidiopsis, Rozella,* etc. In the Plasmodiophorales the zoospores are reported to enter directly. The latter difference may not be important, but the presence of cellulose is fundamentally significant, according to present-day students of phylogeny.

The other species of *Woronina, W. glomerata, W. aggregata, W. elegans,* and *W. asterina,* are not well known, and it is difficult to compare them with the Plasmodiophorales. *Woronina glomerata* parasitizes *Vaucheria* and causes septation of the filaments without hypertrophy. It forms both sporangio- and cystosori, but the resting spores and sporangia are not closely aggregated and compact like in *W. polycystis.* Motile zoospores have not been illustrated, so that nothing is known about the number, position, and relative lengths of the flagella. The zoospores apparently enter the host directly, divide, according to Zopf ('94, p. 54), and form amoebae, which may in turn divide. The amoebae feed on the host protoplasm and engulf starch grains, chlorophyll granules, etc., whereby they may become quite green in color. This food is held in well-defined vacuoles, according to Scherffel ('25), and shortly before the parasite fructifies, the extraneous waste material is extruded as in typical proteomyxean species. The amoebae later unite by fine strands or pseudopods and form a reticulate plasmodium, which may completely fill the host cell. The amoebae may separate again, but at maturity the plasmodium cleaves into segments or "Theilplasmodien," each of which becomes a sorus of zoosporangia or resting spores. This division of amoebae and plasmodia is suggestive of schizogony in the Plasmodiophorales. The resting spores of *W. glomerata,* unlike those of *W. polycystis* and the Plasmodiophorales, function as zoosporangia in germination and produce numerous zoospores. Because of its type of nutrition, Zopf and Scherffel regarded *W. glomerata* as an organism with animal and fungal characteristics and included it with the zoosporic Myxozoidia or Proteomyxa. It may be noted, however, that *W. polycystis* also feeds directly upon the host protoplasm by bodily taking in globules of oil, according to Cook and Nicholson.

Except for the possession of biflagellate zoospores and an intramatrical holocarpic thallus, the other known genera of the Woroninaceae, with the possible exception of *Rozellopsis* Karling ('42b), do not appear to have much in common with the Plasmodiophorales. In the polysporangiate, septigenous species of *Rozellopsis,* the thallus has been described as naked and plasmodium-like, and undergoes segmentation to form numerous zoosporangia which become separated by cross septa in the host. Furthermore, in *R. simulans* the zoospores are anteriorly biflagellate and heterocont, according to Tokunga ('33). However, so little is known about the development and cytology of these species that it is impossible to draw further comparisons. There are nevertheless striking similarities in the development of the Plasmodiophorales and certain species of the Woroninaceae, particularly *W. polycystis,* which suggest a close relationship and common origin. Cook ('33), on the other hand, contended that these simi-

PLATE 16

Fig. 1, 2. Biflagellate zoospores. Fischer, '82.

Fig. 3, 4. Anteriorly biflagellate zoospores. Cook and Nicholson, '33.

Fig. 5, 6. Early infection stages. Fischer, l.c.

Fig. 7. Same. Cook and Nicholson, l.c.

Fig. 8–10. Amoeboid changes in shape and position of young parasite in host cell. Fischer, l.c.

Fig. 11, 12, 13, 15. Successive stages in development of the parasite and its cleavage into a sporangiosorus. Note local hypertrophy and septation of host hypha. Fischer, l.c.

Fig. 14. Vacuolate thallus undergoing centrifugal cleavage. Fischer, l.c.

Fig. 16. Sporangiosorus. Cornu, '72. According to Couch ('39) this figure relates to *P. dioica.*

Fig. 17–20. Maturation, cleavage, and emission of zoospores from a sporangium. Fischer, l.c.

Fig. 21. Small empty sporangiosorus. Cornu, l.c.

Fig. 22. Cleavage of thallus into a cystosorus. Fischer, l.c.

Fig. 23. Mature cystosorus. Cornu, l.c.

Fig. 24. Septate, locally hypertrophied hypha of *Saprolegnia* with five cystosori of various sizes and shapes and two empty sporangiosori. Fischer, l.c.

Fig. 25. Elongate irregular cystosorus. Cook and Nicholson, l.c.

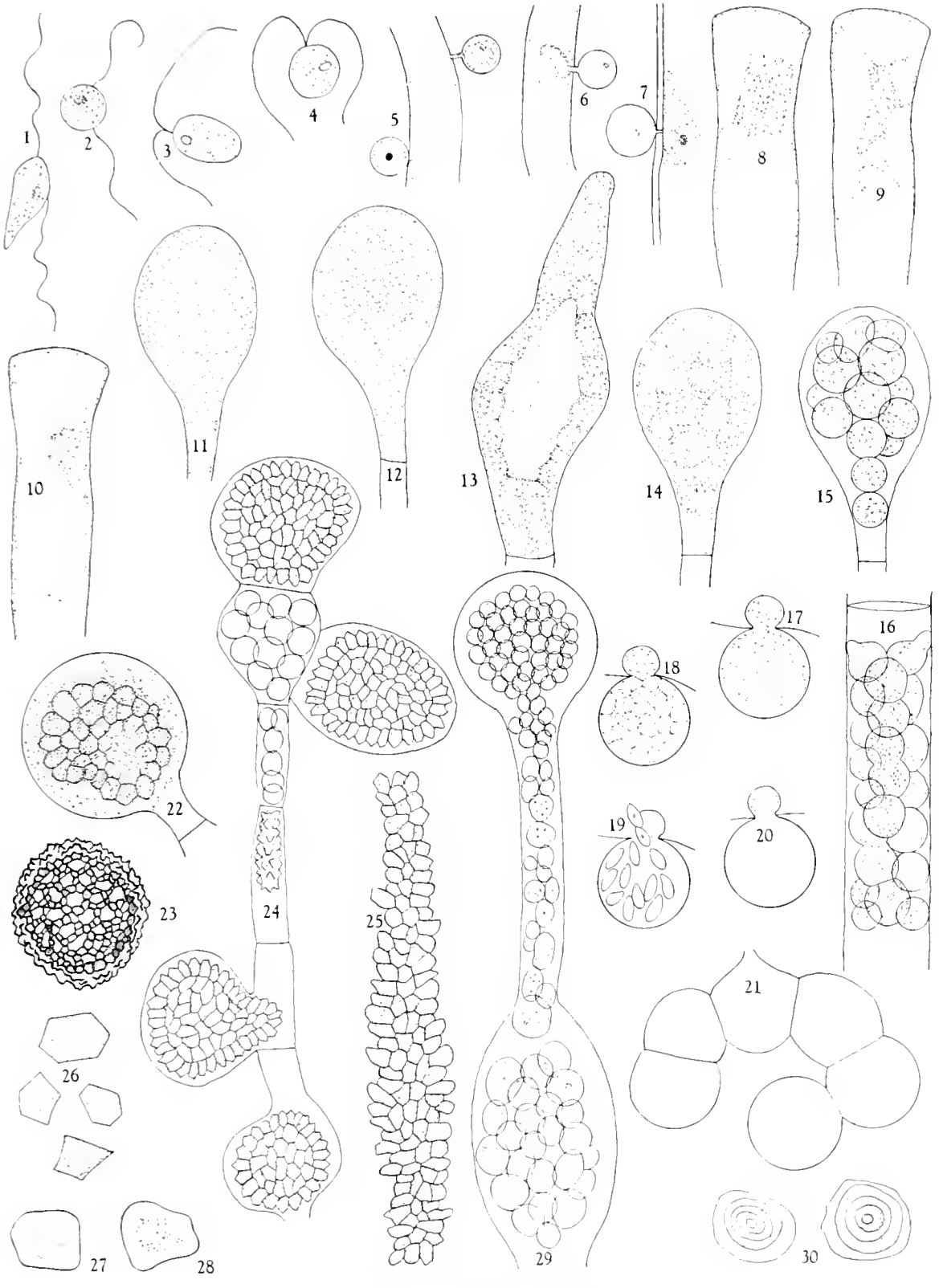
Fig. 26. Variously-shaped resting spores from a cystosorus. Fischer, l.c.

Fig. 27, 28. Thick-walled resting spores. Cook and Nicholson, l.c.

Fig. 29. Germination of cystosorus. Resting spores swelling and vesiculating to become zoosporangia. Fischer, l.c.

Fig. 30. Germination of resting spores. Cook and Nicholson, l.c.

PLATE 16



Woronina polycystis

larities are incommensurable and that the two groups have but little in common. Most of the objections raised by Cook, however, are no longer tenable in the light of more recent discoveries in the Plasmodiophorales.

Plasmodiophorales, Proteomyxa, and Other Protozoa

Inasmuch as the names Monadineae, Myxozoidia, and Proteomyxa are more or less synonymous and have been rather loosely used in the literature, a brief discussion of their terminology is essential before proceeding to the questions of relationship with and origin of the Plasmodiophorales from this group of simple organisms. The term Monadineae was first employed by Cienkowski ('65) for a number of primitive organisms whose vegetative reproductive cell develops into amoeboid or plasmodial thalli which are capable of engulfing solid food particles. Following the feeding and growing stage the thalli develop distinct membranes, discharge the extraneous food material into a large vacuole, undergo cleavage, and form zoospores or small amoebae. At the conclusion of this phase, resting spores are formed. Cienkowski divided these organisms into two groups, Monadineae zoosporeae and Monadineae tetraplastae, depending on whether zoospores or *Actinophrys*-like amoebae are produced. Many of these aquatic monadinaceous species were later included by Klein ('82) in a new family, Hydromyxaceae, but this name was not widely accepted at that time. More recently, however this family was emended by Jahn ('28), raised to ordinal rank, and included as the first order of the Myxomycetes. In 1884 Zopf gave an extended account of the Monadineae in his book on the "Pilzthiere or Schleimpilze" in which he continued Cienkowski's terminology for the whole group but changed the division Monadineae tetraplastae to Monadineae azoosporeae. The following year, however, Lankester created a new class, Proteomyxa, of protozoa to include the Monadineae of Cienkowski and Zopf as well as *Plasmodiophora* and *Tetramyxa*. In 1893 Klebs pointed out that continued use of the term Monadineae in the sense of Cienkowski would lead to confusion inasmuch as this name had previously been applied to a group of flagellates of which *Monas* is the type genus. Zopf ('94) accordingly proposed an alternate name, Myxozoidia, for Cienkowski's Monadineae. Doubtless because Zopf's paper was not published in a prominent journal, his term did not become generally known. Lankester's term was accepted by most protozoologists and has accordingly displaced the terms Monadineae and Myxozoidia in the literature on protozoa. Protophytologists, however, have continued to use Cienkowski's term. According to present-day interpretations the Proteomyxa embraces several families of incompletely known rhizopod-like species, which protozoologists include in the sub-class Rhizopoda of the Sarcodina. For the sake of emphasis and clarity, relationships with the Proteomyxa will be discussed here apart from the Protozoa in general, but such treatment does not imply that this order is to be excluded from the Rhizopoda.

As Zopf early pointed out, the life cycles of certain monadinaceous species, particularly of the family Gymnococcaceae, are similar in many respects to those of the Plasmodiophoraceae, and for this reason he included both families in the same division of the Monadineae. Subsequent studies by de Bruyne ('90), Scherffel ('25), and others have supported Zopf's observations and emphasized these similarities even more fully. As a result of such studies, some of these proteomyxean species are now known to have anteriorly biflagellate, heterocont zoospores,

PLATE 17

Physarella, Fuligo, and Didymium

Fig. 1. Anteriorly uniflagellate zoospore of *Physarella oblonga* with two "basal bodies." Sinoto and Yuasa, '34.

Fig. 2. Biflagellate heterocont zoospores of *P. oblonga* with two "basal bodies." Note tail piece at end of flagella. Sinoto and Yuasa, l.c.

Fig. 3. Biflagellate isocont zoospore of *P. oblonga* with two "basal bodies." Sinoto and Yuasa, l.c.

Fig. 4. Triflagellate heterocont zoospore of *Fuligo septica* with two short flagella attached to one "basal body." Yuasa, '35.

Fig. 5. Biflagellate heterocont zoospore of *F. septica* with "two basal bodies." Yuasa, l.c.

Fig. 6. Biflagellate heterocont zoospore of *D. Xanthopus* with several "basal bodies." Stosch, '35.

Pseudosporopsis, Amylophagus, Gymnococcus, and *Aphelidiopsis*

Fig. 7, 8. Anteriorly biflagellate heterocont zoospores of *Pseudosporopsis* sp. (*Bodo globosus*) with numerous engulfed food particles. Short flagellum extending forward. Scherffel, '25.

Fig. 9, 10. Zoospores of same with contractile vacuoles and nucleus. Scherffel, l.c.

Fig. 11. Anteriorly biflagellate heterocont zoospores of *Amylophagus algarum* with two contractile vacuoles. Long flagellum extending forward. Scherffel, l.c.

Fig. 12. Amoeboid stage of same. Scherffel, l.c.

Fig. 13. Anteriorly biflagellate heterocont zoospore of *P. rotatoriorum* with two contractile vacuoles; long flagellum extending forward. Scherffel, l.c.

Fig. 14. Anteriorly biflagellate zoospores of *Aphelidiopsis epithemiae*. Scherffel, l.c.

Fig. 15. Large plasmodium (?), *A. epithemiae*, with extraneous food material in a large central vacuole. Scherffel, l.c.

Fig. 16. Zoocysts of *A. epithemiae*. Scherffel, l.c.

Fig. 17. Deliquesced zoocysts and emerging zoospores of *A. epithemiae*. Scherffel, l.c.

Fig. 18. Eleven zoosporangia, five of which are filled with zoospores, from a single thallus of *G. Cladophorae*: extruded waste material between sporangia. De Bruyne, '90.

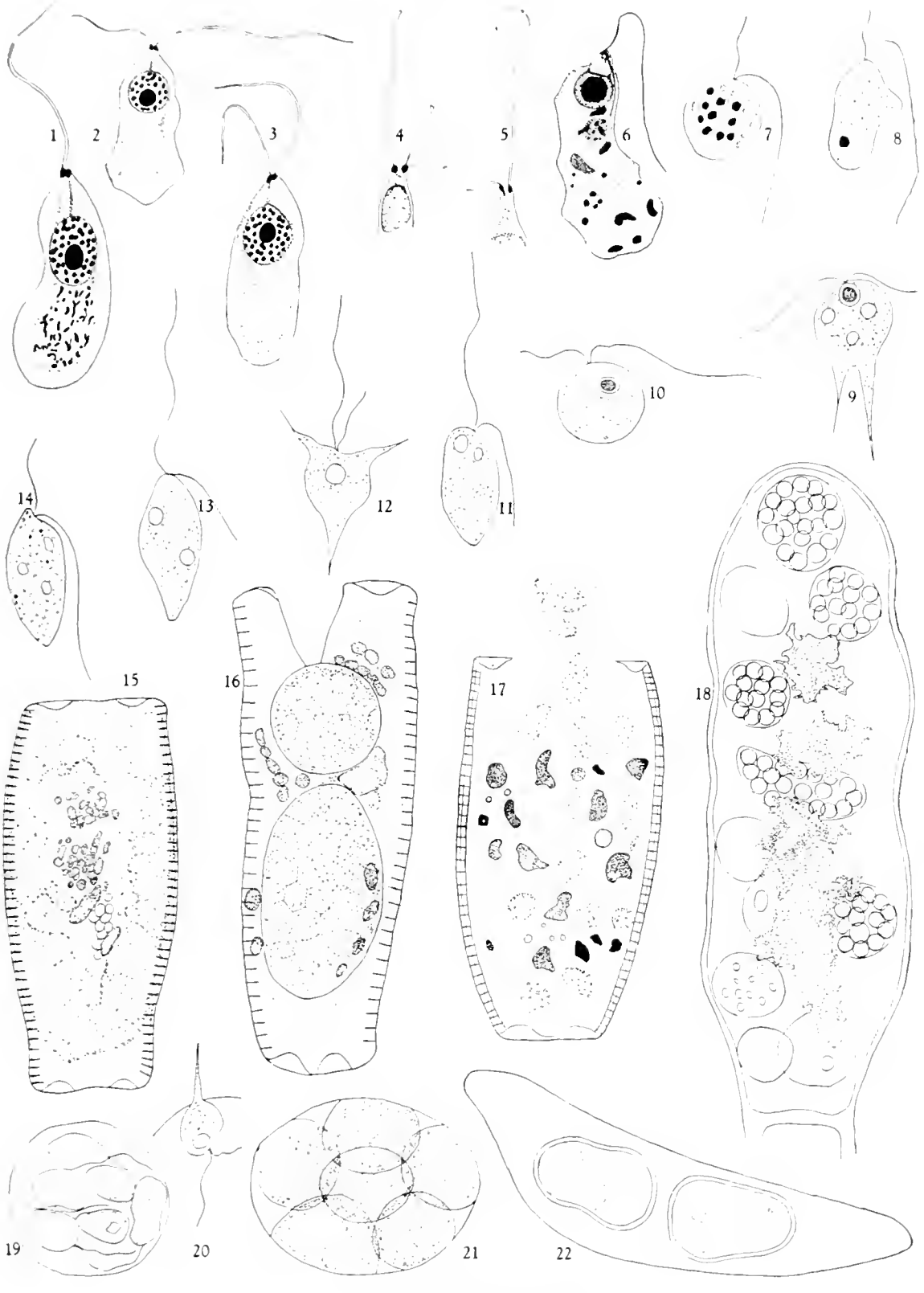
Fig. 19. Zoocyst of *A. algarum*. Scherffel, l.c.

Fig. 20. Emergence of zoospore through zoocyst wall in *A. algarum*. Scherffel, l.c.

Fig. 21. Sporocyst of *P. rotatoriorum* with six resting spores. Scherffel, l.c.

Fig. 22. Resting spores of *Aphelidiopsis epithemiae*. Scherffel, l.c.

PLATE 17



Myxomycetes. Protomyxa

naked plasmodium-like thalli, zoocysts, and sporocysts. When aggregated the latter two structures are comparable with the loose sporangio- and cystosori found in plasmodiophoraceous and woroninaceous species. *Aphelidiopsis, Gymnococcus, Pseudosporopsis* and *Amylophagus* may be taken as examples, and for the sake of more concrete comparisons drawings by de Bruyne and Scherffel of the zoospores and some developmental stages of these genera have been brought together in Plate 17. The zoospores of *Pseudosporopsis* sp. (*Bodo globosus* Stein, fig. 7–10), *Amylophagus algarum* (fig. 11–12), *P. rotatoriorium* (fig. 13), and *Aphelidiopsis epithemiae* (fig. 14), like those of the *Plasmodiophora, Octomyxa,* etc., have two unequal flagella at the anterior end. In *B. globosus* and *A. epithemiae* the short flagellum extends forward and the longer one backward in swimming, while in the other species the relative positions are reversed. The zoospores may become amoeboid, and engulf solid food particles (fig. 7, 8), and include a well-defined contractile vacuole. In the latter two characteristics they appear to differ sharply from the zoospores of the Plasmodiophoraceae, but as has been noted before the zoospores of *Polymyxa graminis* and the young plasmodia of *L. Junci* are said to engulf algae and particles of food.

In all these species, except *A. epithemiae* and *Gymnococcus Cladophorae,* the developing thallus becomes invested with a membrane and forms one zoocyst or zoosporangium (fig. 19). There is no cleavage into segments and development of a sporangiosorus, according to Scherffel. In *A. epithemiae,* on the other hand, the type of development is more like that of the Plasmodiophoraceae. The content of the zoospore enters the host, leaving the empty spore case on the outside, feeds upon the host protoplasm, and develops into an oval vacuolate thallus (fig. 15) which appears to be naked or devoid of a well-defined membrane. At maturity this plasmodium-like thallus cleaves into from 2 to 8 segments (fig. 16) which round up, form thin membranes, and become zoocysts. These vary greatly in size and in the number of zoospores they produce. Small zoocysts may form only 3 to 4 zoospores. No exit papillae for the emission of zoospores are developed, and at maturity the wall deliquesces and disappears (fig. 17) freeing the zoospores simultaneously. In *G. Cladophorae,* however, the wall is thicker, more permanent, and remains after the zoospores have emerged (fig. 18). No exit papillae are present here also, and the zoospores doubtless bore through the sporangium wall as in *A. algarum* (fig. 20). Scherffel did not observe resting spore formation, but his illustrations (fig. 22) suggest that they may be formed in the same manner as the zoocysts. They lie free in the host cell without an enveloping membrane. In *P. rotatoriorum* as many as 8 resting spores are formed in a sporocyst (fig. 21), but in this species they are held together by a membrane. Germination of the resting spores has not been observed.

It is to be particularly noted that the type of nutrition in these species is animal-like. The zoospores, amoebae, and developing thalli engulf chlorophyll granules, starch grains, oil globules, etc., apparently digest them in the food vacuoles, and extrude the waste material shortly before sporogenesis. No conclusive evidence of this type of nutrition has been found in the Plasmodiophoraceae, and this appears to be one of the chief differences between these two groups of organisms at present.

Comparison on the basis of sexuality, time and place of meiosis, alternation of generations, etc., cannot be made, because very little is known about these processes in the Proteomyxa. No good evidence of fusion of amoeboid or motile gametes has been observed in the biflagellate species. Likewise no evidence of schizogony, "promitosis," "akaryosis" or any other reported cytological characteristics of the Plasmodiophoraceae have been observed, but so far *Pseudosporopsis, Aphelidiopsis,* and other similar genera have not been intensively studied from fixed and stained material. It is accordingly premature to draw conclusions on these grounds.

The belief that the Plasmodiophoraceae are related to Protozoa, exclusive of the Proteomyxa which have already been discussed, stems primarily from the views of the protozoologists who have included this family among the primitive animals. Protophythologists in general have opposed this view on the grounds that the Plasmodiophoraceae are fungi. There are, nonetheless, certain specific structural, developmental and cytological similarities among the Rhizopoda and Sporozoa on which this belief is based. The suggested relationship with the Sporozoa relate to similarities in life cycles and asexual reproduction by schizogony, while in the Rhizopoda, exclusive of the Proteomyxa, it concerns the occurrence of "promitosis" and the extrusion of chromidia. The Sporozoa are spore-forming parasites of animals, some species of which may cause marked hypertrophy of the host cell and form galls or cysts. In certain species of the Myxosporidiae the spores give rise to amoebula which penetrate the host tissue, grow in size, and undergo schizogony, cutting off uninucleate schizonts. Each schizont develops into a multinucleate amoeboid plasmodium or trophozoite and divides into sporonts at maturity. The latter grow in size as their nuclei divide several times, become sporoblasts, and form a variable number of spores, which are usually liberated as the host tissue degenerates, and cause secondary infection. In these respects certain sporozoan species resemble the Plasmodiophoraceae, but further than this the similarity is not very striking. However, the occurrence of schizogony is particularly noteworthy. This is a common and widespread method of asexual propagation in the Sporozoa, and has also been reported to occur in most genera of the Plasmodiophoraceae. That its occurrence in both groups together with the production of numerous spores indicates phylogenetic relationship is, however, highly questionable and doubtful, as Maire and Tison ('09) have already pointed out.

The contention that the Plasmodiophoraceae show affinities to the strictly amoeboid Rhizopoda or

Amoebina is based primarily on the reported similarity between the vegetative nuclear divisions in the plasmodium and the promitotic divisions in the *limax* group of *Amoeba*. Cook ('28), as noted elsewhere, held this similarity to be of great phylogenetic significance and accordingly believed that the Plasmodiophorales have originated from the lobosoid amoebae. Horne ('30) severely criticized Cook's view, and after reviewing the variations of nuclear division exhibited by the fungi, algae, and protista, concluded that "the use of criteria relating to the type of nuclear division is of very doubtful value at the present time in discussing the actual relationship between group and group."

The reported similarity of promitosis in certain amoebae and the Plasmodiophorales has been fully presented in Chapter II and need not be discussed further at this point. Suffice it to repeat that Horne, Terby and Webb have refuted the reports of promitosis in the Plasmodiophorales and described the formation of well-defined chromosomes during the vegetative divisions. Furthermore, Miss Terby found that the nucleole does not persist and divide into two parts which are later incorporated in the daughter nuclei as the new nucleoli. Instead, the nucleole may fragment and portions of it become stranded in the cytoplasm between the nuclei, while the daughter nucleoli are formed anew in the telophases as in higher plants. There is accordingly no universal agreement that promitosis, in the strict sense of Nägler, occurs in the Plasmodiophorales. Nor is promitosis, in the modified sense of later workers restricted to the lobosoid amoebae. Intranuclear division with ill- or partly-defined chromosomes and large persistent elongating, constricting, and dividing nucleoli have been figured and described in species of the Rhizomastigina, Thecamoebina, Coccidia, Myxosporidia, Englenoidina and Siphonales. A similar persistence and behavior of the nucleole during division has been recorded by Nemec ('00), Mano ('04), Wager ('04), Lundegårdh ('12), and Tahara ('15) for *Alnus, Phaseolus, Solanum, Cucurbita,* and *Helianthus,* respectively, where the process has been referred to as pseudoprotomitosis. On this basis, according to Cook's line of argument, the Plasmodiophorales are related in varying degrees to a large number of animal and plant families. Persistence and division of the nucleole in the manner described above, therefore, does not appear to be of much significance, and as Doflein, Tischler ('22), Terby ('24), Belar, and others have pointed out, it may be found in various groups of organisms under certain conditions. In light of these data it seems highly doubtful that certain similarities in type of nuclear division are an index of phylogenetic descent and relationship.

It is obvious from this discussion of phylogeny and relationship that the Plasmodiophorales have some developmental phases and cytological characteristics in common with the Mycetozoa, Protozoa, and polysporangiate species of the Woroninaceae. Whether this order has originated directly from such groups or developed along parallel lines with them from a distant common ancestor, however, is still uncertain. Our knowledge of the critical stages in the life cycle of the Plasmodiophorales as well as in the groups with which this order shows affinity is too incomplete to warrant definite conclusions at present. Further intensive study of these stages as well as the discovery of new species will doubtless invalidate many of the present-day beliefs concerning the Plasmodiophoraceae. Likewise the similarities this family has in common with other groups, which now point to definite lines of origin and relationship, may in the future prove to be phylogenetically insignificant.

Nevertheless, the Plasmodiophorales at present appear to be similar to *Woronina polycystis* and the biflagellate heterocont species of the Proteomyxa in zoospore structure, and general type of development. This similarity, of course, does not necessarily mean a common origin and close relationship. It may equally well be nothing more than parallelism in development from separate ancestors. This relationship has, nonetheless, been emphasized rather strongly in the discussions above, primarily with the hope of encouraging intensive research along these lines.

Very little can be said at present about relationships within the order itself, because the life cycles of many species are not fully known. Furthermore the genera are not sharply defined. As is indicated in Chapter III, the relation and arrangement of the resting spores is rather generally regarded as an index of relationships and relative complexity. On this basis *Plasmodiophora* has been regarded as the most primitive genus, because its resting spores are not united in cystosori. *Tetramyxa,* and *Octomyxa,* with spores in tetrads and octads respectively, are accordingly next in line. *Sorosphaera* and *Sorodiscus* at present seem similar to these two genera in that uninucleate spore mother cells or sporonts are delimited in which the meiotic divisions later occur. Whether or not this is an index of relationship is, however, questionable. *Polymyxa* has the most extensive and complex zoosporangial stage of all known genera, but its cystosori resemble those of *Ligniera,* a genus which Cook ('33) regarded as primitive.

BIBLIOGRAPHY: PHYLOGENY

Abe, S. 1933. Bot. and Zool. 1: 1579.

———. 1934. Sci. Rept. Tokyo-Bunrica-Daigaku, sec. B. 1: 193.

Alexieff, A. 1913. Arch. Protistk. 29: 344.

Bessey, E. A. 1935. A text-book of mycology. Philadelphia.

Cadman, Elsie J. 1931. Trans. Roy. Soc. Edinburgh 57: 93.

Calkins, G. N. 1909. Protozoology. Philadelphia.

———. 1933. Biology of the protozoa. 2nd. ed. Philadelphia.

Cavers, F. 1915. New Phytol. 14: 301.

Chatton, E. 1910. Arch. Zool. Exp. et Gen. 5 ser. 5: 239, 267.

Cienkowski, L. 1865. Ark. Mikro. Anat. 1: 203.

Clayley, D. M. 1929. Trans. Brit. Mycol. Soc. 14: 227.

Cook, W. R. I. 1926. *Ibid.* 11: 210.
———. 1928. New Phytol. 27: 243.
———. 1933. Arch. Protistk. 80: 245.
———, and W. H. Nicholson. 1933. Ann. Bot. 47: 851.
Cornu, M. 1872. Ann. Sci. Nat. Bot. ser. 5, 15: 1.
Cotner, F. B. 1930. Sci. n. s. 71: 670.
Couch, J. N., J. Leitner, and A. Whiffen. 1939. Jour. Elisha Mitch. Sci. Soc. 55: 399.
Couch, J. N. 1939. *Ibid.* 55: 409.
De Bary, A. 1884. Vergleichende Morphologie der Pilze.
De Bruyne, C. 1890. Arch. de Biol. 10: 43.
Delage, Y., and E. Herouard. 1896. Traité de Zoologie Concrete 1: 76.
Doflein, F. 1901. Die Protozoen als Parasiten und Krankheitserreger. Jena.
———. 1909. Lehrbuch der Protozoenkunde. Jena. 3rd ed. 1911. 4th ed. 1916.
———, and E. Reichenow. 1928. Lehrbuch der Protozoenkunde. 5th ed. Jena.
Fischer, E. 1882. Jahrb. Wiss. Bot. 13: 286.
Galiano, E. F. 1921. Morfolgia y biologia de los protozoas. Madrid.
Gaumann, E. A. 1926. Vergleichende Morphologie der Pilze. Zürich.
———, and C. W. Dodge. 1928. Comparative morphology of fungi. New York.
Gilbert, H. C. 1935. Amer. Jour. Bot. 22: 52.
Gwynne-Vaughan, H. C. I., and B. Barnes. 1926. The structure and development of fungi. Cambridge. 2nd ed. 1937.
Harper, R. A. 1900. Bot. Gaz. 30: 217.
Hartog, M. 1906. Protozoa. Cambridge Nat. Hist. 1: 89. London. 2nd ed., 1909. 3rd ed., 1922. 4th ed., 1936.
Hertwig, R. 1919. Lehrbuch der Zoologie. 12th ed., Jena.
Howard, F. L. 1931. Am. Jour. Bot. 18: 116.
Jahn, E. 1904. Ber. Deut. Bot. Gesell. 22: 84. 1907, *Ibid.* 25: 53. 1908, *Ibid.* 26a: 342. 1911, *Ibid.* 29: 231.
———. 1914. Zeitschr. f. Bot. 6: 877.
———. 1928. Engler and Prantl, Nat. Pflanzf. 2: 304.
———. 1933. Ber. Deut. Bot. Gesell. 51: 377. 1936, *Ibid.* 54: 517.
Jones, P. M. 1928. Arch. Protistk. 62: 313.
Kambly, P. E. 1939. Am. Jour. Bot. 26: 88.
Karling, J. S. 1942a. *Ibid.* 29: 33.
———. 1942b. Mycologia 34: 205.
Klebs, G. 1893. Zeitschr. wiss. Zool. 55: 281.
Klein, J. 1882. Bot. Centralbl. 11: 254.
Kniep, H. 1928. Die Sexulität der niederen Pflanzen. Jena. Pp. 253, 258.
Kränzlin, H. 1907. Arch. Protistk. 9: 170.
Kudo, R. R. 1931. Handbook of Protozoology. Baltimore.
Kunkel, L. O. 1915. Jour. Agr. Res. 4: 265.
Lankester, R. 1885. Encyl. Brit. 9th ed. 19: 839.
———. 1909. A treatise on zoology 1: 11.
Lister, A. 1893. Jour. Linn. Soc. 29: 529.
Lotsy, J. P. 1907. Vorträge über Botanische Stammengeschichte 1: 403. Jena.
Lundegårdh, H. 1912. Cohn's Beitr. Biol. Pflanz. 11: 373.
Maire, R., and A. Tison. 1909. Ann. Mycol. 7: 248. 1911, *Ibid.* 9: 240.
Martins Mano, T. 1904. La Cellule 22: 57.
Minchen, E. A. 1912. An Introduction to the Protozoa. London. 2nd ed. 1922.
Nägler, K. 1909. Arch. Protistk. 15: 1.
Nawaschin, S. 1924. C. R. Acad. Sci. Russie 1924: 173.
Nemec, B. 1900. Funfstück's, Beitr. Wiss. Bot. 4: 37.
Olive, E. W. 1907a. Trans. Wisc. Acad. Sci. Arts, Letters 15: 753.
———. 1907b. Sci. n. s. 25: 266.
Pascher, A. 1918. Ber. Deut. Bot. Ges. 36: 377.
Pavillard, J. 1910. Progr. Rei. Bot. 3: 475.
———. 1912. Ann. Mycol. 10: 218.
Pinoy, E. 1908. C. R. Soc. Biol. 94: 630.
Prowazek, S. 1902. Österr. Bot. Zeitschr. 52: 213.
———. 1905. Arb. Kais. Gesundheit. 22: 396.
Rhumbler, L. 1923–1925. In Kükenthal ard Krumbach's Handbuch der Zoologie 1: 106.
———. 1939. Protozoology. Baltimore.
Rosen, E. 1893. Cohn's Beitr. Biol. Pflanz. 6: 237.
Scherffel, A. 1925. Arch. Protistk. 52: 1.
Schroeter, J. 1886. Cohn's Krypt. Fl. Schlesiens. 3: 133.
———. 1897. Engler and Prantl. Die Nat. Pflanzenf I, 1: 7.
Schünemann, E. 1930. Planta 9: 645.
Sinoto, Y., and A. Yuasa. 1934. Bot. Mag. Japan Bot. Soc. 48: 720.
Skupienski, F. X. 1917. C. R. Acad. Sci. Paris 165: 118. 1918, *Ibid.* 167: 31. 1926, *Ibid.* 182: 150. 1927, *Ibid.* 184: 1341.
———. 1928. Acta Soc. Bot. Poloniae 5: 255.
Smith, E. C. 1929. Am. Jour. Bot. 16: 645.
Stevens, F. L. 1913. The fungi which cause plant disease. New York.
———. 1925. Plant disease fungi. New York.
Stosch, H. A. 1935. Planta 23: 623.
———. 1937. Ber. Deut. Bot. Gesell. 55: 362.
Strasburger, E. 1884. Bot. Zeit. 42: 305.
Tahara, M. 1915. Bot. Mag. Tokyo 29: (1).
Tokunaga, Y. 1933. Trans. Sapporo Nat. Hist. Soc. 13: 20.
Tubeuf, K. F., and W. G. Smith. 1897. Diseases of Plants. London.
Vouk, V. 1911. Oesterr. Bot. Zeitsch. 61: 131.
Wager, H. 1904. Ann. Bot. 18: 29.
Wettstein, R. 1935. Handbuch der Systematische Botanik. 4th ed., p. 199. Leipzig and Vienna.
Wilson, M., and Elsie J. Cadman. 1928. Trans. Roy. Soc. Edinburgh 55: 555.
Winge, O. 1913. Ark. f. Bot. 12, No. 9: 26.
Woronin, M. 1878. Jahrb. Wiss. Bot. 11: 568.
Yuasa, A. 1935. Bot. Mag. Japan Bot. Soc. 49: 538.
Zopf, W. 1884. Encykl. der Naturwiss. 3: 129.
———. 1888. Abh. Naturf. Ges. Halle 17: 1.
———. 1894. Phys. Morph. Nied. Organismen 2: 3.

Chapter VI

Diseases Caused by Species of Plasmodiophoraceae

ALTHOUGH ALL known species of this family are parasitic only two members are economically important as pathogens of food crops. As noted elsewhere, *Plasmodiophora Brassicae* and *Spongospora subterranea* cause diseases of crucifers and potatoes respectively, which are commonly known as club root and powdery scab. The other species parasitize fungi, algae, cryptogams, and wild or seldom cultivated higher plants.

CLUB ROOT OF CRUCIFERS

Club root is a destructive root disease of wild and cultivated crucifers which is world-wide in distribution in temperate climates and known throughout the world by a large number of common names. In England, Scotland, and Wales it is known as finger and toe disease, anbury, hanbury, ambury, club root and clubbing; in Russia as herma or Kapoustnaja kila; Kropfkrankheit des Kohles, Kohlhernie, Klumpenfuss, Knotensucht, Fingerkrankheit, Kelch, Galle, Knolle, Huas, Kuss, etc., in Germany, Switzerland and Austria; Gros pied, maladie du Chou, and hernie du Chou in France; Tubereulosi dei cavoli and Mal de gozzo dei cavoli in Italy; Knoelvoet in Holland; Kwab, Kwabbe, Kwabbeziekte, Knol, Knolziekte, Klinker, Knoop, Knuist, Knobbel and Kwabbel in Belgium; Kaalbrok in Denmark; Klumprotsjuka in Sweden; Dik Voet in South Africa; club foot and club root in U. S. A.; and by various other names in other countries. According to Vanderyst ('04, p. 548) the name Vingerziekte used by Woronin and numerous subsequent writers for the disease in Belgium is unknown in that country.

From the economic standpoint club root is the most important disease of cultivated crucifers. In badly infested fields entire crops may be destroyed unless stringent control measures are employed. In Germany, England, Russia, the U. S. A., and other countries in Europe, Asia, and Africa 50 to 100 per cent destruction of turnips, swedes, cabbages, etc., has been reported (Brunchorst, '87; Rostrup, '93, '94; Halsted, '93–'99; Eyeleshymer, '94; Laubert, '05; Remy and Lüstner, '11; Georgeson, '16; Gleisberg, '20; Korff and Böning, '27, and others). Woronin ('78) reported that in 1869 the loss in the vicinity of St. Petersburg alone amounted to more than $20,000,000, while Herpers ('25) estimated that the annual loss in Germany runs into millions of marks. In New York State alone a loss of several thousand tons of cabbage were reported by Haskel and Martin in 1918. Edson, Miller and Wood ('35, '36, '37) have subsequently reported losses of 5 to 100 per cent in cruciferous crops throughout the U. S. A. The most significant fact about club root is that it spreads rapidly, and once it has become established in the soil, it makes the fields almost useless for crucifer cultivation for a number of years.

The origin of club root is unknown, but its symptoms had been well described more than a century before Woronin showed it to be due to a plasmodiophoraceous organism. According to Böhner ('22), the disease is as ancient as its hosts. The occurrence of spongy, fungus-like roots (radices fungosae) of crucifers noted by Albert the Great as early as the 13th century is supposed to relate to club root, and his control practice of avoiding fresh stable manure and the disposal of chaff appears to have been acquired from the Roman Pallatius, according to Böhner. The disease was well known in Spain in the 15th century where cabbages were described as being syphilitic (see Woronin, '78, p. 552), and the swellings were thought to be due to the organism causing syphilis (Ruiz Diaz de Isla). The first report of its occurrence in England was made by Ellis in 1736, who believed the disease was contagious and due probably to an excess of barnyard manure. Adam discussed its widespread occurrence in England in 1789, and it was subsequently reported in Scotland from 1829 to 1831 by Farquharson, Abbay, and Birne who thought it to be due to unsatisfactory soil conditions or unbalanced fertilizer practices. Abbay saw the disease as early as 1801, and Anderson stated that it first became troublesome in Scotland about 1813. Renard reported that it was first observed on cauliflower in 1820 in France. By 1853 it was fairly abundant around Hamburg, Wurzburg, in the Rhine valley and other parts of Germany (N. N. '53), and from 1855 on it appeared in various parts of Norway (Jorstad, '30). Other workers, including Curtis ('43), Kühn ('58), Henderson ('67), Sorauer ('74), Slingerhand ('94), and others (see Woronin '78, pp. 552–554) believed it to be due wholly or in part to various insects and other animals. Buckman ('54), however, claimed that club root was due to reversion to the original wild forms. By 1872 the disease had become so widespread and destructive around St. Petersburg that the Royal Russian Gardening Society in St. Petersburg offered a prize for the solution of the cause and control of hernia. Woronin began to study the disease independently of this offer in 1873, and two years later he announced that it is caused by a plasmodiophoraceous organism to which he subsequently ('78) gave the name *Plasmodiophora Brassicae*.

Symptoms

Club root disease is usually characterized by marked enlargement of the infected roots (Pl. 2, fig. 1), and in exceptional cases the galls on cabbage may reach the size of a man's fist and appear greasy-gray and pale-yellow in color. In most cases the clubs are

regularly spindle-shaped, but when several infections occur together the swellings may fuse and produce irregular growths or compound spindles (fig. 3). According to Küster ('11) and M. T. Cook ('23) these galls are kataplasmic, since the affected tissues usually remain parenchymatous and do not undergo differentiation. Other root symptoms have also been reported. According to Appel and Werth ('10), no hypertrophy occurs in radishes, and the disease is here characterized only by darkened and decayed areas. Honig ('31) found similar symptoms on *Lunaria biennis*. Ravn ('22) and Pape ('25) likewise reported the occurrence of deep wounds or lesions on turnip roots which were filled with spores. According to Pape, such symptoms appear when the galls or nodular excrescences on the roots decay.

In a study of 104 species from 28 genera, Cunningham ('14) found definite types of hypertrophy and symptoms more or less characteristic for certain crucifers and classified them accordingly:

1. Complete clubbing of main and lateral roots. *Brassica oleraceae*.
2. Clubs on main root, laterals free. *Sisymbrium altissimum*.
3. Clubs on lateral roots, main root free. *Sisymbrium officinale* and *Erysimum cheiranthoides*.
4. Clubs on main and lateral roots with club-free rootlets above the diseased portion. *Lepidium sativum*.
5. Clubs as tumors of the roots. *Raphanus sativus*.
6. Dark, decomposing spots on the roots. *Raphanus sativus*.

In the last category true hypertrophy does not occur. The disease is here characterized by cracks, fissures, and darkened areas in the host tissue which turn black, decay, and serve as sites of secondary infections by other fungi. As has been noted above, Appel and Werth claimed that these are the characteristic symptoms of the disease on radishes, but Cunningham found them only on the Everlasting radish, in addition to spindle-shaped swellings of the rootlets.

Club root disease may also stimulate branching of the roots and shoot and lead to the production of buds where they do not normally occur, as has been described by Caspary, Woronin, Favorski, and Kunkel. The secondary roots may attain a length of several inches or become stunted as short knobs. On the other hand, the production of secondary rootlets may be greatly inhibited, according to Laubert ('05) and Schlumberger ('14). The diseased buds on infected roots and shoots are often unable to respond normally to gravity, and they may grow downward and horizontally as well as upward. In the latter instances the infected buds may push up above the surface of the ground and give rise to thick, distorted, fleshy, and abnormally succulent leaves and petioles, so that the disease may occasionally manifest itself above ground in the shoot, petioles, and leaves. In addition to these above-ground symptoms, club root causes yellowing of the leaves, wilting on hot days, and in the case of cabbage, atrophy, or complete lack of head development. Seedlings which are infected early usually die within a few weeks. The wilting of large diseased plants is partly due to hypoplasia of the xylem region and to splitting up of the woody cylinder by infection and expansion of the medullary rays.

All galls or swellings on roots of crucifers, however, are not due to *P. Brassicae*. Nematodes, insects, and other factors may cause malformations which are superficially very similar to club root, and unless microscopic examination of the tissues is made, these galls may be easily mistaken for those of the finger-and-toe disease.

Anatomically, the causal organism of club root affects the cortical parenchyma most conspicuously, but it also produces marked changes in the cambium, xylem, and medullary rays. When roots of considerable size are infected the amoebae and small plasmodia migrate through the cortical parenchyma into the cambium. Here they follow the path of least resistance, according to Kunkel and Larsen, and spread up, down, and around the central cylinder through the delicate thin-walled cambium cells and form thus a cylinder of infected tissue. From the cambium they may travel laterally into the cortex, medullary rays, and xylem. Their migration up and down in the cambium ceases after a while, and the distance of the infection in these directions determines the ultimate length of the spindle-shaped club. Each club, in Kunkel's opinion, is a morphological unit which has resulted primarily from the abnormal growth of the cambium. In comparatively old infected roots the medullary ray cells divide a number of times and enlarge and thus form large bands of pathological tissue which split and force the xylem tissues apart, until the latter becomes distorted and shifted out of their natural position. Separated from each other in this manner, the vascular bundles grow out fan-wise instead of remaining wedge-shaped and are no longer able to function normally. Plasmodia and amoebae have frequently been found in the tracheids, but they do not seem to have any appreciable effect on the normal functions of such differentiated cells. In young roots medullary ray infection is less common, and most of the abnormal growth occurs in the region of the cambium and the cortex. The xylem, nonetheless, may fail to differentiate properly and is often supplanted by a mass of partially differentiated cells.

As is shown by figure 4, one of the most striking appearances in sections of diseased roots and shoots is the presence of more or less isolated groups of hypertrophied infected cells which Nawaschin named "Krankheitsherde." He believed that these groups arise by repeated anti- and periclinal division of one or more originally infected cells, whereby the plasmodia are passively distributed in a radial direction around the region of infection. Chupp also reported that a single amoeba might give rise to as many as six such groups by multiplication and migra-

tion from cell to cell. His account was subsequently confirmed by Kunkel who believed that a single infection may lead to the formation of thousands of separate and distinct "Krankheitsherde." Kunkel assumed that as a plasmodium migrates from cell to cell it may divide, whereby portions are left behind and become established here and there in the tissue and give rise to groups of infected cells.

Cellular Interrelations Between Host and Pathogen

Plasmodiophora Brassicae has a pronounced effect on infected and healthy cells. Infection may be temporary or permanent, and if the plasmodium migrates out of a cell before stimulating much change, the latter may recover and continue to function normally. Permanently infected cells, however, may expand to more than 10 to 20 times their normal size. In the early stages of infection the presence of the parasite does not inhibit nuclear (Pl. 2, fig. 5) and cell division (fig. 6), so that some cells may function normally in this respect for a short time. Other cells may begin to enlarge directly after infection without dividing. Occasionally, cell division may be affected to the extent that the cell wall is only partly developed across the mother cell (fig. 6). Eventually the power to divide is lost completely, and the infected cell gradually expands to its large size. Prowazek found that karyokinesis may continue after cell division has ceased, resulting in binucleate cells. Lutman also found abnormal types of mitosis which appeared to be a modified form of amitosis.

The first visible effect of the parasite on the host nucleus is an enlargement of the nucleus as a whole followed by an increase in the number of nucleoli, according to Lutman (fig. 8–11). By the time the parasite is mature, the host nucleus has lost its regular outline, and the nucleoli lie (fig. 11) in clear spaces surrounded by a distinct membrane, an appearance which led Prowazek to assume that smaller nuclei may be formed in a mother nucleus. In the final stages of degeneration the chromatic material collects into irregular strands (fig. 12) and assumes a peripheral position in the distorted and hypertrophied nuclei.

The relation between the protoplasts of host and pathogen appears to be very intimate, and little or no visible antagonism is exhibited. The amoebae and young plasmodia of the parasite lie embedded in the host protoplasm (fig. 5, 6, 26, 28), and in the living condition the two are indistinguishable, according to Woronin, Nawaschin, Lutman, and others. This close association together with the fact that the infected host cells may continue to divide and function normally for some time led Nawaschin, Gaylord, and Vanderyst to believe that there is a symbiotic relationship between the host and pathogen during the latter's early developmental stages. The host cytoplasm has been described as becoming more vacuolate as the plasmodium enlarges, but part of the early change is probably due to the great increase in volume of the host cell whereby the cytoplasm is thinned out. Later, however, as the plasmodia mature and approach sporogenesis the protoplasm is almost completely gone. Infected cells develop an unusually large amount of transitory starch, according to Walker, Halsted, and Nawaschin, which may be grouped around the nucleus as Lutman has shown. These grains may later be found in the plasmodium (fig. 74) and are apparently wholly or partly digested before sporogenesis. Reed ('11) noted an appreciable increase in calcium, magnesium, potassium, phosphoric acid, sulphuric acid, etc., in diseased cabbage roots. The increase was greatest in the case of potassium, which he attributed to an accumulation of protoplasm and starch in diseased tissues. Nicoloff and Stefanova ('22), however, found that roots of diseased cabbage plants were high in protein and lower in phosphorus and potassium than those of healthy plants.

Noninfected cells are also stimulated to divide by the presence of the parasite and may often enlarge considerably. This is particularly true of medullary ray cells, which may expand until they have lost all characteristics as such. The nuclei of these cells enlarge also and keep pace to some extent with the increase of cell size. According to Kunkel, the stimulus travels in advance of the infection, so that increased cell division may be noted before the parasite reaches a particular, undifferentiated tissue, which suggests that a growth-stimulating substance is released by the causal organism and travels ahead of the plasmodium. Nawaschin, on the other hand, believed that the division of noninfected cells around the "Krankheitsherde" is due to the stimulus of mechanical outward pressure exerted by the enlarging parasitized cells.

Kunkel suggested that the limitation of the parasite in groups of cells might be due to a protective substance or antitoxin produced by the infected cell which diffuses out into the adjoining healthy cells and renders them immune to attack. Levine and Levine ('22) believed that the surrounding cells are not only immune but present a reactive protective barrier against the spread of the parasite. The question of whether or not infected plants can recover from club root and become immune has often been debated. Woronin ('78), Eycleshymer ('91), Laubert ('05a) and Müller-Thurgau and Osterwalder ('23) maintained that recovery is impossible, but Massee ('96), Mathieu-Sanson ('97), Appel and Schlumberger ('13), Schlumberger ('14), and Wahling ('22b) reported varying degrees of recovery when infected plants were treated with a 2 per cent potash solution, milk of lime, planted in ore mud, and sterile soil, and watered with sulfur and solibar solutions. Müller and Osterwalder transplanted infected plants to heavily limed soil, but found no inhibitory effects or recovery. Hönig ('31) believed that if infected plants are transplanted to sterile and disinfected soil the progress of the disease may be halted, but such plants can recover only if they are sufficiently healthy to begin to grow anew.

Entrance and Spread of *P. Brassicae* in the Host

Actual penetration of *P. Brassicae* into the host was not observed by the early workers, but most of them assumed that it occurs only when the plants are young and susceptible. Hönig and Rochlin, however, subsequently demonstrated its entrance through the walls of root hairs and epidermal cells, although Woronin, Chupp, Cook, Schwartz, and others had previously held that the amoebae gain entrance through the root hairs (fig. 28, 29) and migrate into the deeper lying tissues. W. G. Smith ('84), on the other hand, maintained that the parasite enters as a plasmodium. Favorski reported that infection may take place through ordinary epidermal cells and stated that Woronin's figures of amoebae in root hairs relate to *Olpidium Brassicae*. Kunkel found that old plants are as susceptible as young ones and that infection of old roots is very common. He further refuted the claim that root hairs are of any importance as avennes of infection and concurred with Favorski's belief that Woronin had figured thalli of *O. Brassicae* and *O. borzii* in the root hairs instead of *P. Brassicae*. Cook and Schwartz, Hönig, Rochlin, and others, however, have subsequently demonstrated quite definitely that *P. Brassicae* occurs in root hairs and thus confirmed the observations of Woronin and Chupp. Kunkel, nonetheless, showed that old plants are susceptible and may become infected as long as they live. Infection through mechanical wounds and ruptures caused by adventitious roots and by the removal of lower leaf petioles at the time of transplanting is fairly common, according to Larson ('34). The enlargements, however, which are formed at the region of injury on the stem are definite spheroid galls in contrast to the spindle-shaped clubs on the roots.

As to the spread of the parasite in the host tissues and the channels involved, it is now generally agreed that it occurs in two ways: by migration of amoebae and young plasmodia from cell to cell, and by passive distribution of the parasite through repeated divisions of infected cells. Woronin contended that amoebae and plasmodia migrate only through pits and sieve plates, while Atkinson believed that amoebae are able to spin out into such fine threads that they can enter the roots along with nutrients in solution. Eyclesheimer found plasmodia in xylem vessels and thought therefore that they may travel in the fibrovascular bundles. Nawaschin believed that migration of amoebae from cell to cell is impossible after secondary thickening begins in the roots, and hence distribution by division of infected cells is the principal method of dissemination in old roots. Subsequently, Lutman figured and described the passage of small plasmodia from cell to cell, and since that time Chupp, Kunkel, Honig, Rochlin, and others (fig. 31–33) have demonstrated its occurrence. Cook and Schwartz, more than a decade later, however, still expressed doubt as to its occurrence. Fedorintschik ('35) believed that in the early stages of the disease, migration of amoebae is the principal method of distribution in the host tissues, but after the plasmodia have formed and begun to mature, further spread is by division of infected cells. While it is now generally believed that division of the host cell greatly increases the number of infected cells, it nevertheless appears to play a minor role in distributing the parasite throughout the roots and shoots.[1]

Dissemination of *P. Brassicae* in Nature

The club root organism is readily disseminated in nature in various ways and by numerous agents. It was formerly believed (Atkinson, '89; Carruthers, '93; Müller and Osterwalder, '19) that the motility of the zoospores in moist soil spread the disease, but Chupp ('17) has presented evidence to show that zoospores and amoebae rarely travel more than five inches. It has also been claimed (Carruthers, Ravn, '08, and others) that wind is an important agent of dissemination, but this factor apparently operates only in the case of light, dry, loose soils and where strong winds prevail. It has been demonstrated in heavier and more compact soils that unless the pathogen is transferred by some other agent, wind does not usually spread it from one field to another. Rains and water are doubtless more important, particularly on rolling land where the water following a heavy rain runs off quickly and carries the spores to lower-lying fields. According to Naumov ('25), however, dispersal in a radial direction by such means is not very extensive. Müller-Thurgau and Osterwalder ('23) reported that in the course of a year club root does not spread laterally more than 1½ to 2 meters in the ground. Earthworms have also been found to be active in the dissemination of club root in small gardens (Gleisberg, '22; Bremer, '24; Fedorintschik, '35). The spores may be carried in the mucilage on the skin or in the intestinal tract, and virulent forms of *P. Brassicae* have been found in the excreta of worms. Ground moles, root nematodes and insects feeding on diseased roots doubtless spread the disease to some extent (Favorsky, '10; Esmarch, '24; Beyer, '25; Chupp, '25; Erickson, '26), but how important they are as active disseminators is not known.

[1] In a paper presented before the meeting of the American Phytopathological Society at Dallas, Texas, December 1941, Walker reported system infection of cabbage and distortion of buds, stem, and leaves as follows: "Under greenhouse conditions when cabbage seedlings are grown in soil infested with *P. Brassicae* the pathogen, after infecting the root, may migrate through the cambium into the stem. There is relatively little cambial proliferation in the internodal regions above the third or fourth leaf. Dormant buds at the leaf scars, however, are stimulated to grow and become invaded by the pathogen. They become malformed due to hyperplasia. The organism may reach the growing point in young plants and cause extreme distortion of stem and leaves. When plants are inoculated at above ground leaf nodes, the pathogen may migrate down the stem, leaving no evidence of proliferation in its path until the hypocotyl is reached, where a typical club is formed. There is evidence that the reaction of the host is influenced by the nutrient supplied to it." (Phytopath. 32: 18)

Dispersal by the dung of livestock fed with diseased roots is very common. The spores remain alive during passage through the digestive tract, and if animals which have been fed on diseased crucifers are let out to pasture, the spores are disseminated in the droppings. Gibbs ('31) found that the spores may remain viable in fresh cattle droppings for at least fifteen weeks. They also remain alive for long periods of time in dung piles around stables, and it has long been known that the application of such manure to virgin soil introduces the parasite. Transport of infected soil on farm implements, laborer's, horse's and livestock feet, etc., is also effective in spreading the disease.

Numerous wild cruciferous plants are susceptible to club root, as has been shown by Halsted ('96–'99), Ravn ('08), Cunningham ('12), Ssacharoff ('16), Naumova ('26), Gibbs ('32), Rochlin ('33), Jamalianen ('36) and others, and these hosts often harbor and perpetuate the disease in the absence of cultivated crucifers. Such wild infected hosts have been found in grass pastures, wayside ditches, river beds, gardens, and cultivated fields, and their presence on infected soil reduces the effectiveness of crop rotation in club root control.

Environmental Factors

The degree of infection, development and severity of club root depends to a large extent on environmental factors, but the manner and extent to which each factor operates are not clearly understood. The disease is commonly believed to be favored by wet, poorly-drained, acid soils and temperatures slightly higher than those optimum for host root development, but reports to the contrary have often been made. Motte ('33), for instance, reported that club root is most prevalent in light soils and during the dry season in Denmark.

As to spore germination, many workers have found it occurs mostly abundantly in acid media. Bremer ('23, '24, '26), however, reported that H-ion concentration is not the sole determining factor. He found that strong alkalinity inhibits germination of the spores without killing them and that germination occurs over a pH range of 5.4 to 7.5 but not at pH 8.0. Honig ('31), on the other hand, reported that spores germinate as well in alkaline as in acid solutions; all of which indicates that other little-known soil factors operating in combination are equally as important as H-ion concentration.

Most workers have, nonetheless, found a fairly close correlation between incidence of infection and pH range (Massee, '96; Christensen, Harder and Ravn, '11; Ravn, '12–'13; Hiltner and Korff, '16; Neger, '17; Atkins, '22; Bremer, '24–'28; Lindfors, '24, '25; Naumov, '25; Ludwigs, '25; Riehm, '25; Tessenow, '26; Gleisberg, '26; Chupp, '28; Brönnle, '28; Martin, '28; Blunck, '29; Schaffnit and Meyer, '30; Beaumont and Staniland, '33; Wilson, '34, etc.). Lindfors ('24) observed a marked decline in percentage of infection with an increase in soil alkalinity. In a pH range of 7.1 to 7.5, 85 per cent of the plants were diseased while at pH 7.8 to 8.0, all plants remained healthy. Naumov ('25) found that infection occurs most readily at pH 6.0 to 6.5 with the optimum near neutrality, although infection of seedlings took place within a range of 5.7 to 8.4. In a more intensive study of the problem in 1927 he further found that percentage of infection is not consistently correlated with the pH range, as is shown below:

300 gms.	BaO_2 per 100 cc. soil					pH. 6.8	Infection	0.00%
100 "	"	"	"	"	"	pH. 7.3	"	0.00%
60 "	"	"	"	"	"	pH. 7.2	"	0.00%
30 "	"	"	"	"	"	pH. 6.9	"	14.30%
15 "	"	"	"	"	"	pH. 7.4	"	20.90%

Blunck ('29) likewise found infection occurring at pH. 8.8. Further inconsistencies in the literature on the effects of raising the pH value is shown by the reports of Martin ('28), Schaffnit and Meyer ('30), Wilson ('34), and others that club root can be effectively controlled or serious loss prevented by adjusting the pH of the soil to 7.4 and above. Chupp ('28) also reported that infection does not ordinarily occur in soils with pH ranges above 7.2 to 7.4.

In 1930 Wellman made a survey of 116 club root infested fields in Wisconsin and found a pH range of 5.0 to 7.8. In Lithuania, Vilkaitis ('33) found the range to extend from 4.0 to 7.6. By the addition of certain chemicals to the soil Wellman modified the pH value experimentally and found that raising the H-ion concentration did not consistently inhibit the disease as is shown below:

pH value	$Ca(OH)_2$	$CaCO_3$	K_2CO_3
7.1	Diseased	Diseased	Diseased
7.2	"	———	"
7.3	Healthy	———	"
7.4	———	Diseased	———
7.5	Healthy	"	———
7.6	"	"	Diseased
7.7	"	———	———
7.8	———	Diseased	———
7.9	———	Healthy	———
8.0	Healthy	———	———
8.1	"	———	Diseased

It is to be particularly noted that addition of sufficient amounts of K_2CO_3 to bring the pH up to 8.1 did not inhibit the disease. In thoroughly infested fields treated with lime Wellman further found that 3 per cent of cabbages were destroyed at pH 8.1, and 54 per cent and 49 per cent destroyed at pH 6.7 and 7.5 respectively. Wellman accordingly concluded from his experiments that club root occurs in such a wide pH range that to consider H-ion concentration alone as an important factor in the occurrence of the disease is highly questionable.

Since that time other workers have also shown that club root may occur in a high pH environment. Beaumont and Staniland ('33) reported that while infection is most common in acid soils, swedes and broc-

coli may become badly clubbed in soils adequately provided with lime. In 1934, however, they reported that turnips and swedes are unaffected by club root in soil the pH value of which was raised to 6.76 and 7.02 by liming and concluded that pH 6.6 is the probable limit for the disease. Larsen and Walker ('34) also observed that the addition of calcium hydroxide and calcium or magnesium carbonate in doses sufficient to raise the pH to 7.1 and above did not generally inhibit development of club root in silty clay loam soils. In the greenhouse, however, infection was reduced by the addition of these substances sufficiently to bring the pH up to 7.0. At the pH 7.2 or above infection was completely inhibited. Whitehead ('36) likewise noted that the disease is generally less prevalent in alkaline soils but he also found a high percentage of infection in cabbage, cauliflower and Brussel sprouts in soils with pH ranges of 7.45 to 7.81.

It is obvious from the data presented above that H-ion concentration in the soil is not the sole determining factor in infection, development, and severity of club root. As Naumov and others have pointed out the intensity of infection is intimately associated with many external and internal factors, such as degree of soil infestation, moisture content, anatomical structure of the hosts, specific and varietal susceptibility, etc.

Temperature does not appear to be as important as other factors in spore germination, infection, and development of club root, because these processes may occur under a fairly wide range of temperature. It was commonly believed that outbreaks of disease were most severe in cold countries and during the cool seasons in warm regions, but this belief was not based on exact experimental data. As to spore germination Chupp ('17) found that it does not occur at room temperature (16° to 21° C) and that the optimum lies between 27° C and 30° C. He, nevertheless, obtained host infection at room temperatures, which indicates that temperature was not the only important factor in his experiments. Wellman, on the other hand, found that spores will germinate within a range of 6° C to 27° C, with an optimum range of 18° to 25° C. Honig ('31) likewise found that spores may germinate readily below 21°. As to the direct effect of soil temperature alone on infection and club root development very little experimental evidence is available, but temperature doubtless operates indirectly in conjunction with other soil factors. In carefully controlled tests Montieth ('24) showed that club root develops at 9° to 30° C. One case of clubbing was found at 35°, but it occurred on the main stem near the surface of the soil where contact with air probably lowered the temperature. Clubbing was most severe at 25°. Montieth concluded that the temperature range over which the disease occurs is more or less the same as that required by the host and that temperature itself is not a limiting factor in club root development. In similar controlled tests Wellman found that no clubbing occurs below 12° and above 27°. The optimum temperature for greatest infection and disease production ranged from 18° to 24°, with the peak for severity slightly above 24°. In soil temperatures of 12°, 15° and 27° a fair percentage of plants became infected, but clubbing was distinctly inhibited. The optimum temperatures for spore germination, infection and development of the disease determined by Wellman are 5° higher than those which Tisdale (Jour. Agric. Res. 25: 55) found to be optimum (20°) for cabbage root development.

Soil moisture is more significant than temperature in relation to infection and severity of club root. The early student of this disease as well as later investigators, including Halsted, Ravn, Cunningham, Chupp, Whitehead, Reed and others, noted that the disease is most prevalent in low lying, poorly drained soils and severest after periods of wet weather, and concluded that soil moisture is perhaps the most important determining factor. These conclusions, however, were based more on general observations than on direct experimental evidence. Montieth demonstrated experimentally the dependence of club root on high soil moisture and showed that cabbage could be grown free of the disease in heavily infested soil by keeping the moisture down to 45 per cent of the total water holding capacity. At 60 per cent club root was uniformly present. He believed that the failure of club root to develop in infested soils with low moisture content is probably due to insufficient water for spore germination. Montieth's results have been by and large confirmed by Wellman and Naumov. Wellman, however, demonstrated that continued high soil moisture is not necessary for infection and development of the disease. Plants which had been exposed only 18 hours to infested soil with 80 per cent moisture content became badly diseased when transplanted to relatively dry infested soil. He believed that even in a dry season a heavy rain or a few moderate rains at short intervals might raise the moisture content sufficiently to insure infection. Wellman's results may be the explanation of Motte's report that club root is prevalent during the dry seasons in Denmark. Naumov ('33) likewise found that cabbage seedlings became infected within a range of soil moisture from 45 to 100 per cent of the total water-holding capacity, with the optimum at 80 per cent. At 30 per cent no development of the disease occurred.

The physical character of the soil has also been regarded as a significant factor in club root infection, development, and severity. Sandy, humus-rich, clayey soils favor the disease, according to McAlpine ('03), Bos ('04), Janson ('20), and Naumov ('28). In Belgium, Vanderyst ('04) reported that club root occurs abundantly on sandy soils, is generally present in soils rich in shale, sparse on limey and clayey soils, and unknown on soils rich in lime. Humphrey ('92) and Read ('11) found the disease to be most severe on heavy soils and those rich in humus. Ravn ('08) reported that turnips were more susceptible than cabbage on sandy soil, while on clayey soil the degree of susceptibility was reversed. Hayunga ('19), Janson ('20) and Bremer ('23) found club

root to be less severe on marsh and heath soils. Soils poor in lime generally favor development of the disease, according to Eyckeshymer ('91), Massee ('96), Laubert ('35a) Burkhardt ('15), Triesehmann ('17), Bos ('18), Bremer ('23–'28) Kindshoven ('24), Hall ('10), and Atkins ('22). Naumov ('27) found that soils with a lime (in terms of oxide) content of 0.4 per cent or more are generally free of the disease, but Honig ('31) reported that in the vicinity of Munich soils with a 58 per cent lime content were heavily infested with club root. Herpers ('29) and Honig observed that the disease is very abundant in soils which heat up readily.

The physical character of the soil also influences the infective ability of the fungus spores according to Fedotova ('28). In ordinary grey garden soil with 40,000 spores per cc. of soil, 66 per cent of the plants became infected, while in black, greenhouse dirt with 100,000,000 spores per cc. only 12.5 per cent of the plants were clubbed. Naumov ('28) also found that in clayey soils 20,000,000 spores per cc. of soil were necessary for optimum infection, while in humus-rich soils 100,000,000 were essential. He, furthermore, reported that in the vicinity of Leningrad the spores do not remain viable in the soil in the absence of hosts for more than three years unless fairly high temperatures and humidity are maintained.

The observations of most of the workers mentioned above were not correlated with exact data on the water-holding capacity and acidity of the respective types of soil, and it is quite probable that the increased infection and severity of club root reported on clayey, heavy soils and those rich in humus are due not so much to the physical nature of the soils as to their high acidity and water-holding capacity.

Hosts and Degree of Infection

Club root was first observed on cultivated crucifers, but later it became evident that wild species of the mustard family also are susceptible to this disease. Woronin reported hypertrophied roots of *Iberis* in 1878, and some years later Magnus ('93) and Henning ('96) found other genera and species attacked by *P. Brassicae*. Halsted ('92–'99) appears to be the first to have undertaken a more extensive study of the host range, and since that time this phase of the disease has been intensively investigated in various parts of the world. Club root is confined to species of the mustard family, and although reports of its occurrence on plants outside of this family are to be found in the literature, they have subsequently been proven false. The number of hosts is large, and in the following table are listed the cruciferous species which have been examined for the presence of club root. Included also is the degree of infection found by investigators who have studied the host range of *P. Brassicae*. Previous authors have usually arranged the genera and species according to sub-families, but for the sake of convenience they are listed in alphabetical order below.

Index to Authors Cited and Degree of Infection of Hosts

Appel = Appel and Werth ('10)
Cun. = Cunningham ('14)
Clint. = Clinton ('06)
Da. = Davis ('25)
Erick. = Ericksson ('96, '26)
Gi. = Gibbs ('32)
Gl. = Gleisberg ('23)
F. S. = F. S. ('20)
Hal. = Halsted ('92–'99)
Ham. = Hammarlund ('15)
Henn. = Henning ('96)
Hon. = Honig ('31)
Höst. = Höstermann ('21)
Jam. = Jamalainen ('36)
Katt. = Katterfeld ('23)
Mag. = Magnus ('93)
Mass. = Massee ('96)
Mü. = Müller-Thurgau and Osterwalder ('23)
N. N. = Anonymous ('53)
Naum. = Naumann ('13)
N. = Naumov ('14–'28)
Rain. = Rainio ('30a)
Ravn. = Ravn ('08)
Roc. = Rochlin ('33)
Ros. = Rostrup ('93)
Schl. = Schleyer ('07)
Sit. = Sitensky ('98)
Ssach. = Ssacharoff ('16)
Svec. = Svec. ('23)
Weiss. = Weiss ('18)
Wor. = Woronin ('78)

0 = no infection
+ = weak infection
++ = medium infection
+++ = severe infection

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Aethionema arabicum	N. ('24) 0
A. buxbaumii	Katt. 0
A. cappadicum	N. ('25) 0
A. rotundifolium	N. ('15) 15%
Alliaria officinalis	Mü. 0; Ravn; Jam. 0
Alyssum alpestre var. *odoratum*	N. ('13) 10%; Roc. 47%
A. alpestre	Hal.; Ravn.
A. alyssoides	Gl. 20%; N. ('12)+; N. ('13)+, Cun. 51.4%;
A. argenteum	Jam. 0
A. bornmülleri	Gl. 0; Jam. 0
A. benthami compactum	Jam. 10.5%–11.6%
A. calycinum	Gl. 71.43%; N. ('12)+; N. ('13)+
A. campestre	Jam. 3.8%
A. condensatum	Cun. 33.4%
A. corymbosum	Jam. 31%
A. desertorum	N. ('25) 0; Roc. 57%
A. edentulum	N. ('15) 50%
A. fischerianum	Jam. 0
A. gemonense	N. ('15) 5%
A. idaeum	Jam. 14.5%
A. maritimum = *Lobularia maritima*	Hal.+; Ravn. Cun. 41.2–50%; N. ('15) 5%; Jam. 0

A. minimum Gl. 4.76%; N. ('15) 0
A. moellendorfianum Cun. 100%
A. montanum Gl. 0; N. ('12)+; Cun. 22.2%; Jam. 18.8%
A. podolicum Jam. 3.9%
A. rostratum Cun. 86.7%
A. saxatile Hal.; N. ('14) 3%; Cun. 32%
A. serpyllifolium Cun. 8.3%
A. sinuatum N. ('15) 0; Gl. 100%
A. Stribrnyi N. ('15) 0
A. umbellatum Gl. 0
A. Wierzbickii Cun. 0
Arabis albida N. ('14) 0; Jam. 0
A. albida var. *grandiflora* N. ('14) 0; Jam. 0
A. albida var. *nana* N. ('14) 0
A. albida var. *umbrosa* N. ('13) 60%
A. Allionii Jam. 0
A. alpestris N. ('13) 50%
A. alpina Cun. 52.4%; N. ('12)+; Roc. 27%–18%
A. arenosa N. ('12)+
A. bellidifolia Jam. 1.9%
A. brachycarpa Hal. +
A. Canadensis Hal.
A. coerulea Jam. 0
A. glabra Hal.
A. halleri Cun. 0
A. hirsuta N. ('12)+; N. ('13) 80%; Jam. 1.5%
A. holboelli Cun. 50%
A. laevigata Hal. +; Ravn.
A. muralis var. *collina rosea* Jam. 0
A. pendula N. ('14) 0
A. petraea N. ('12)+
A. procurrens N. ('15) 0; Jam. 0
A. pumila N. ('14) 0; N. ('25) 80%
A. Stelleri N. ('25) 44%
A. suecica N. ('15)+; 72.2%
A. Turrita Jam. 7.4%
Aubretia Bougainvillei Cun. 0
A. deltoidea Roc. 0
A. eyrei Cun. 0
A. graeca Cun. 0
A. hendersoni Cun. 0
A. Leichtlini Cun. 37.5%
A. olympica Roc. 0
A. pinardi N. ('15) 0; F. S. ('20)+
A. purpurea Cun. 67.7%
Barbarea arcuata Gl. 0
B. bracteosa Roc. 0
B. intermedia Gl. 0
B. lyrata Roc. 0
B. plantaginea Roc. 65%
B. praecox Gl. 0; N. ('12)+; N. ('15) 0
B. rupicola Roc. 99%
B. stricta Cun. 4.3–7%; Gl. 0; Gi. 0
B. verna Gi. 0
B. vulgaris Cun. 0–70.1%; Ham., Gl. 0; N. ('14) 0; N. ('25) 1.6%; Gi. 0; Rain. 66.7%; Jam. 3.5%
B. vulgaris fol. *variegatis* Roc. 0; Jam. 0
Berteroa incana N. ('14) 0; N. ('24) 0; N. ('25) 7.9%; Jam. 0
B. mutabilis N. ('15) 0
B. obliqua N. ('15) 0
Biscutella auriculata N. ('24) 0; N. ('25) 0; Katt. 25%; Roc. 40%
B. cichorifolia N. ('15) 0; N. ('24) 0
B. didyma N. ('25) 0; Katt. 13%
B. laevigata N. ('14) 0; Jam. 11%
B. leiocarpa N. ('15) 0
Brassica arvensis Cun. 99.8%; Gi. 100%; Roc. 100%
B. balearica N. ('15) 30%
B. cernua Gl. 100% N. ('25) 33%; Katt. 100%
B. chinensis N. ('25) 100% Katt. 100%
B. cretica N. ('15) 40%
B. elongata Jam. 20%
B. incana N. ('15) 80%
B. insularis N. ('15) 40%
B. junci Cun. 99%; Gl. 100%; N. (25) 96%
B. macrocarpa N. ('15) 80%
B. napus Wor.; Ravn.; Cun. 83.7%—49.2%; Gl. 0
B. napus var. *oleifera* Cun. 83.7%; Roc. 11%; Jam. 84.8%
B. napus var. *esculenta* Ssach. 0; Roc. 0
B. nigra Hal. +; Ravn. +; Cun. 28.2%; Gl. 20%; N. ('15) 0; Hon. 4.5–62.1%; Roc. 0; Jam. 3.4%
B. oleracea Ravn.; Cun. 94.2—81.6%; Gl. 16.6%; Gi. 0–100%; Roc. 100%
B. oleracea var. *acephala* Cun. 92.8%
B. oleracea botrytis Cun. 88.8%
B. oleracea var. *capitata* Cun. 93.1%
B. oleracea gemmifera Cun. 88.2%
B. rapa L. (= *B. campestris?*) Wor. +; Ravn.; Cun. 100—1.3%; Gl. 0; Ham.: N. ('25) 16.6%; Gi. 75–100%; Roc. 35–50%; Jam. 62.5%
B. pekinensis Da.; Ikeno ('29)
B. pe-tsai Cun. 100%
B. rapifera Hal.
B. robertiana N. ('15) 50%
B. sabularia N. ('15) 100%
B. Tinei N. ('15) 60%
B. tournefortii N. ('15) 0
B. sp. 1 N. ('14) 30%
B. sp. 2 N. ('14) 20%
Braya alpina Jam. 53.3%

Bunias orientalis	N. ('12)+; N. ('13) 2 out of 2; N. ('14) 0; N. ('25) 0; Jam. 28.6%
Camelina dentata	Cun. 100%; Gl. 100%
C. foetida	Katt. 100%
C. linifolia	N. ('15) 90%
C. macrocarpa	Cun. 94.5%
C. sativa	Hal. +; Ravn.; Cun. 80%; Gl. 100%; Mü., 63%; N. ('14) 0; Jam. 91.9%
C. sp.	N. ('14) 100%
Capsella bursa pastoris	Wor.; Hal. ++; Mass.; Ravn. +; Cun. 40.8%; Höst. 62%; Katt. 0; Mü. 95.7%; N. ('12) +; N. ('13) +; N. ('24) +; N. ('25) +; Gi. 78–100%; Rain. 32.5%; Jam. 46%
C. heegeri	N. ('24) 50%; Katt. 90%
Cardamine heterophylla	Gi. 50–100%
C. pratensis	Ravn.; Cun. 100%; Erik. ('26)
Carpoceras sibiricus	N. ('25) 0
Carrichtera vella	N. ('24) 80–100%; Katt. 100%; Roc. 100%
Cheiranthus Allioni	Cun. 76.2%; Gl. 20%; Mü. 99%
C. alpinus	Gl. 0; Jam. 0
C. annus	Hal.
C. Cheiri	Hal.; Sit.; Ravn. +; Naum. +++; Sor. ('21); Höst. 85%; Mü. 99%; Gl. 1.3%; Gi. 78%; Jam. 0
C. incanus	Cun. 0
C. maritimus	Cun. 43.6%
C. scoparius	N. ('15) 50%
C. semperflorens	Mü. 100%
C. Senoneri	N. ('25) 28%
Clypeola jonthlaspi	N. 2 out of 2
Cochleari Amoracia	N. N.; Schl. ('07); Roc. 0
C. anglica	Gl. 100%
C. danica	Gl. 95.59%; Roc. 75%; Jam. 8.6%
C. groenlandica	Gl. 97.56%
C. officinalis	Gl. 86.54%; Mü. 50%; N. ('14) 0; Roc. 0; Jam. 10%–59.3%
Conringia orientalis	Cun. 40.8%, 8%; N. ('15) 20%; N. ('24) 0; Katt. 100%
Coronopus didyma	Gi. 0
C. procumbens	Gi. 0
Crambe abyssinica	N. ('25) 17%; Katt. 100%
C. cordifolio	Cun. 100%; N. ('12)+; N. ('15) 0
C. hispanica	Cun. 6.3%; N. ('15)+; Jam. 20%–98.1%
C. maritima	Cun. 68.2%; N. ('15) 0
C. tartarica	Cun. 100%
Descurainia sophia	Ham.; Gl. 41.67%; N. ('12) +; N. ('13) +; N. ('24) 20%; Katt. 100%; Gi. 75%; Jam. 8.3%
Diplotaxis erucoides	N. ('25) 3.5%
D. muralis	N. ('24) 10%; Gi. 4–100%
D. tenuifolia	Gi. 19–100%
Draba aizoides var. *oblongata*	N. ('14) 0
D. aizoon	N. ('14) 0
D. altaica	N. ('25) 0; Jam. 3%
D. amplexicaulis	Jam. 0
D. androsaxea	Cun. 0
D. armata	Cun. 0
D. borealis	N. ('14) 0
D. caroliniana	Cun. 0
D. corsica	N. ('14) 0
D. dorneri	Roc. 33%
D. fladnizensis	Cun. 50%
D. frigida	N. ('14) 0
D. oxycarpa	Jam. 28.6%–37.5%
D. glacialis	N. ('14) 0
D. rupestris	Jam. 14.7%
D. grandiflora	N. ('15) 0; Jam. 72%
D. haynaldii	Jam. 0
D. hirsuta = *repens* (?)	N. ('12)+
D. hirta	N. ('12)+
D. hispanica	N. ('14) 0
D. incana	N. ('25) 0; Jam. 0
D. johannis	Jam. 9.8%
D. nemorosa	N. ('12) 0
D. montana	Jam. 0
D. nivalis	N. ('14) 0
D. norvegica D. scandinavica	Roc. 26%; Jam. 1.9%–60%
D. pyrenaica	Roc. 63%
D. subamplexicaulis	Roc. 44%; Jam. 0–10%
D. tomentosa	Jam. 0–22.2%
D. verna	N. ('24) 80%
D. wahlenbergi	N. ('14) 0; N. ('15) 0
Eruca cappadocica	N. ('25) 0; Katt. 100%
E. orthosepala	N. ('15) 50%
E. sativa (*vesicaria*)	Cun. 96.3%, 63%; Jam. 18%–27.3%; N. ('15) 90%; N. ('15) 50%; N. ('23) 50%; Roc. 30%; Ravn.; Sit.
Erucastrum obtusangulum	Gl. 6.85%; N. ('25) 0; Katt. 100%
Erysimum altaicum	N. ('15) 0; N. ('25) 2.5%
E. asperum	Hal.; Ravn. +; Cun. 50%
E. aurantiacum	Jam. 48.9%
E. Barbarea	Cun. 76.3%
E. cheiranthoides	Hal. ++; Mass.; Ravn.; Cun. 72.1%; Ham.; Gl. 100%; N. ('12)+; N. ('13)+; Rain. 13.9%; Jam. 11.1%

E. comatum Cun. 0
E. crepidifolium Sit.; Ravn.
E. helveticum Cun. 2.1%; Jam. 4.8%
E. hieraciifolium Gl. 45%; N. ('14) 30%; Katt. 0
E. leptophyllum N. ('15) 20%
E. ockroleucum Cun. 57.2%
E. orientale Gl. 25%
E. parviflorum Cun. 85.7%
E. perowskianum Hal.; Ravn. +; Naum. 0; N. ('15) 0–1.3%; Roc. 62%
E. pulchellum (*E. rupestre*) N. ('15) 0; Jam. 4.3%
E. strictum Appel.; Gl. 14.29%
E. virgatum N. ('15) 1%
Heliophila amplexicaule N. ('24)+; N. ('25) 0; Roc. 46%
Hesperis alpina Roc. 0
H. fragrans Roc. 0
H. lutea Roc. 100%; Jam. 11.3%
H. matronalis Hal. +; Cun. 32.1%; Ham.; Gl. 5.36%; Mü. 100%; N. ('14) 50%; N. ('24) 75%; Jam. 2.8%
H. matronalis var. *nivea* N. ('15) 0
H. matronalis var. *nana* N. ('14) 0; Roc. 0
H. runcinata N. ('15) 0
H. tristis Jam. 10%
H. violacea Jam. 23.6%
Iberis amara Cun. 87%; Gl. 100%; N. ('25) 0; Roc. 18–51%; Jam. 41.4%
I. coronaria Cun. 73.7%
I. gibraltarica Mü. 24.2%
I. hybrida Cun. 52.6%
I. intermedia Gl. 100%
I. lagascana Cun. 47.3%
I. odorata Cun. 41.6%; N. ('25) 0; Jam. 85.1%
I. pinnata Gl. 4.42%; N. ('25) 0; Roc. 82%
I. sempervirens Cun. 43.5%; N. ('25) 0; Jam. 10.5%
I. taurica N. ('15) 30%; N. ('25) 0
I. Tenoreana Jam. 34.3%
I. umbellata Wor.; Hal. ++; Ravn.; Cun. 92%; N. ('15) 0; Gl. 99.14%; Roc. 73%; Jam. 60.7%
I. zenoreana Cun. 2.3%
I. sp. Cun. 100%
Isatis glauca Cun. 68.4%; F. S. ('20) 33%; Roc. 0
I. tinctora Hal.; Cun. 42.5%; Roc. 17%; Jam. 0–9.5%
I. undulata N. ('25) 0; Roc. 0
Jonopsidium acaule N. ('24) 0
Koniga libyca N. ('25) 0
Lepidium apetalum Cun. 52%
L. campestre Hal.; Ravn.; Cun. 42.8%; Gl. 0; Gi. 0–100%
L. draba Jam. 0
L. hirtum N. ('15) 0
L. intermedium Hal.
L. latifolium Jam. 0
L. menziesii Hal.; Ravn.
L. micrantum N. ('13) 50%
L. montanum Hal.
L. perfoliatum N. ('15) 0
L. reticulatum N. ('25) 0
L. ruderale N. ('25) 90%; Gi. 50–75%
L. sativum Cun. 1.8%; N. ('13) 0; Katt. 0; Gl. 0; Mü. 0; N. ('25) 0; Roc. 0; Jam. 0
L. tenuicaule Gi. 0
L. virginicum Hal. ++; Ravn.; Cun. 23%; N. ('25) 17%; Gi. 10%
Lunaria biennis Hal.; Cun. 97.2%; Naum. +; Mü. 100%; Hon. +; Gi. 27–100%
Malcomia africana N. ('25) 0
M. flexuosa N. ('15) 0
M. graeca N. ('15) 0
M. maritima Ravn.; Roc. 9%; Jam. 4.3%–11.1%
Matthiola bicornia Hal. +; Cun. 7.9–4.3%; Gl. 0; N. ('15) 0; Jam. 0
M. fenestralis Gl. 0
M. incana (= *annua*) Wor. +; Hal. 0; Ravn.; N. ('15) 0; Höst. 0; Gl. 0; Mü. 0; Gi. 0
M. oyensis N. ('25) 0
M. tricuspidata Cun. 0; Gl. 0
Melanosinapis communis N. ('24) 0 out of 1; N. ('25) 0
Meniocus ehrenbergii N. ('24)+; Katt. 0
Myagrum perfoliatum N. ('15) 1 out of 1
Nasturtium amphibium Ham. (after Naumann. '13)
N. officinale Gi. 0
N. palustre Hal.; Ham.; Ham.; Gi. 0
N. silvestre Hal.; Mag.; Ham.
Neslia paniculata Cun. 100%; N. ('12)+
Notoceras canariense N. ('24) 75%; Katt. 75%
Peltaria alliacea Jam. 0
P. turmena Jam. 0
Raphanus maritimus N. ('15) 0; N. ('25) 0
R. sp. Gi. 0
R. niger N. ('15) 0
R. odessanus N. ('15) 0; Katt. 15%
R. oleiferus Honig 85–87%
R. radicula Cun. 53.4%; Ssach. 10%
R. raphanistrum Hen. +; Ravn.; Ham.; Mü. 36.4%; Svec. ++; N. ('12) +; Weiss +; N. ('25, '28); Rain. 14.8%; Jam. 39.1%
R. rostratus N. ('14) 1%

R. sativus	Hal. +; Sit. +; Clint. +; Ravn. +; Cun. 37.4%; N. ('13)+; Gi. 0
R. sativus var. *B. niger*	N. ('25, '28) 0; Ssach. 0
Rapistrum hispanicum	N. ('24) 0 out of 1; N. ('25) 1 out of 1; Katt. 60%
R. perenne	Jam. 88.9%
R. rugosum	N. ('24) 2 out of 2
Ricotia lunaria	N. ('24) 1 out of 1
Roripa amoracia	Hal.
R. palustris	Ravn.
R. silvestris	Ravn. Jam. 18.9%
Senebiera coronopus	N. ('24) 0; N. ('25) 0
S. pinnatifida	N. ('24) 0; N. ('25) 0
Sinapis abyssinica	N. ('15) 30%
S. alba (*B. alba*)	Hal. +++; Ravn.; Cun. 100%; Gl. 100%; Mü. 100%; N. ('15) 100%; Rain. 61.4%; Roc. 81%; Jam. 95.6%
S. apula	N. ('25) 90%; Katt. 100%
S. arvensis (*Brassica sinapistrum*)	Hal.; Ros.; Clint.; Roc. 100%; Jam. 58.4%; Mass. +; Ravn.; Cun. 100%; Ham. +; N. ('12)+
S. chinensis	N. ('25) 50%; Roc. 50%
S. geniculata	N. ('15) 0
S. turgida	N. ('15) 95%
Sisymbrium Alliaria	Mass. +; Gl. 0
S. altissimum	Hal.; Cun. 38.3%; N. ('15) 0
S. asperum	N. ('15) 0
S. austriacum	Appel; Gl. 1.25%; N. ('14) 5%; Jam. 12.5–44%
S. bursifolium	N. ('15) 0
S. crepidifolium	Hal.
S. cumingianum	Jam. 21.4%
S. hirsutum	Gl. 25%
S. incisum	Cun. 81.3%
S. irio	Roc. 64%
S. loeselii	Gl. 1.31%
S. officinale	Hal.; Ravn.; Cun. 10%; Erick.; N. ('12)+; N. ('13) 3%; N. ('24) 100%; Gi. 55–100%
S. orientale	Gi. 11–100%
S. Pallasii	N. ('15) 0
S. persicum	Gl. 0
S. polyceratium	N. ('15) 1%
S. sinapistrum	N. ('15) 70%
S. strictissimum	Appel; N. ('15) 0; Jam. 10%
S. taraxacifolium	N. ('15) 30%
S. Thalianum	N. ('24) 10%
S. vulgare	Ravn.
Sophia pinnata	Hal.; Cun. 53.9%
Succovia balearica	N. ('15) 100%; N. ('24) 100%; Katt. 100%
S. balearica Medis	Roc. 100%
Thlaspi alpestre	N. ('15) 20%; Roc. 3%; Jam. 27.3%
T. arvense	Hal. +++; Ravn. +; Cun. 97.6%; Gl. 25.81%; N. ('13) 99%; N. ('24) 66%; N. ('25, '28) 99%; Rain. 99.5%; Jam. 97.6%
T. bellidifolium	N. ('14) 0; Jam. 2.2%
T. kovacsii	N. ('14) 0; N. ('25) 0; Jam. 0–13.7%
T. oliveri	Roc. 0
T. perfoliatum	N. ('15) 40%
Thlaspi sp.	N. ('14) 0
T. violascens	Jam. 9.5%
Turritis glabra	N. ('14) 0; Roc. 79%
Tysanocarpus curvipes	N. ('24) 100%
Visicaria gracilis	N. ('15) 1%

So far 318 species in 59 genera of crucifers have been examined for club root, and among these all but 89 species and 8 genera were found to be infected. It is to be noted that the percentage of infection reported by the various workers for the same species varies considerably. Such differences are largely due to the small number of plants examined. In some species, particularly wild crucifers, the percentages are based on the examination of only two or three plants. This is also true of some of the genera and species which have been reported to be uninfected. Doubtless when a larger number of plants have been examined these species also will be found to be susceptible to club root.

As has been noted elsewhere, club root is limited to the mustard family, and all reports of its occurrence in species outside of the Cruciferae have been disproven. In 1910 Marchand reported a disease of melon, celery and sorrel in France which he thought was caused by *P. Brassicae*, but subsequent examination of these plants by Grignon ('10) showed that the swellings on the roots were caused by the nematode, *Heterodera radicicola*. Griffon and Maublanc ('12) later confirmed Grignon's observations.

Several attempts have been made to infect plants closely allied to the mustard family with *P. Brassicae*, but these have been unsuccessful. In 1897 Halsted tested the following species in New Jersey:

Abutilon abutilon	*Malva rotundifolia*
Agrostemma Githago	*Melilotus alba*
Argemone mexicana	*Papaver* sp.
Chelidonium majus	*Reseda odorata*
Erodium cicutarium	*Saponoria officinalis*
Hibiscus trionum	*Silene noctiflora*

No indication of club root was found in any of these species. Potts ('35) likewise found that non-cruciferous plants, including *Reseda odorata, Carydalis glauca, Fumaria officinalis, Allium schonprösum, Urtica pillulifera* and *Spinacia oleracea*, are unsusceptible.

Control of Club Root

Because of the great economic importance of club root extensive attempts to control the disease have been made for almost a century and a half, but so far no completely effective measures have been developed. The resting spores of the causal organism are produced in prodigious quantities, have a fairly high degree of resistance, and may remain viable in the soil without the presence of host plants for seven to eight years; all of which makes effective control very difficult. Control is furthermore hampered by the wide range of wild and cultivated hosts which harbor *P. Brassicae* and the fact that crucifers are susceptible as long as they are alive.

Control measures against club root have involved sanitary practices, sterilization of the seed, disinfection of soil in seed beds, applications of fungicides to the soil in fields, adjustment of the soil reaction, addition of lime, judicious use of basic fertilizers, soil drainage, crop rotation, eradication of wild cruciferous hosts, and the development of resistance varieties or races of cultivated crucifers.

One of the main factors which makes club root difficult to eradicate in the soil is the longevity of the resting spores. Longevity is not influenced by grazing, crop rotation, plowing, or the application of carbonate of lime and sulphur to the soil, according to Gibbs ('39). Fedorintschik ('35) found that soil from fields which had not been sown to crucifers for seven years contained enough viable spores to infect 26.6 per cent of aseptically grown cabbage seedlings after transplantation. In cabbage fields rested one year the viability of the resting spores was reduced from 81.2 per cent to 43.7 per cent, but in one field rested five years the reduction was only 10 per cent. Plowing the fields two or three times a year has no effect on resting spore viability, according to Fedorintschik. In badly infested fields up to 100 million spores per cc. of soil have been found (Naumov, '28), which may extend to and infect plants at depths of 10 to 30 cms. in soils of various types (Gibbs, '32; Motte, '34; Potts, '35; Fedorintschik). The intensity of attack is directly correlated with the number of spores in the soil, according to Gibbs ('31b) and Fedorintschik, but Naumov ('28) found but little evidence of correlation in Russia. Gibbs found that one plant out of 42 became infected when there were approximately 25,500 spores per seed box, and 43 out of 44 when 530,000,000 spores per box were present. According to Fedorintschik's calculations, less than 10,000 spores per cc. of soil cause isolated attacks on lateral roots but does not reduce the crop weight of cabbage. More than 10,00 spores may cause 50 per cent infection of lateral roots but no reduction in crop weight, while 300,000 spores per cc. of soil usually leads to over 50 per cent infection of the whole root system and reduces the crop weight to 50 per cent.

In the soil the spores are also fairly resistant to fungicides in concentrations low enough to avoid injury to the host. Bremer ('35) found that a 0.5 per cent solution of uspulun poured over spores in the soil killed only 24 per cent to 38 per cent, and that 5 days were required to kill the spores when immersed directly in a 0.25 per cent solution of the same fungicide. Likewise, relatively strong solutions of formalin were ineffectual. Fedovota ('29) found that treatment with 0.1 per cent mercuric chloride has little or no toxic effect on the spores. On the other hand, Honig ('31) reported that 0.001 per cent mercuric chloride when applied directly to the spores caused general plasmolysis, while higher concentrations were more or less ineffective. He also found that solutions of $MgSO_4$, NaCl, KNO_3 and NH_4Cl in molar concentrations of 1:100, 1:10,000, 1:100,000 plasmolysed the spores within 4 weeks. Immersion of spores for 30 minutes in 70° C. water and heating the soil 5 to 30 minutes at 60° to 80° C., renders them inactive (Naumov, '28; Vladimirskaya, '30; Anony., Ger., '39). Polyakoff ('39) reported that immersion of spores for 5 minutes in *condensate* (containing 5 per cent formalin) kills the spores, and that this solution added to the fields at the rate of 1.8 by volume of soil reduces infection 70 to 100 per cent. Desiccation has a marked effect on spore viability, according to Naumov ('25). Spores kept in a relative dry cellar over winter caused infection of seedlings the following spring, but a year later they were no longer viable. If desiccated completely the spores lose their infective power within a year.

SANITARY PRACTICES

Since the spores of *P. Brassicae* will survive passage through the digestive tract of animals and may be carried to the fields in contaminated manure, it is obvious that diseased roots should be thoroughly boiled before being fed to livestock. Stable and liquid manure should be avoided as much as possible, since it is conducive to club root development if applied directly to a crop of crucifers. If it is to be used at all it should be applied during the season preceding a susceptible crop. If contaminated it should be sterilized or disinfected before application to the soil. Vincent, Herviaux, and Cöic ('38) advocated the addition of 90 kg. nitrogen in the form of cyanamide to stable manure before using. It is interesting to note in this connection that Naumov ('28) reported that, contrary to all expectations, the addition of stable manure to the soil exerted a slight detrimental action on the parasite.

Other sanitary practices involve collecting and burning diseased plants. These should not be allowed to rot in the soil or in piles, since this liberates the spores in the soil again. Plowing under of diseased plants to various depths has been advocated. Frank ('96), Potter ('97), L. R. Jones ('01), Laubert ('05a), Köck ('11) and Lindner ('11) recommended a depth of 1 meter; Naumann ('13), Neger ('17), Trieschmann ('17) and Ludwigs ('25) 80 cms.; and Müllers (Honig, '31) 20 to 30 cms. The latter depth is obviously inadequate, since it has been shown that infection may occur at 30 cms. Esmarch ('24) contended that burial is worthless and that burning is the only safe method of disposal.

Young seedlings may often be infected and not show recognizable symptoms of the disease at the time of transplanting. Careful examination of the plants at the time of removal from the seed beds is therefore essential if there is any suspicion that the disease may be present. Should a single seedling from a seed frame show symptoms of club root it is advisable, in the opinion of Schlumberger ('14), Chupp ('25) and Gleisberg ('26), to avoid or destroy all plants from that particular bed, since it is only rarely that infected seedlings recover.

Seed, Seed Bed and Seedling Disinfection.—Seeds of infected crucifers occasionally bear the fungus spores externally, and in such cases seed sterilization is necessary. Soaking seeds in tillantin B and 0.25 per cent to 0.5 per cent uspulun for one-half to one hour before planting has been reported by Mothes ('25), Brönnle ('26) and Leines ('26) to reduce the incidence of infection if followed by fungicidal treatment of the soil. Such seed treatment, however, is worthless unless it is followed by seed bed disinfection.

Various fungicides and chemicals as well as heat have been used in seed bed disinfection. Heating the soil $\frac{1}{2}$ hour at 60° C. or above kills the spores, according to Vladimirskaya, Jorgensen, and Shewell-Cooper. Commercial formalin (1 part to 40), 0.05 per cent to 0.2 mercuric chloride (1 to 2 gals. per sq. yd.), 0.1 per cent to 0.5 per cent liquid ceresan, corrosive sublimate (1 oz. in 2–10 gals. water), 0.5 per cent uspulun solution, uspulun and solibar mixed (1 to 5), 10 per cent solution of washing soda, folosan and brassican (18 oz. per cubic yard of soil) mixed with lime, carbolic acid, mustard oil, etc., applied 1 to 5 times to seed beds have been reported to reduce or completely control seedling infection by the following workers: Anony. (Australia, '40), Sommer ('22), Jorstad ('23), Bremer ('23–'24), Darnell-Smith ('24), Kindshoven ('24), Chupp ('25), Hofferichter ('26), Clayton ('26), Blunck ('28), Osterwalder ('29), Preston ('30), Hoffman ('32), Jorgensen ('33), Gibbs ('34), Woodman, Benchley and Hanley ('34), Küpke ('35), and Smieton ('39).

Effective control has been reported from the use of uspulun on seed beds, but some workers have claimed that it is less satisfactory than mercuric chloride. According to Clayton ('26) the spores of *P. Brassicae* in the soil are fairly sensitive to mercuric compounds, but such substances have been found to be more or less toxic to the host, especially in dry hot weather, and may reduce the crop to some extent. Wellman ('30), however, found that mercury compounds used according to Clayton's methods were ineffective in Wisconsin unless applied in concentrations high enough to be injurious to the host. Copper carbonate and sulphate, and carbonates and sulphates of calcium were likewise ineffective. Hydrated lime worked into the soil at the rate of 1,500 pounds to 5 tons per acre gives good control in seed beds, according to Wellman. Motte ('34) found that the fungus spores rarely exceed a depth of 20 cms. in the soil, and as a control measure for seed beds he advocated removal of the upper 25 cms. of soil.

Seedling disinfection alone before or at planting has not proven generally practical in controlling club root. Dipping seedlings up to the collar in weak solutions of uspulun, mixtures of uspulun and solibar solutions (1:5), mercuric chloride, 0.1–1.5 per cent liquid ceresan, etc., before planting has been recommended by Kindshoven ('24), Preston ('29), Rabbas ('30), Küpke ('35), and others, but Jamalainen ('36) asserted that seedling treatment at and after planting is ineffective. While such disinfectants may inactivate the spores in the soil adhering to the roots and root hairs, they obviously cannot destroy the amoebae and plasmodia within the tissues, if such stages are already present, without killing the host. It is doubtful that enough fungicide will remain on the roots during transplantation to kill or inactivate the spores which may be present in the plant holes. Seedling treatment, as recommended above, must obviously be followed by soil disinfection in the field to be effective.

The addition of 1, 2, and 25 gms. uspulun dust per plant hole (Esmarch, '25; Blunck, '28), 1 liter of .25 per cent uspulun solution, 10 liters of .20 per cent uspulun, tillantin B, and germisan per plant (Lindfors, '25; Hertel, '26; Rabbas, '30), 10–15 gms. humus carbolineum per plant (Popp, '25), $\frac{1}{2}$ pt. .01 per cent (or 1 oz. in 6 gals. water) corrosive sublimate per plant (Preston, '27; Holmes-Smith, '30), chloropierin in plant holes (Anony., Rhode Island, '39), $\frac{1}{2}$ pint .062–.1 per cent mercuric chloride per plant (Preston, '29; Olgilvie and Mulligan, '34; Smieton, '39), and other chemicals have been reported to reduce or completely control infection. Preston ('28) found that $\frac{1}{2}$ pt. per plant of .2 per cent methyl green, malachite green, methyl violet, and Brilliant green applied at planting was ineffective. Likewise clubicide and Cheshunt Brown compounds as well as .2–.5 per cent formalin and .2 per cent lysol were unsatisfactory for seedling treatment at and after transplanting.

Soil Disinfection in the Field.—In attempts to combat club root in the field by soil disinfection a wide assortment of chemicals, fungicides and special remedies have been employed as is shown in table 2 and the accompanying pages. In pots, seed beds, small gardens, and experimental plots these substances are fairly effective, but with the exception perhaps of uspulun they have not proven commercially satisfactory and expedient in the field. As Larsen and Walker ('34) have pointed out, greenhouse pot tests are not always a true index of what may be expected in the field. The cost of materials and expense of application often outweigh the beneficial results obtained, and in many instances the fungicides directly injure or reduce the crop. According to Motte ('33) very little is now being done to combat the disease in Denmark beside avoiding manure, using basic fertilizers, and growing resistant varieties.

TABLE 2. *Showing the effects of fungicides, chemicals, and special remedies on club root in the field.*

	Uspulun	Mercuric chloride	Formalin	Sulphur	Corrosive sublimate	Carbolineum and humus	Lime sulphur dust
Anon., U. S. A., '22	.....	.....	Effective	Effective	.....	.....	.....
Anon., Ger., '39	.....	0.15 gm. per l. of soil—effective	1.5 cc. per l. of soil— ineffective	.....	.....	.....	.....
Appel, Schlumberger, '13, '14	Effective	.....	10% sol.—ineffective	.....	.....	Ineffective	.....
Arker, '35	.25% sol., 500 l. per acre— fair control	.....	.....	.....	.....	.....	.....
Blunck, '19, '28, '29	20 gms. per plant hole—effective	.....	.04% sol.; 4.1 l. per sq. m.—ineffective	.....	.....	Ineffective	.....
Braun, '26, '27	Plus basic fertilizers—effective	.....	.....	.....	.....	.....	.....
Bremer, '23, '28	1–0.5 gms. per kg. soil—effective	.....	.....	.....	.....	6–10 cc. florium per kg. of soil—effective	.....
Brick, '13, '16	.....	.....	1% sol.; 5 l. per sq. m.—ineffective	Ineffective	.....	.....	Ineffective
Bronnle, '26	0.5% sol.—effective	.....	.....	.....	.....	.....	.....
Burkhardt, '15	.....	.....	Effective	.....	.....	.....	.....
Clupp, '25, '28	.....	.....	Effective	320–640 lbs. per acre—ineffective	1 oz. per 10 gals. water—good control	.....	.....
Clayton, '26	.....	1:1,200–1:2,000 sol. 1 gal. to 30 ft. row—effective	.....	.....	.....	.....	.....
Dankler, '19	.....	.....	.....	.....	.....	Effective	.....
Eggemeyer, '20	.....	.....	.....	Ineffective	.....	.....	Ineffective
Esmarch, '25	.....	.....	.....	.....	.....	.....	Effective
Flachs, '30	Effective	.....	.....	.....	.....	.....	.....
Flachs, Kronberger, '30	Effective	.....	.....	.....	.....	.....	.....

Gibbs, '31, '39		0.4% acidulated; 2 gals. per sq. yd. good control		300 lbs. per acre—ineffective			
Gloyer, Glasgow, '24					1:1,280 and 1:2,000 sol. effective		
Halsted, '96 '99		0.05% sol. 5,280 gals. per acre slightly effective		300–1,200 lbs. per acre—ineffective	33 lbs. per acre ineffective		
Hammarlund, '15			1% sol. 10 l. per sq. m. effective				
Herpers, '25	Effective						
Hiltner, Korff, '16						Effective	
Hollrung, '23				Ineffective (?)			
Holmes-Smith, '30 '31					0.05% sol.—excellent control		
Honig, '32	100, 200, 300 gms. per sq. m. reduced infection 42.4%						
Hostermann, '21	0.1 gm. per l. of soil good control						
Jamalainen	0.25% sol.—effective	0.1% sol.—effective	10%, 100 cc. per sq. m.—effective			Effective	
Jorgensen, '33	Effective		Effective				
Kindshoven, '24, '28	120 gms. per sq. m. good control		Effective				
Kirshner, '27			Effective				
Koblischek, '29	Plus alkaline fertilizers effective						
Kohne, '28	70 gms. per sq. m. very effective						
Knorr, '20							Effective
Leines, '26	Dust less efficient than solutions						
Lindfors, '21, '25	0.25% sol. 1 l. per plant good control	Effective	0.1% sol.; 10 l. per sq. m. ineffective				

TABLE 2. *Concluded.*

	Uspulum	Mercuric chloride	Formalin	Sulphur	Corrosive sublimate	Carbolineum and humus	Lime sulphur dust
Loew, '12	.....	.....	.....	.....	.....	Effective	.....
Ludwigs, '25	Effective	.....	.....	.....	.....	.....	.....
McLeod, Howatt, '34	.....	10–15 lbs. per acre —effective	.....	.....	.....	.....	.....
Merkenschlager, '24	Effective	.....	.....	.....	.....	.....	.....
Müller, Osterwalder, '19, '23	Fairly effective	.....	3% sol.; 5 l. per sq. m.—ineffective	Ineffective	.....	.....	Ineffective
Olgivie, Mulligan, '34	.....	½ pt. per pl. of 1:1,600 sol.—effective	.....	.....	.....	.....	.....
Osterwalder, '29	.....	.....	.....	Ineffective	.....	.....	.....
Palmer, '41	.....	1%-.075% sol.—effective	.....	.....	.....	.....	.....
Popp, '19	.....	.....	10%; 400 cc. per 4 sq. m.—ineffective	.....	.....	Effective	.....
Preston, '27–'31	0.5% sol.—fairly effective	0.1% sol.—effective	.....	.....	½ pt. per plant of 1 oz. to 6 gals. water—effective	.....	.....
Rabbas, '30	0.25% sol.—good control	.....	.....	.....	.....	.....	.....
Riehm, '24, '25	Dust mixed with soil—good control	.....	.....	.....	.....	.....	Good control
Schaffnit, Lustner, '20	Effective	.....	.....	.....	.....	Effective	Effective
Trieschmann, '17	.....	.....	Effective	.....	.....	.....	.....
Vielhauer, '20	.....	.....	.....	.....	.....	Effective	.....
Vilkaitis, '33	Partially effective	.....	.....	.....	.....	.....	.....
Vladimirskaya, '30	.....	Effective	.....	.....	.....	.....	.....
Vogt, '24	Effective	.....	.....	.....	.....	.....	.....
Wahling, '22	Effective	.....	.....	.....	.....	Effective	.....
Wilson, '34	.....	Effective	.....	.....	.....	.....	.....

The results shown in table 2 are contradictory in several cases. This is doubtless due in many instances to outside factors such as those which influence the effectiveness of lime and other basic fertilizers. Differences in time and methods of application, degree of soil infestation, soil moisture, etc., obviously operate here also. As is shown in table 2 uspulun has been extensively used, especially in Europe, and when applied at rates of 0.5 to 1 gm. per kg. of soil or 120 to 300 gms. per sq. m. in the field two weeks or more before planting it is the most effective and practical of all fungicides for the control of club root, according to the data in the literature. On the other hand, numerous workers have reported it to be unsatisfactory. It may be used as a solution and poured over the soil or as dust mixed with fertilizers, but Honig ('31) stated that its effect is less certain and complete when used in solution. Whether or not uspulun will prove practical in large-scale operations is uncertain, according to Blunck ('29), but Honig claimed that its practicability in this respect has already been demonstrated. In combination with solibar, lime and other basic fertilizers its use may be greatly extended, but even when mixed with soil alone it is too expensive for practical purposes, according to Riehm ('25).

The effect of uspulun on the parasite and host is not definitely known. Whether it kills the spores or prevents germination is uncertain. Bremer ('23) found that 1 gm. per kg. of soil destroys about one half of the spores, and held that it acts primarily in killing the amoebae. Honig ('31) believed that uspulun may possibly stimulate spore germination and kills the amoebae as they emerge, or that it increases the resistance of the host, along with a weakening of the amoebae.

Mercuric chloride is generally reported to be effective, but whether or not it is economically practical in large-scale operations is still uncertain. *Formalin* has been extensively employed, but the results obtained are very conflicting, as is shown in table 2. Its efficacy in the field is doubtful, and Hammarlund ('15), Burkhardt ('15), and Lindfors ('24) stated that it is too expensive for commercial use.

Sulphur has proven ineffective, and in only a few instances has corrosive sublimate reduced infection. *Bordeaux mixture* is also of little or no value in combating club root. *Carbolineum* alone and mixed with various types of humus, however, has been reported to be fairly satisfactory.

In addition to fungicides listed in table 2 various other chemicals, substances, and remedies have been used in combating club root. These have been used singly or in combination, and with or without alkaline fertilizers, but here again the results obtained are contradictory and generally unsatisfactory.

Segetan, a mercury compound, is ineffective according to Osterwalder ('29).

Cresol (2 kg. per l. of water) applied at the rate of 2.5 l. per c. m. of soil is effective, according to Loew ('12), but 50 gms. per cu. m. of soil has no effect.

Liquid ammonia, 1 per cent solution, has no inhibitory properties (Osterwalder, '29).

Sulgine is worthless, according to Lindfors ('24).

Soot or *lampblack* has been used in England to control club root, according to Woronin ('78), but Eggemeyer ('20) found it to be useless.

Petroleum was reported to be effective by Pfeiffer and Staes ('02). Müllers (Honig, '31) got 80.44 per cent healthy plants by its use in Germany.

Chloropicrin in the plant holes or added to the soil reduces infection in cabbage to 4 per cent or less (Anony., R. I., '39).

Pure carbolic acid added to the soil completely eliminates club root from experimental plots, according to Jorgensen ('33).

Mustard oil (3 cc. per l. of soil) gives complete control (Anony., Ger., '39).

Parachlorbenzine gives only partial control and injures the host plants (Vladimirskaya, '30).

Germisan, 20 gms. in 10 liters of water per plant is not effective, or only partially so (Hertel, '26; Vilkaitis, '33).

Sulcan is less satisfactory than Beka-Wurzelschutz (Esmarch, '25) but sufficiently effective for practical purposes.

Potassium and *calcium permanganate* applied directly to the soil are ineffective. (Müller and Osterwalder, '23, '24; Osterwalder, '29).

Mercurous chloride is less satisfactory than mercuric chloride, according to Bailie and Muskett ('33), but Preston's ('31) earlier report contradicts their results. Palmer ('41), however, secured striking control in cabbages with mercurous chloride suspended in water with the aid of gum arabic at the rates of 5 and 7.5 lbs. per acre.

Folosan (pentachlornitrobenzine) and *brassisan* (trichlornitrobenzine), 18 oz. per c. yd. of soil, are superior to an equal concentration of mercuric chloride in seed boxes, but in the field they are less effective (Smieton, '39). All three compounds check growth to some extent, but nonetheless give good control. Folosan and brassisan are more effective when used with lime. Brown ('35) likewise found brassisan to be effective against club root.

Semesan is equally as effective as mercuric chloride (Clayton, '26).

Liquid ceresan, 0.1–0.15 per cent, applied to seed bed at time of planting, to seedlings a day or two before transplanting, and 6 to 8 days after setting out gives excellent control, according to Küpke ('35).

Tillantin B in solution sprinkled over seed beds gives complete control, according to Mothes ('25). Hertel ('26), however, reported that 20 gms. per 10 l. water poured over each plant or used as dust in conjunction with uspulun are ineffective. Likewise, tillantin B dust alone (100 to 150 gms. per sq. m. of soil) has no effect on club root, according to Blunck ('28).

Cheshunt compound, clubicide, and 0.2 per cent *lysol* are ineffective, according to Preston ('28). *Carbon bisulphide* when applied to the soil is also ineffective (Müller and Osterwalder, '24).

Copper sulphate powder applied at the rate of 600 and 1,200 lbs. per acre (Halsted, '96), or as a solution (1:1,664, 1 gal. per 30 ft. row) directly to growing plants (Gloyer and Glasgow, '24) has no effect on club root. Müller and Osterwalder ('24) got similar negative results.

Bewley's solution (2 oz. copper sulphate and ammonium carbonate) applied in a concentration of 1 oz. to 2 gal. water increases infection (Gloyer and Glasgow, '24).

"Höchst mittel," according to Hertel ('26) reduces infection considerably, but Blunck ('28) found that 150 gms. per sq. m. of soil is ineffective. He also found Elhardt's *Wurzelschutz* and *florium* (150 g. per sq. m.) to be of little value against club root.

Copper carbonate is reported to be fairly effective by Naumov ('27) and an anonymous worker in the U. S. ('22), but Vladimirskaya ('30) got only partial control with it.

Red copper oxide is fairly effective, according to Naumov ('27) and McLeod and Howatt ('34).

Lime copper dust increases infection, according to Gloyer and Glasgow ('34).

Sodium carbonate, 3,000 lbs. per acre is ineffective, according to Halsted ('96). Lindfors ('24) confirmed Halsted's results, but Naumov ('27) and Vilkaites ('33) found it to be slightly effective.

Bordeaux mixture alone in amounts up to 5,280 gals. per acre or mixed with corrosive sublimate has little or no effect on club root, according to Halsted ('96, '99), but later an anonymous worker (U. S. A., '22) reported it to be effective.

Sodium chloride, 300 to 600 lbs. per acre, has no appreciable effect on club root, according to Halsted ('96). Naumov ('97), however, found that calcium and barium salts (K_2CO_3, NaOH, KOH, and $Ba(OH)_2$) are to some degrees effective, while $CaCl_2$ and $BaCl_2$ are of little value. Wellman ('30), on the other hand, reported that K_2CO_3 does not inhibit club root.

Radium, x-ray, and *ultraviolet* light treatments are reported by Petri ('24) to be effective in reducing club root infection.

LIMING

Liming the soil before planting appears to be the most widely used and practical control measure in the field, although numerous workers have failed to secure satisfactory results by such treatment. Who first discovered the efficacy of lime is not known, but Ellis reported that before 1742 farmers in England had been using clay or marl for dressing diseased fields before planting turnips. In 1831 Farquharson advocated the addition of powdered lime shells to manure before using, while Abbay (1831) recommended the addition of 256 bushels of "knottingsley" lime per statute acre as a control measure. Subsequent workers, including Anderson ('55), Hunter ('57), A. Voelcker ('59), and Henderson ('67), of this early period also noted the great prevalence of club root in lime-free soils and reported varying degrees of control with the addition of lime, ground oyster shells, and flour of bone to the soil, but they found that the effectiveness of these substances varied markedly and that all kinds of lime were not equally effective. At the close of the 19th century numerous other pathologists, including J. A. Voelcker ('94), Eycleshymer ('91), Sommerville ('94–'97), Massee ('95), Halsted ('96–'99), Seltensperger ('96), Potter ('96–'97), Sitensky ('98), Gilchrist ('98–'00), L. R. Jones ('01), and others reported varying beneficial results from the use of lime. Halsted, in particular, carried out an extensive series of tests in America, and after seven years of field experimentation concluded that air-slaked lime at the rate of 75 bushels per acre was commercially satisfactory as a control measure. Later, however, Cunningham ('14) reported that 150 bu. per acre were necessary for effective control. Extensive experiments along the same line were carried out in Denmark by Ravn and his associates ('02–'11) with calcium carbonate and calcium oxide in quantities varying from 2.5 to nearly 10 tons per acre. They found that the largest treatments were the most effective, and although infection still occurred the crops produced were commercially satisfactory. Following these long-time experiments of Halsted and Ravn, beneficial results from the use of lime in the field have been reported by numerous workers, including the following: Diakanoff ('11), Brick ('13), Cunningham ('14), Georgeson ('16), Bos ('18), Weiss ('18), Popp ('19), Müller-Thurgau and Osterwalder ('19, '23), Janson ('20), Whitehead ('22, '36), Jorstad ('23), Katterfeld ('23), Harter and Jones ('23), Bremer ('23–'24), Hollrung ('23), Anony. (Nova Scotia, '23), Montieth ('24), Darnell-Smith ('24), Lindfors ('24), Kindshoven ('24), Naumov ('25, '27), Tennent ('25, '30), Siemaszko ('25), Riehm ('25), Gleisberg ('26), Tessenow ('26), Vaughan and Wellman ('26), Appel ('27), Chupp ('28), Martin ('28, '34), Blunck ('28), Wellman ('30), Rabbas ('30), Gibbs ('31, '32), Anony. (Germany, '31), Kreuzpointer ('31), Beaumont and Staniland ('33, '34), Nielsen ('33), Wilson ('34), Potts ('35).

Arker ('35), Brown ('37), Murphy ('37) and Bennett ('39).

On the other hand, unsatisfactory and inconclusive results from the use of lime as a control measure have been reported by the following workers: Potter ('97), Hiltner ('08), Naumann ('12, '13), Appel and Schlumberger ('13), Schlumberger ('14), Pettera ('17), Janson ('20), Eggemeyer ('20), Vielhauer ('20), Vogel ('22), Whitehead ('22), Lindfors ('24), Esmarch ('25), Korff and Böning ('27), Flachs and Kronberger ('30), Vilkaitis ('33), Motte ('33), Bailie and Muskett ('33), and Jamalainen ('36).

The amount of lime used and recommended by many of these workers varies greatly, and this may partly explain some of the inconsistencies in the results obtained. The investigators listed below have used and advocated the following quantities of lime in the control of club root:

Abbay (1831), 256 bu. per acre.
Hunter ('57), 14–16 tons per acre.
Sommerville ('94), 700 lbs. per acre in drills with seed.
J. A. Voelcker ('94), 2 tons per acre.
Stewart ('95), 90 bu. per acre.
Mathieu-Sanson ('97), 400 liters per acre.
Hawk ('98), 6–8 tons per acre.
McAlpine ('03), 0.32–0.67 liters per sq. m.
Laubert ('05), 1.5 kg. per sq. m.
Schlumberger ('14), 2–3 kg. per sq. m.
Burkart ('15), 0.5–0.6 gm. per sq. m.
Neger ('17), 0.5–1.0 kg. per sq. m.
Trieschmann ('17), 2–3 kg. per sq. m.
Popp ('19), 0.5–0.6 kg. per sq. m.
Böhmer ('22), 1.4 kg. per sq. m.
Höstermann and Noak ('23), 0.5–0.6 kg. per sq. m.
Darnell-Smith ('24), 150 bu. per acre.
Herpers ('24), 0.25 kg. per sq. m.
Beyer ('25), 0.5 to 0.6 kg. per sq. m.
Tessenow ('26), 400 gms. per sq. m.
Gleisberg ('26), 0.5–0.6 kg. per sq. m.
Kirschner ('27), 1–2 kg. per sq. m.
Blunck ('29), 1–2 kg. per sq. m.
Albert ('31), 1–4 tons per acre.
Anony. (Australia, '40), 2 tons hydrated lime per acre.
Stubbs ('41), 1–2 tons per acre.

The majority of workers listed above did not specify the kind of lime used, and it is impossible to determine whether they used pure calcium hydrate, air-slaked lime, carbonate of lime, etc., or calcium cyanamide. Since all kinds of lime are not equally effective in controlling club root many of the differences in results reported in the literature are doubtless due to this factor. Soil differences, degree of spore infestation, environmental conditions, soil moisture, variations in technique and time of lime application before planting, use of manure and acid fertilizers with lime, etc., are factors which may influence the effectiveness of lime, and unless they are kept as constant as possible in experimental work, differences in results are certain to occur. That such factors are important is well shown by the precautions recommended for the use of lime. Schlumberger ('14), for instance, claimed that lime is effective only if the soil is thoroughly aerated at the time of application, while Larsen and Walker ('34) reported that aeration in relation to liming increases infection. They also found that fluctuations of soil moisture at a relatively low moisture content influenced the degree of infection in limed soils. Appel and Schlumberger ('14) noted that liming becomes less effective on a given plot the second year, and Lindfors ('24) asserted that lime is ineffective if the disease is already present. If not, lime is a good club root inhibitor. Murphy ('27) maintained that lime does not take effect until the third or fourth year after application, and Kreuzpointer ('29) stated that liming and other control measures are worthless if stable and liquid manure are used in conjunction. All of these reports as well as others to be found in the literature, show that several factors operate and influence the inhibitory properties of lime.

Some of the workers who have specified the kind and quantity of lime used are listed in table 3, which is obviously very incomplete because much of the European and Asiatic literature has not been available since the present war began. Table 3 shows quite clearly that the amount of lime used and recommended as well as the effects produced vary greatly. Calcium hydrate is generally believed to be the most effective, but Walker and Larsen ('35) found that calcium cyanamide is about twice as effective as $Ca(OH)_2$ in reducing infection in cabbages. Martin ('34) and Haenseler and Moyer ('37) have likewise found calcium cyanamide to be effective when used alone, and when used in combination with calcium hydrate the decrease in clubbing was even greater. Wellman ('30) got complete inhibition with calcium hydrate, and found that limes consisting of $CaCO_3$, and $CaSO_4 \cdot 2H_2O$ are not good club root inhibitors. On the other hand, limes which are of CaO or $Ca(OH)_2$ composition are good inhibitors. The effectiveness of air-slaked lime varies greatly. The relative amounts of hydrate and carbonate in air-slaked lime varies considerably depending on the conditions under which the oxide is slaked, and this factor doubtless influences its effectiveness. Burnt quick lime (CaO) is usually beneficial, but calcium carbonate is generally regarded as ineffective. Although Massee and Carricklee reported gas lime to be inhibitory it has been found to be of little or no value (Halsted, '96–'99). Calcium chloride not only fails to arrest club root infection but also reduces the crop materially. Raw ground limestone is reported to be effective (L. R. Jones, '01), but Wellman ('30) found no inhibitory effects by its use. Later, however, Larsen and Walker ('34) reported that finely ground dolomitic limestone distinctly inhibited infection when applied in sufficient quantity to bring the pH up to 6.9, and completely prevented infection at pH 7.2 and 7.6.

In Germany and other countries of Europe a patented preparation called Steiner's remedy, consisting of relative proportions (Popp, '19b) of lime, ashes, and refuse or waste, has been used with considerable success in controlling club root. In addi-

TABLE 3. *Showing the types and quantities of lime recommended and their effects on club root.*

	Calcium hydrate (hydrate of lime)	Slaked lime	Calcium cyanamide $CaCN_2$	Calcium carbonate $CaCO_3$	Calcium oxide (ground quicklime)	Quicklime CaO	Gas lime	Calcium chloride	Raw ground limestone
Anon. ('23), Germany	Good control								
Anon. ('33), Conn., U. S. A.	1½–2½ tons per acre—good control								
Appel, Schlumberger, '13–'14								Ineffective	
Bailie and Muskett ('33)		No satisfactory control							
Beaumont and Staniland ('33, '34)	10 tons per acre—complete control					8 tons per acre—effective control			
Becker ('37)			Effective						
Blunck ('28)	1–2 kg. per sq. m.—good control								
Bremer ('23)	30 gms. per k. soil—effective control								
Carrieklee ('03)							Complete control		
Chupp ('26)	3,000 lbs. per acre—effective control								
Cunningham ('14)		(Air slaked) 150 lbs. per acre—good control						650 lbs. per acre—not effective	
Eriksson ('26)				Not effective					
Esmarch ('25)		Not very satisfactory						Not effective—heavy reduction in crop	
Gibbs ('31, '32, '39)		(Air slaked) 2 tons per acre—good control		3–5 tons per acre—effective control					

Haensler, Moyer, '37	3,000 lbs. per acre—good control at pH 7.3	.....	1,200 lbs. per acre good control; 1,200 lbs. $CaCN_2$ + 3,000 lbs. $Ca(OH)_2$ best control	.....	.....	.....	.....	.....	.....
Hall ('04)	.....	.....	.....	.....	.....	3–4 tons per acre—effective control	.....	.....	.....
Halsted ('97)	.....	37.5–150 lbs. per acre good control	.....	.....	.....	.....	150 bu. per acre—not effective	.....	.....
Harter and Jones ('23)	.....	75 lbs. per acre good control	.....	.....	.....	.....	.....	.....	.....
Jamalainen ('36)	.....	2 kg. per sq. m.—not effective	.....	.....	.....	.....	.....	.....	.....
Jones ('01)	.....	.....	.....	.....	.....	.....	.....	.....	(Stone lime) 80 bu. per acre—good control
Kindshoven ('24, '28)	.....	.....	¼ kg. per cubic m.—good control	Not very effective	.....	.....	.....	.....	.....
Kreuzpointer ('31)	.....	.....	Complete control	.....	.....	Complete control	.....	.....	.....
Larsen and Walker ('34)	2–4 tons per acre—effective control	.....	.....	Effective control if sufficient to bring pH to 7.0–7.2	Effective control if sufficient to bring pH up to 7.0–7.2	.....	.....	.....	Effective control at pH 6.9, 7.2, and 7.6
Loew, '12	.....	.....	.....	.....	.....	.....	.....	300 gms. per sq. m.—effective	.....
Martin ('34)	3,000–6,000 lbs. per acre 96.9–100% control	.....	600–1,200 lbs. per acre; 60–94.4% control	.....	.....	.....	.....	.....	.....
Massee, '96, '03	.....	.....	.....	.....	.....	.....	Effective	.....	.....
Motte ('33)	.....	Not effective	.....	.....	.....	.....	.....	.....	.....
Muller and Osterwalder ('19, '23, '24)	1.5 gms. per sq. m.—good control	.....	Not effective	.....	.....	.....	.....	.....	.....

TABLE 3. *Concluded.*

	Calcium hydrate (hydrate of lime)	Slaked lime	Calcium cyanamide $CaCN_2$	Calcium carbonate $CaCO_3$	Calcium oxide (ground quicklime)	Quicklime CaO	Gas lime	Calcium chloride	Raw ground limestone
Nielsen ('33)	.....	.....	.....	18,000 kilo. per hectare—good control	.....	18,000 kilo. per hectare—good control	.....	.....	.....
Naumov ('27)	.....	2 gms. per 100 cc. soil—no effect	.....	Not effective	.....	.....	.....	.....	.....
Potts ('35)	.....	.....	.....	.....	.....	Effective control	.....	.....	.....
Preston ('30–'32)	2 tons per acre—good control	.....	1 ton per acre—ineffective	.....	.....	.....	.....	.....	.....
Preston ('03)	.....	No control	.....	.....	.....	.....	.....	.....	.....
Rabbas ('30)	.....	.....	.....	.....	.....	1 kg. per sq. m.—effective control	.....	.....	.....
Ravn ('10)	.....	(Air slaked) 2 tons per acre—effective control	.....	.....	.....	.....	.....	.....	.....
Renard ('35)	.....	.....	300–500 kg. per hectare—good control	.....	.....	.....	.....	.....	.....
Shewell-Cooper ('32)	.....	.....	300 lbs. per acre—good control	.....	.....	.....	.....	.....	.....
Tennant ('30)	.....	.....	.....	.....	.....	16 tons per acre—good control	.....	.....	.....
Vilkaitis ('33)	.....	Inconclusive results	.....	.....	.....	.....	.....	.....	.....
Vladimirskaya ('30)	.....	.....	.....	.....	.....	.....	.....	Only slightly effective	.....
Vogel ('22)	.....	.....	Not effective	.....	.....	.....	.....	.....	.....
Walker and Larsen ('35)	3,000–4,000 lbs. per acre—good control	.....	440–880 lbs. per acre—good control	.....	.....	.....	.....	.....	.....
Wellman ('30)	1,500–2,000 lbs. per acre—good control	2–2½ tons per acre—not effective	.....	Not effective	.....	Effective	.....	.....	2–4½ tons per acre—not effective
Wilson ('34)	Effective control	.....	.....	.....	.....	.....	.....	.....	.....

tion, it is said to lead to a more richly branched and filamentous root system on the host (Appel and Schlumberger, '13, '14; Müller and Osterwalder, '23). It is generally applied 10 cms. deep on infested soil (Naumann, '13; Schlumberger, '14; Hiltner and Korff, '16; Popp, '19; and Höstermann and Noak, '23), or at the rate of 1,000 cu. m. per hectare. Naumann ('12, '13), Schlumberger ('14), Hiltner and Korff ('16), Neger ('17), Popp, Schmid ('19), Schaffnit and Lustner ('20), Höstermann and Noak ('23), and others have reported beneficial effects from the use of this preparation. Appel and Schlumberger ('13, '14), however, found it to be unsatisfactory, and claimed that its effectiveness is dependent to a large degree on the weather. While Steiner's remedy has proven to be fairly effective in controlling club root, the detailed and bothersome method of preparing and applying it as well as the high cost of materials and transportation have not made it popular and commercially profitable.

As to the most suitable time and method of applying lime for club root control various practices and recommendations are to be found in the literature. Eycleshymer ('91), Halsted ('96, '97), Laubert ('05), Schlumberger ('14), Chupp ('25) and Koblischek ('29) advocated application 3 to 6 months before planting, while Erickson ('26) recommended 6 to 18 months. Gibbs ('32) found that if planting took place too soon (less than 3 months) after liming, club root occurred $2\frac{1}{2}$–4 inches below the surface, while in fields planted 12 months after liming infections were present only at 9–$11\frac{1}{2}$ inches. Gibbs accordingly advised at least 3 and up to 12 months between planting and liming. As to the method of application, Sommerville ('94), Seltensperger ('96), Roger ('12), Koblischek ('29), and others recommended drilling lime in with the seed or applying it directly to the plant holes at the time of transplanting. According to Ravn ('12, '13) it is immaterial whether the lime is hoed in or plowed under. Since infection appeared to be prevented to the depth to which lime penetrates, Cunningham ('14) advocated mixing the lime down to 6 to 9 inches in the soil. Müller and Osterwalder ('23) also advised a thorough mixing of lime and soil.

The manner in which lime operates against club root is not very well understood. Most workers have believed that it prevents spore germination by raising the H-ion concentration of the soil, since the spores germinate more readily in acid media, according to numerous investigators. Likewise, it has been reported that infection rarely occurs above pH 7.2 to 7.4 (Chupp, '28). That an increase in pH is not the sole determinating factor is indicated by the report of Honig ('31) that spores will germinate equally well in alkaline media, and by the fact that club root may occur in a high pH environment. Whether or not lime has a direct toxic and lethal effect on the resting spores is uncertain. Bremer ('23) refuted this idea, but later Whitehead ('36) claimed that lime is toxic. Naumov ('27) found that calcium hydrate has no immunizing effect on cabbage, and concluded that its controlling effect is due to direct action on the parasite itself. Seedlings which had been grown for two months in sterile soil with a very high lime content were as susceptible as the controls when transferred to infested soil. Naumov also found that salts of other metals, particularly barium and magnesium, have a controlling effect, and from these experiments he concluded that the deciding factor in the inhibitory action of the salts on the parasite is not so much on the nature of the metallic ion as the presence of free hydroxyl ions in the soil. In the case of calcium cyanamide, Walker and Larsen ('34) stated that its toxicity is not due only to the basic substances formed from it but also to the CH_2 anions in the soil before hydrolysis is complete.

Basic Fertilizers

While the nature of the inhibitory effect of lime is not clearly known, it is nonetheless obvious from experimental work that lime makes the soil environment unfavorable for club root infection and development. Any fertilizer, therefore, which neutralizes this effect is to be avoided. The selection of a fertilizer to be added to the soil previously or sown with the seed determines to a great extent whether or not liming will be effective. Acid fertilizers in general and particularly superphosphates, basic superphosphates, superphosphates and carbonate of lime, turnip manures, etc., have been found to nullify the effects of lime and stimulate club root. The substitution of basic fertilizers and their use with lime is accordingly essential and has been widely advocated and practiced. A review of the literature shows, however, that the results have not always been strikingly beneficial or commercially satisfactory. In table 4 are listed the most commonly used of these fertilizers and their effect on club root.

In addition to the fertilizers listed in table 4 others have been used with varying success. *Calcium* and *potassium nitrate* give favorable results, according to Brezhnev ('34). Kindshoven ('24, '28) likewise secured good results from calcium nitrate when used at the rate of 50 gms. per sq. m. of soil. Sodium nitrate is effective when used in combination with lime, according to Murphy ('27).

Ammonium sulfate is ineffective as a fertilizer in combating club root, according to Whitehead ('25) and Osterwalder ('29).

Magnesium carbonate reduces infection in well-watered soil when used in sufficient amounts to raise the pH to 7.0 and usually inhibits the disease at pH 7.2 or above (Larsen and Walker, '34).

Gypsum stimulates the development of club root, according to Lindfors ('24), and in the opinion of Wellman ('30) is completely ineffective as a control.

"*Schlick*" or ore slime is a good fertilizer to be used against the disease, according to Hayunga ('12, '19), Appel and Schlumberger ('13), and Schaffnit and Lustner ('20).

TABLE 4. *Showing the effects of basic fertilizers on the control of club root.*

	Kainit (potash)	Basic slag	Calcium hydroxide and calcium cyanamide (Beka-Wurzelschutz)	Calcium hydroxide and waste (Herniol)
Arker ('35)	Favorable results	Favorable results	.....	.....
Blunck ('29)	.....	.....	Unfavorable results	.....
Deutelmoser ('26)	.....	.....	.....	Effective
Eriksson, '13	2,000 kg. per hectare—favorable	.....	.....	.....
Farsky, '26	Favorable results	Favorable results	.....	.....
Flachs and Kronberger ('30)	Favorable	.....	.....	.....
Gibbs ('32)	.....	Basic slag + lime—favorable results	.....	.....
Halsted, '95–'99	500, 1,000, 2,000 lbs. per acre—poor results	.....	.....	.....
Hayuna, '12, '19	Favorable results	.....	.....	.....
Hiltner, '08	Unfavorable results	.....	.....	.....
Hiltner, Korff, '16	.....	.....	.....	Favorable results
Kindshoven, '24, '28	.....	.....	.....	More or less unfavorable
Kreuzpointer, '23	.....	.....	Favorable results	.....
Magin, '02	.....	Good results	.....	.....
Miller, '23	.....	.....	Favorable results	.....
Motte, '33	Inconclusive results	.....	.....	.....
Murphy, '27	.....	Basic slag + lime—effective control	.....	.....
Naumann, '13	.....	8–12 kg. per acre + lime—good results	.....	.....
Osterwalder, '29	Unsatisfactory results	.....	Unfavorable results	.....
Schmidt, '22	.....	.....	.....	Favorable results
Vogel, '22	.....	.....	.....	More or less unfavorable
Wagner, '09	.....	8–18 kg. per acre + lime—good results	.....	.....
Whitehead, '25	.....	.....	Favorable results	.....

Haage's remedy has been found to be of little value by Appel and Schlumberger ('13) and Naumann ('13).

Superphosphate fertilizer stimulates club root development, according to Ravn ('10, '12), Osterwalder ('29) and Gibbs ('32), but McAlpine ('03) and Flachs and Kronberger ('30) reported favorable results from its use.

Jassen's remedy (calcium carbide dust and calcium cyanamide) is ineffective, according to Müller-Thurgau and Osterwalder ('23).

Saltpeter, superphosphate and *potash* as a combination fertilizer increases turnip yields, according to Ravn ('10), but also stimulates club root development.

Various kinds of ashes have also been tried as fertilizers in relation to club root, with varying success. Lime, peat, and briquett ashes are effective according to Ponkler ('96), Mathieu-Sanson ('97), Seelhoff ('12), K. M. ('19) and Straube ('22). Wood ashes were reported by N. N. ('93), Massee ('96) and Katterfeld ('23) to be effective against club root, but Halsted ('96, '99) and Schlumberger ('14) found them to be useless.

The beneficial effects of alkaline fertilizers as contrasted with acid ones on club root has been shown in experiments involving so-called complete fertilization. Kindshoven ('24) succeeded in reducing infection from 30 per cent to 2 per cent by application of an alkaline fertilizer consisting of calcium cyanamide, basic slag and 40 per cent potash at the rate of 50 gms. per sq. meter. Honig ('31) likewise got striking results in comparing the effects of alkaline and acid fertilizers on infection of kohlrabi in pots of heavily infested soil, as is shown below.

		Percentage of infection
Alkaline		
5.25 gms.	sodium nitrate	0– 20
10.50 "	basic slag	
13.11 "	calcium carbonate	
2.65 "	potash	
Acid		
4.2 gms.	ammonium sulfate	
8.94 "	superphosphate	90–100
17.81 "	gypsum	
5.25 "	Kainit	
	Controls	26– 31

Plants treated with the alkaline fertilizers showed 0 to 20 per cent infection, while those watered with acid fertilizers were 90 to 100 per cent infected.

Gibbs ('32) also found a marked difference in club root development when basic slag was compared with superphosphate in conjunction with lime, as is shown below:

Treatment	Percentage of Infection: Seeds drilled with basic slag	Seeds drilled with superphosphate
Control—no lime	59	95
3 tons commercial ground limestone per acre	22	53
3 tons superfine ground limestone per acre	10	87
2 tons air-slaked lime per acre	0	78
2 tons burnt lime per acre	0	36
2 tons water-slaked lime per acre	3	82

Kirschner ('27) has likewise advocated the use of complete basic fertilizer composed of basic slag and potassium nitrate in conjunction with calcium cyanamide to control club root in the field. In this connection may be noted Pryor's ('40) study on the effect of sulphur, nitrogen, and potassium nutrition on club root development in susceptible, resistant and immune strains of crucifers under controlled greenhouse conditions in Wisconsin. Varying nutrition has a pronounced effect on disease development in susceptible plants but does not influence resistance in immune varieties, according to Pryor. An abundance of potassium or nitrogen and a deficiency of sulphur or nitrogen increased the disease in susceptible plants. The percentage of infection was decreased markedly by a potassium deficiency. In the case of resistant plants club root was increased somewhat by a high supply of nitrogen; increased further by a deficiency of sulphur or nitrogen, and definitely decreased by lack of potassium.

Soil Drainage

Since club root is frequently most severe on low, wet and water-logged soils, proper soil drainage has often been advocated as an effective cure. Anderson ('55) and Ravn ('08) cited several instances where club root had been markedly checked by drainage, and Montieth ('24) and Naumov ('33) have demonstrated by controlled experiments that crucifers can be grown free of the disease in thoroughly infested soil by keeping the soil moisture down to 30 to 40 per cent of the water-holding capacity; all of which indicates the effect of excessive water in the development of club root. However, there is considerable evidence to show the maintenance of proper soil moisture by drainage is not in itself effective. Severe clubbing has often been found on high, well-drained soil and in fields which were carefully under-drained with tile. Furthermore, Wellman ('30) has shown that club root may occur generally in roots which are exposed to only 18 hours of excessive soil moisture. During the last two decades it has become increasingly obvious that other soil factors, relative acidity, humus content, etc., are involved and influence the efficacy of drainage as a curative measure. While soil drainage aerates and improves the physical condition of the soil, it cannot be relied upon alone as an inhibitor, but must be used in conjunction with other control measures to be effective.

Crop Rotation

Crop rotation is now generally recognized as essential in combination with other control measures against club root. Since the type of soil most favorable for intensive cultivation of crucifers is relatively limited, farmers and gardeners have a tendency to grow these crops on the same land for several successive years. If club root is present, such practice obviously leads to heavy infestation with fungus spores, and unless stringent control is exercised the land may become worthless for crucifers within a few years. The earlier students of club root, including Heinzelmann ('82), Eyeleshymer ('91), Laubert ('05a), Köck ('11), Burkhart ('15), and Ludwigs ('25) advocated only 2 to 3 years between successive crucifer crops, but since it has been shown that the resting spores of *P. Brassicae* may remain alive in the soil without hosts up to 7 and 8 years, it is obvious that a long rotation period is necessary for heavily contaminated fields. Jorstad ('23) recommended 5 to 6 years, Lindfors ('24) 4, Siemaszko ('25) 4 to 5, De Andres ('29) 3, Nielsen ('33) 6 to 8, Motte ('33) 7, Fedorintschik ('35) 4, Gibbs ('39) 6, and Stubbs ('41) 4 years or more between successive crops of crucifers. Short intervals are apparently ineffective if Fedorintschik's observations that soil not planted to crucifers for seven years contain enough viable spores to infect 26.6 per cent aseptically grown cabbage seedlings are correct. The practice of liming during rotation is of questionable value in light of Gibb's ('39) observation that the addition of lime and sulphur does not affect spore longevity. Crop rotation is further complicated by the fact that wild cruciferous hosts or weeds are also susceptible to club root and may keep the fungus alive during the rotation interval.

Various crops have been advocated as beneficial in rotation. Halsted ('99) reported a five-fold increase in turnips on land which had been planted to buckwheat the previous season, but these beneficial results were not evident the second year. Pettera ('17)

maintained that *Thysostegia virginica, Achillea pharmica, Astilbe* sp., and *Pyrethrum* have an inhibitory effect on club root and inactivates the spores within three years. Müller-Thurgau and Osterwalder ('23) also found that the spores remain viable only three years if beans are rotated with cabbage. According to Murphy ('27) turnips should be alternated with carrots. Blunck ('29) found that beans were particularly favorable as an alternate crop. Arker ('35) advocated rotation with beets, and Fedorintschik recommended rotation with grass and clover during the last two years of the interval to avoid plowing.

Eradication of Wild Hosts

Numerous cruciferous weeds are susceptible to club root, as Halsted ('92–'99), Ravn ('08), Cunningham ('12, '14), Ssacharoff ('16), Naumov ('26), Gibbs ('32), Rochlin ('33), Jamalainen ('36), and others have shown, and these hosts may harbor and perpetuate the disease in the absence of cultivated crucifers. Infected weeds have been found in grass pastures, wayside ditches, river beds, gardens, and cultivated fields (Halsted, '98; Gibbs, '32), and their presence on infected soil reduces the effectiveness of crop rotation in club root control. Even when only a few weeds are present in infected fields enough spores will be produced and perpetuated to infect subsequent cruciferous crops. Eradication of wild crucifers is therefore highly essential as a control measure and has been advocated and practiced to some extent as such, but in certain places it is not always practical. As Gibbs has pointed out, eradication is impractical in cereal grain crops, grass lands and pastures. In crop rotation on cultivated fields, eradication is obviously important, but unless it is combined with other control measures such as liming and growing resistant varieties of crucifers to keep down spore multiplication, its effect is limited.

Other special control measures involving winter ridging of the land and hilling up the soil around cabbage stalks have been practiced without consistent success. In the autumn of 1898 Halsted plowed infected plots deeply and piled the soil up in long 2 ft. high ridges to expose the spores to the maximum weathering during the following winter months. Less clubbing was present on the ridged land (31 per cent) the following season than on the level plots (38 per cent), but the small difference does not justify ridging as a satisfactory remedy for club root, according to Halsted. He also tested the effect of shading on the disease in turnips and found that it does not have an inhibitory effect. Hilling up the soil around cabbage stalks leads to increase of adventitious roots on the stalk above the infected portion, according to Cunningham ('14). Such adventitious roots are comparatively free of clubbing, and since they occur above the diseased and useless main root the nutriments which they absorb are readily available to the developing heads. Cunningham found that hilling increased the yield ten-fold in some plots during 1912, but in the following year no beneficial results were attained.

Resistant Varieties

Cultivated and wild crucifers vary in degree of susceptibility to *P. Brassicae,* and several cultivated strains and varieties have been developed which are fairly resistant to club root. A certain measure of control may accordingly be achieved by the cultivation of these varieties. Particularly promising are the results obtained by Olsson ('39, '40) in breeding resistant varieties of swedes and turnips in Sweden. The data on relative degree of resistance, however, are often conflicting, and in certain varieties where some investigators have reported complete immunity, others have found 100 per cent susceptibility. These differences in results are doubtless due in part to variations in experimental conditions and methods employed. As has been shown elsewhere, soil types and moisture, H-ion concentration, number of spores in the soil, etc., are important factors in infection, and unless they are kept constant in experimental work, it is difficult to determine the inherent degree of susceptibility or resistance of a particular variety or strain. Doubtless many of the reported cases of immunity relate to plants which have escaped infection in the field. The literature relating to varietal susceptibility is nonetheless very extensive, and in a brief treatise of this nature no attempt will be made to enumerate and discuss all the data relative to this subject.

The range of susceptibility in turnips is very great and some varieties are reported to vary from 100 per cent susceptibility to almost complete resistance. No varieties, however, have been developed or found which are consistently immune. Southern Curley Top, Rutaraga, and Large Flat Green were reported by Cunningham ('14) to be particularly susceptible. In the first named variety clubbing was so extensive that the turnip root was converted into a system of branched hypertrophied rootlets. On the other hand, the following commercial strains have been reported to be relatively resistant:

Bruce
Bruce Purple Top Yellow
Bruce Wallace
Dale's Hybrid
Early Snowball
Early White Milan
Golden Ball
Green Top
Hinkenborstel
Irvine's Green Top Yellow
May
New Bronze Top
Ostersundom
Pomeranian Tankard
Purple Top Milan
Purple Yellow Top
Purple Yellow Top Aberdeen
Rutabaga
Scarlet Kashmyr
Seefeld
Snowball
Svalöv's Yellow Tankard
Victor
Weibull's Immune
Weibull's Sekel
White
White Fleshed May
White Milan
Yellow Aberdeen
Yellow Bruce

by Halsted ('99), Ravn ('11), Cunningham ('14), Anony., (Nova Scotia, '23), Lindfors ('24, '25), Tennent ('25, '30, '34), Gibbs ('31), Findlay ('31), MacLeod ('31), Hendrick ('32), Beaumont and Staniland ('33, '34), Walker and Larsen ('34), Walker ('36), Olsson ('39, '40), Brezhnev ('39) and Pryor ('40). EARLY WHITE MILAN and EARLY SNOWBALL showed only 1.1 per cent to 0.6 per cent susceptibility, according to Cunningham.

Swedes in general are reported to be more resistant than turnips, but they likewise exhibit a wide range of susceptibility and resistance. The following varieties:

Balmoral	Ostergöta
Bangholm	Otofte
Bangholm Herning	Sweet German
Bangholm Studsgaard	Sweet Russian
Danish Varieties 4 and 25	White Necklace
	White Russian
Green Top Swedish	White Swede
Majrova	Wilhemsburger
May	

have been reported by Ravn ('11), Cunningham ('14), Anony., (Nova Scotia, '23), Linfors ('24, '25), Whitehead ('22, '25), Hockey ('26), Güssow ('26), Tennent ('25, '30, '34), Davis, Griffith, and Evans ('28), Osterwalder ('29), Gibbs ('31), Findlay ('31), MacLeod ('31), Beaumont and Staniland ('33, '34), Walker and Larsen ('34), Olsson ('39, '40), Bennett ('39), and Pope ('39) to be relatively resistant. Ravn ('11), Lindfors, Güssow, and Pope, however, found that the so-called resistant BANGHOLM PURPLE TOP, STUDSGAARD BANGHOLM, WILHELMSBURGER, and YELLOW TANKARD varieties may be 100 per cent infected and completely destroyed.

Cabbages, likewise, show a wide range of susceptibility to club root, and none of the commercial varieties are highly or completely resistant, according to Cunningham, Lindfors, Walker and Larsen, Jamalainen, and others.

All Seasons	Dark Red Erfurt
Amager	Hvidkaal
American Savoy	Mammoth Red Rock
Blomkaal	Perfection Savoy
Braunsweig Gribkova	Rodkaal
Braunsweig Hos Hos	Volga
Brunswick	White Russian
Copenhagen	

have been reported by Ravn ('08), Cunningham ('12, '14), Naumov ('25, '28), Rochlin ('33), and Fedorintschik ('35) to be particularly susceptible. All of these varieties may show 98 per cent to 100 per cent clubbing in badly infested soil. On the other hand,

Blue	Large Late Flat Dutch
Bodenkohlrabi	Late Moscow
Bronka	Red
Grönkaal	Short Stemmed Amager
Henderson's Early Summer	Slovianka
	Stone Maxon
Hollander	Valvatievka

are said to be less susceptible (Cunningham, '14; Höstermann, '22; Harter and Jones, '24; Osterwalder, '29; Tedin, '33; Motte, '33; Fedorintschik, '36; Brezhnev '39).

Radishes are also very susceptible to club root, and it is doubtful whether any completely immune commercial varieties exist. Halsted ('99) reported GIANT STUTTGART, LONG BLACK SPANISH, NEWCOMB WHITE, and YELLOW SUMMER TURNIP to be wholly free from clubbing. Cunningham found that susceptibility varied from 92.2 per cent in LONG SCARLET RADISH to 5.6 per cent in GIANT STUTTGART. In addition to the latter variety, EARLY SCARLET TURNIP, DELIKATESS, DREIENBRUNNEN, IMMUNE, LONG BLACK PARIS WINTER, RUBIN, and SAXA have been reported by Cunningham ('14), Gleisberg ('23) and Jamalainen ('36) to be fairly resistant.

Other commercial cultivated crucifers have not been so extensively studied for varietal resistance as those noted above. Cunningham and Jamalainen found all varieties of kohlrabi to be very susceptible, but Schaffnit reported that the varieties which he studied were relatively immune. Honig ('32) tested five varieties of kohlrabi and found the following incidence of infection: GELBE SCHMALZ 7.3 per cent, WEISSE SCHMALZ 27.7 per cent, WEISSE WESTER 0 per cent, GELBE WESTER 32.6 per cent, APFEL GELB 6.2 per cent. Among Brussel sprouts, HERCULES is fairly resistant (Jamalainen, '36). All varieties of cauliflower are equally susceptible, according to Cunningham, Lindfors, and Jamalainen. MARROW STEM and DREINENBRUNNEN CURLEY KALE are said to be highly resistant by Osterwalder, and Beaumont and Stanland, while APRIL QUEEN and VICTORY broccoli were found to be resistant by Bailie and Muskett. Rape shows almost 50 per cent susceptibility, according to Cunningham, but Gleisberg reported it to be immune to club root. Ssacharoff and Rochlin also reported *B. Napus* var. *S. esculenta* to be immune. One variety of *B. Rapa* showed 100 per cent susceptibility, while another exhibited only 10.9 per cent clubbing, according to Cunningham. Gleisberg, however, reported *B. rapa* to be unsusceptible. Mustard is reported to be highly susceptible by Höstermann, while Cunningham, Naumov, Motte and Rochlin found black mustard to be highly resistant or completely immune. Chinese cabbage shows 100 per cent susceptibility, according to Naumov and Katterfeld.

Nature of Susceptibility and Resistance.—The differences in degree of infection exhibited by the wild and cultivated crucifer varieties listed above were believed by some workers to be partly due to the presence of more or less virulent biological strains of *P. Brassicae* which are specific for certain hosts. Considerable doubt has been expressed about the presence of such strains and it is rather generally believed that relative susceptibility and resistance are largely inherent host characters. The nature of resistance is not yet well understood, but Ssacharoff believed it to be due to substances in the cell sap.

He found that resistance was correlated with a low sugar content in the cell sap and a pungent, bitter taste of the expressed juice, while the cell sap of susceptible plants was comparatively rich in sugar content. Whitehead ('25), however, asserted that the factor determining resistance is not related to total dry matter or sugar in the roots. Further observations and experiments on the nature of resistance were made by Rochlin in 1933, who tested 47 wild and cultivated species belonging in 14 genera of the Cruciferae for their susceptibility to *P. Brassicae*. He found that the reaction varied from complete immunity in some species to susceptibility in others, independently of their taxonomic position, as Cunningham had previously shown. All gradations of susceptibility occurred in one and the same genus. Rochlin also made a comparative anatomical study of the roots of numerous species and found that in the early stages of growth immunity or susceptibility is not correlated with any marked differences in root structure. In adult plants, however, the penetration and spread of *P. Brassicae* is hindered to some degree by the development of cork layers, collenchyma, and by the compact structure of the wood layers.

The degree of resistance exhibited by a species or variety is directly correlated with the amount it contains of those glucosides which on fermentation with myrosin produce highly pungent mustard oils, according to Rochlin. Chief among such glucosides in crucifers are *sinigrin* (particularly abundant in *B. nigra* and horseradish and in smaller amounts in *Sinapsis juncea, B. rapa, B. Napus,* etc.), *gluconasturtiin* (in *Barbarea praecox* and *Nasturtium officinale*), *glucotrapaeolin* (in *Lepidium sativum*), etc.), and *glucocochlearin* (in *Cochleari officinalis*). *Sinablin,* a glucoside present in *B. alba,* which does not yield a pungent mustard oil, was found to be of no protection against infection with *P. Brassicae*.

An indication of the possible use of active glucosides or their derivatives as fungicides is shown by the results obtained by Rochlin in a small experimental plot in which seeds of the very susceptible Brunswick cabbage were sown in highly infected soil in pots, some of which were abundantly watered with a water extract from *B. nigra* seeds. Only 20 per cent of the seedlings became infected and showed a very slight swelling of the roots, while all the control seedlings were infected. Considerable doubt has been thrown on Rochlin's theory of the nature of resistance by the subsequent studies of Walker ('36), Walker, Link, and Marcell ('36). These workers found that some collections of *B. nigra* are very susceptible and that there is no correlation between mustard content and resistance.

From the practical standpoint, Rochlin suggested the possibility of controlling club root by crossing cruciferous species deficient or meager in active glucosides with those which contain greater amounts of these substances. Pryor investigated this possibility by direct experiments involving variation of the mustard oil content of crucifers and noting their susceptibility to the disease. All mustard oils in crucifers contain sulphur and nitrogen, while their glucosides also contain potassium. Thus, by lowering or increasing these nutrient elements, it is possible to change the mustard or sulphur oil content of experimental plants. From the results obtained by this procedure, Pryor concluded that sulphur oils do not inhibit or prevent infection and development of club root in crucifers—thus refuting the observations of Rochlin.[1]

Geographical Distribution of Club Root and Bibliography of Literature

Club root is now world wide in distribution, and the countries from which it has been reported up to the present time are listed below. The number of publications on the occurrence, distribution, hosts, life-history, cytology, relationships, eradication and control of *P. Brassicae* and club root is quite large and many of them are to be found in local journals which are not readily available. In the bibliography which follows many such publications have doubtless been overlooked and omitted.

ALASKA

Alberts, H. W. 1930. Rept. Alaska Agric. Exp. Sta. 1930: 6.

Georgeson. 1914. *Ibid.* 1914: 27. 1915, *Ibid.* 1915: 39. 1917, *Ibid.* 1917: 8. 1919, *Ibid.* 1919: 21. 1927, *Ibid.* 1927: 10.

ARGENTINA

Marchionatto, J. B. 1929. Phys. Rev. Soc. Argentina de Cien. Nat. 9: 455.

AUSTRALIA

Anony. 1940. Agr. Gaz. New South Wales 51: 559.

Darnell-Smith, G. P. 1924. Agric. Gaz. New S. Wales 35: 180, 488.

McAlpine, M. D. 1898. Proc. Linn. Soc. N. S. Wales 1898: 82.

———. 1901. Dept. Agric. Victoria 1901.

———. 1903. Dept. Agric. Victoria 1903.

Noble, R. J. Intern. Bull. Pl. Protect. 5: 202.

Stubbs, L. L. 1941. Jour. Dept. Agric. Victoria 39: 208.

AUSTRIA

Anonymous. 1933. Bundesant. f. Pflanzensch. Mitteil. 167.

Köck, G. 1911. Landesamtbl. Erzherogtums Österr. a. d. Enns 1911, no. 1.

Kornauth, K. 1913. Zeitschr. Landw. Versuchsw. Österr. 17: 395. 1915, *Ibid.* 19: 180.

[1] In a paper presented before the December 29, 1941, meeting of the American Phytopathological Society, Dallas, Texas, W. J. Hooker (see Phytopathology 32: 9) reported that two mustard oils (allyl isothiocyanate and beta phenyl ethyl isothiocyanate) were consistently effective in preventing spore germination at 80 ppm. and sometimes at as low concentrations as 10 ppm. of allyl isothiocyanate and 5 ppm. of beta phenyl ethyl isothiocyanate. Concentrations of both oils below the toxic level were found to be capable of stimulating spore germination.

Piehler, F. 1919. Wien. Landw. Zeit. 1919: 383.
Stift, A. 1905. Österr. Ungarische Zeitschr. Zucker u. Landw. 1905: 9.

BELGIUM

Terby, J. 1923. Mém. Roy. Acad. Belg. 7: 1–28.
———. 1924a. Bull. Roy. Bot. Soc. Belgium 56: 48.
———. 1924b. Bull. Roy. Acad. Belg. 5 ser. 10: 519.
———. 1932. Mém. Roy. Acad. Belg. 11: 120.
Vanderyst, H. 1904. Bull. Agric. 20: 533.

BULGARIA

Nicoloff, I., and M. Stefanova. 1922. Zentralbl. f. Agrikultchemie 51: 101–102.

CANADA

Anonymous. 1923. Rept. Superint. Kentville, N. S. Exp. Sta. 1922. Canad. Dept. Agric. Domin. Exp. Farm.
Clark, J. 1924. Rept. Superint. Charlottetown. P. E. I. Exp. Sta. 1923. Canad. Dept. Agric. Domin. Exp. Farm.
Güssow, H. T. 1925. Rept. Domin. Botanist for 1924. Div. Bot. Canad. Dept. Agric. pp. 27–28.
Hockey, J. F. 1926. Rept. Domin. Botanist for 1925. Div. Bot., Canad. Dept. Agric. pp. 29. 1927, *Ibid.* 1927: 28. 1928, *Ibid.* 1928: 139.
Ledingham, G. A. 1934. Nature 133: 531.
McRostie, G. P. 1936. Rept. Canad. Seed Grow. Assn. 1935–'36: 31.
McLarty, H. R. 1929. Rept. Domin. Botanist for 1928. Div. Bot., Canad. Dept. Agric. p. 142.
MacLeod, D. J. 1931a. Rept. Domin. Botanist for 1930. Div. Bot. Canad. Dept. Agric. p. 25. 1931b, *Ibid.* p. 181.

CEYLON

Petch, T. 1906. Trop. Agric. 25: 839.

CZECHOSLOVAKIA

Bandys, E. 1914. Zeitschr. Pflanzenkr. 24: 342.
Bubak, F. 1902. Zeitschr. Landw. Versuchsw. Österr. 5: 675. 1904, *Ibid.* 7: 731.
Farsky, O. 1926. Ochr. Rost. 6: 114.
Köhne. 1928. Landw. Fachpresse Tschechoslow. 6: 204.
Milovidov, P. F. 1931. Arch. Protistk. 73: 1. 1933. *Ibid.* 81: 138.
Nemec, B. 1913. Sekarske Rozhledy. Abt. f. Immunität u. Seriol. 1913: 181.
Schmidt. 1922. Land. u. Forstw. Mitt. Böhmen 1922: 21.
Sitensky, F. 1896. Vestnik. kräl. Ceske Spolecenosti nauk Trida Matem. Proiodov. 1896: 8–20.
Svec, F. 1923. Ochr. Rost. 3: 18–19.
Uzel, H. 1914. Wiener Landw. Zeit. 1904: 917.
———. 1907. Zeitschr. Pflanzkr. 17: 85.
———. 1908. Zeitschr. f. Zuckerind in Böhmen 32: 622. 1910. *Ibid.* 34: 319.

DENMARK

Anonymous. 1922. Stat. Forsoksvirk. Plantek. Medd. 95: 1.
Christensen, O. 1903. Vort Landbrug. 22: 157.
Christensen, H. R., P. Harder, and F. K. Ravn. 1909. Tidsskr. Landbr. Planteavl. 16: 430.
———. 1911. Zeitschr. Pflanzenkr. 21: 124.
Ferdinandsen, C. 1923. Tidsskr. f. Landkon. 1923: 256.
———, and S. Rostrup. 1921. Tidsskr. Landbr. Planteavl. 27: 697.
Gram, E., and S. Rostrup. 1922. Tidsskr. Landbr. Planteavl. 24: 236.
———, C. A. Jorgensen, and S. Rostrup. 1928. *Ibid.* 34: 778.
Hennings, P. 1895. Verh. Bot. Verein Prov. Brandenburg 37: LVIII. 1896, *Ibid.* 38: 58..
———. 1922. Kgl. Landtbr. Akad. Handl. Tidsskr. 1922: 26.
Jorgensen, C. A. 1922. Tidsskr. Landbr. Plant. 39: 316.
Mortensen, M. L., S. Rostrup, and F. K. Ravn. 1908. Tidsskr. Landbr. Plant. 15: 153. 1910, *Ibid.* 17: 306. 1911, *Ibid.* 18: 317.
Motte, M. H. 1933. Jour. d'Agric. Prat. 97: 177. 1935. *Ibid.* 99: 93.
Nielsen, N. J. 1933. Tidsskr. Landbr. Plant. 39: 361.
———, and C. J. Christensen. 1914. Tidsskr. Landlingetspl. 21: 87.
Ravn, F. K. 1905a. Dansk. Landbrug. 1: 39.
———. 1905b. Gartner-Tidende 1905: 109.
———. 1905c. Beret. Foren. Jyd. Landbofor. 31: 89. 1906, *Ibid.* 32: 86. 1907, *Ibid.* 33: 166.
———. 1907a. Smaaskr. udg. of Dansk Landbr. 1907.
———. 1907b. Beret. om lokale Markforsoy i Jylland 1906: 85.
———. 1908. Tidsskr. Landbr. Plant. 15: 527.
———. 1909. Zeitschr. Pflanzenkr. 19: 473. 1910a, *Ibid.* 20: 15.
———. 1910b. Tidsskr. Landbr. Plant 17: 163. 1911a. *Ibid.* 18: 357.
———. 1911b. Biol. Arb. 1. Eng. Warmung 1911: 167.
———. 1913. Zeitschr. Pflanzenkr. 23: 140. 1917, *Ibid.* 27: 144.
———, and A. Madsen-Mygdal. 1906. Udg. of de Samvirk Landbofor. i Fyns Stift, p. 14.
———, and A. Madsden-Mygdal. 1909. Zeitschr. Pflanzenkr. 19: 473.
Rostrup, E. 1871. Landmans-Blad 1871: 57, 71.
———. 1884. Meddel. Bot. Foren. Kobenhavn 1: 119.
———. 1891. Tidsskr. Landokon. 3 ser. 10: 498. 1892, *Ibid.* 11: 326. 1893a, *Ibid.* 12: 625.
———. 1893b. Zeitschr. Pflanzenkr. 3: 146.
———. 1894a. Tidsskr. Landbr. Plant. 1: 131.
———. 1894b. Zeitschr. Pflanzenkr. 4: 285.
———. 1894c. Bot. Tidsskr. 19: 201.
———. 1896. Tidsskr. Landbr. Plant. 3: 123.

———. 1897. Zeitschr. Pflanzenkr. 7: 158. 1898, *Ibid.* 8: 278.
———. 1899. Tidsskr. Landbr. Plant. 6. 1900, *Ibid.* 7: 13–32. 1901, *Ibid.* 8: 109. 1902, *Ibid.* 9: 115. 1903, *Ibid.* 10: 361. 1904, *Ibid.* 11: 395. 1905, *Ibid.* 12: 352. 1906, *Ibid.* 13: 79.

ENGLAND

Adam, E. 1789. Practical essays on agriculture, vol. 2.
Anonymous. 1868. Country Gentleman's Mag. 1: 40.
———. 1911. Gard. Chron. 3 ser. 49: 150.
———. 1912. Worcester County Exp. Gard., Droitwich. Ann. Rept. 1912.
———. 1933. Bull. Min. Agric. Fish. 68. 1934, *Ibid.* Bull. 79.
Beaumont, A., and L. N. Staniland. 1933. Ninth Ann. Rept. Seale-Hayne Agric. Coll., Newton Abbot, Devon. 1932. 1934, *Ibid.* 1933.
Bennett, F. T. 1939. Ann. Appl. Biol. 26: 837.
Berkeley, M. J. 1856. Gard. Chron. 1856: 500.
Biffen, R. 1927. Jour. Roy. Agric. Soc. England 187: 346.
Brown, W. 1935. Jour. Pom. 13: 247. 1937, *Ibid.* 15: 69.
Buckman, 1854. Jour. Roy. Agric. Soc. England 15: 125.
Carieklee, H. T. W. 1903. Gard. Chron. 3 ser. 34: 163.
Carruthers, N. 1893. Jour. Roy. Agric. Soc. England 3 ser. 4: 334.
Collinge, W. E. 1911a. Jour. Land Agent's Soc. 10: 1–4.
———. 1911b. Gard. Chron. 50: 150.
Cooke, M. C. 1903. Jour. Roy. Hort. Soc. 27: 801.
———. 1906. Fungoid pests of cultivated plants. London.
Cook, W. R. I. 1933. Arch. Protistk. 80: 179.
———, and E. J. Schwartz. 1930. Phil. Trans. Roy. Soc. London B 218: 283.
Curtis, J. 1843. Jour. Roy. Agric. Soc. London 4: 100.
Ellis, W. 1742–'44. The modern husbandman, or the practice of farming. 4 vols. London.
Gilchrist, D. A. 1898. Ann. Rept. Distr. of Grants Agric. Education. 1897/98: 89; 1898/99: 110; 1899/1900: 96.
———. 1905. County of Northumberland Bull. No. 3, New Castle.
———. 1913. County Northumberland Educ. Comm. Bull. 19: 86.
Hall, A. D. 1904. The soil. London. 2nd ed. 1910. 3rd ed. 1920.
Holmes-Smith, E. 1930. Gard. Chron. 89: 371. 1931, *Ibid.* 90: 35.
Marshall. 1795. Rural economy of Norfolk. 2nd ed. London.
Massee, G. 1895. Proc. Roy. Soc. 57: 330.
———. 1896. Rev. Mycol. 1896: 22.
Milburn, T. 1853–'55. Jour. Agric. 1853–'55: 73.
Ogilvie, L., and B. O. Mulligan. 1934. Fifth Ann. Rept. Agric. and Hort. Res. Sta. Long Ashton, Bristol for 1933: 98.
Potter, M. C. 1896a. Gard. Chron. 19: 332.
———. 1896b. Jour. Newcastle Farm Club. 1896.
———. 1896/97. Nature 55: 33.
Potts, G. 1935. Trans. Brit. Mycol. Soc. 19: 114.
Preston, N. C. 1926. Rept. Advis. Dept. Harper Adams Agric. Coll. Newport, Salop 1: 9. 1927, *Ibid.* 2: 2. 1929, *Ibid.* 4: 4. 1930, *Ibid.* 5: 5.
———. 1934. Jour. Min. Agric. 41: 329.
Preston, W. P. R. 1903. Gard. Chron. 3 ser. 4: 293.
Russel. 1857/59. Jour. Agric. n. s. 1857/59: 529.
Shewell-Cooper, W. E. 1932a. Gard. Chron. 91: 387. 1932b, *Ibid.* 92: 83.
Smieton, M. J. 1939. Jour. Pomol. 17: 195. 1931, *Ibid.* 90: 35.
Smith, W. G. S. 1883. *Ibid.* 20: 625.
Smith, E. H. 1930. *Ibid.* 87: 371.
Sommerville. 1894. Jour. Roy. Agric. Soc. England 3 ser. 5: 808. 1895, *Ibid.* 6: 749.
———. 1897. Ann. Rept. Distr. Grants Agric. Education. 1896/97: 39.
Stephens. 1828/29. Quart. Jour. Agric. 1: 429.
Voelcker, A. 1854. Jour. Roy. Agric. Soc. England 1 ser. 20: 101.
Voelcker, J. A. 1894. *Ibid.* 3 ser. 5: 318. 1898. *Ibid.* 3 ser. 8: 650.
Wakefield, E. M., and W. C. Moore. 1896. Trans. Brit. Mycol. Soc. 20: 97.
Woodman, R. M., G. H. Brenchley, and F. Hanley. 1934. Jour. Soc. Chem. Ind. 53, 4: 35.

FINLAND

Anonymous. 1925. Nachrichtenbl. Deut. Pflanzenschutzel. 5: 106.
Jamalainen, E. A. 1936. Valt. Naatal. Julk. 85: 1–36.
Karsten, P. A. 1884. Soc. Fauna et Flora Fennica 10.
Rainio, A. J. 1930a. Valt. Naatal. Julk. 23: 275. 1930b, *Ibid.* 23: 306.

FRANCE AND COLONIAL AFRICA

Dufrenoy, J. 1923. Rev. Bot. Appl. 33: 241.
Foex, E., and E. Marchal. 1931. Ann. Epiph. 17: 1–61.
Gay, A. 1913. Jour. d'Agric. Prat. 77: 816.
G. M. 1920. La Terre Vaud. 1920: 192.
Griffon and Maublanc. 1912. Bull. Soc. Mycol. France 26: 469.
Mathieu-Sanson. 1897. Rev. Hort. 69: 394.
Maire, R. 1917. Bull. Soc. Hist. Nat. l'Afrique du Nord. 8: 134.
———, and A. Tison. 1909a. C. R. Acad. Sci. Paris 150: 1768.
———, and A. Tison. 1909b. Ann. Mycol. 7: 226.
Mangin, L. 1902. Rev. Hort. 74: 432.
Marchand, E. F. L. 1910. C. R. Acad. Sci. Paris 150: 1348.
Noel, P. 1902. Natur. 1902, no. 374: 226.

Oger, A. 1897. Rev. Hort. 69: 213.
Passy, P. 1914. Jour. d'Agric. Prat. 78: 87.
Pinoy, E. 1905. C. R. Soc. Biol. 58: 1010.
———. 1907. Ann. Inst. Pasteur 21: 686.
Prillieux, E. 1895. Maladies des Plantes Agricoles 1. Paris.
Renard, P. 1935. Vie Agric. rur. 24: 167.
Seltensperger. 1896. Rev. Mycol. France 1896: 23.
Vanderyst, H. 1901. Bull. l'Agric. 1901: 535.
Vincent, V. H., J. Herviaux, and Coic. 1936. C. R. Acad. Agric. France 22: 851. 1938. *Ibid.* 24: 83.
Vercier, J. 1930. Rev. Horti. Paris 102: 86.
Weschieder. 1898. *Ibid.* 70: 566.

GERMANY

Anonymous. 1914. Gartenwelt 18: 96.
———. 1922a. Prov. Sachs. Monatschr. Obst.-, Wein. -u. Gartenb. 33: 34. 1922b. *Ibid.* 33: 50.
———. 1922c. Prakt. Ratg. Obst. -u. Gartenb. 37: 87.
———. 1922d. Handelsbl. Deut. Gartenb. 37: 180.
———. 1923a. Landw. Wochenschr. Westfalen u. Lippe 80: 130.
———. 1923b. Deut. Obst. -u. Gemusebau Zeit. 69: 99.
———. 1923c. Deut. Erwerbsg. 1: 92.
———. 1923d. Prakt. Ratg. 1923.
———. 1923e. Wegweis. Obst. -u. Gartenb. 31: 67.
———. 1923f. Prakt. Obst. -u. Gartenb. 15. no. 3.
———. 1926. Landw. Jahrb. 64. suppl. 11: 67. 1931. *Ibid.* 74. suppl. 1: 1–110.
———. 1932. Mitt. Biol. Reichanst. f. Land. -u. Forstw. 43.
———. 1939. Landw. Jahrb. 87: 567.
Appel, O. 1914. Mitt. d. Mährens Landw. Landesvers. Brüm 1914: 39.
———. 1928. Handbuch der Pflanzenkr. 2. Berlin.
———, and E. Werth. 1910. Mitt. Kgl. Biol. Anst. Land. -u. Forstw. 10: 176.
———, and O. Schlumberger. 1913. *Ibid.* 14: 18. 1914. *Ibid.* 15: 13.
Arker, H. 1935. Nachr. Schäd. Bekampf. Leverkusen 10: 162.
Aue, W. 1913. Erfurt Führer Obst. -u. Gartenb. 13: 315.
Bayer, F. 1922. *Ibid.* 22: 115.
Becker, J. 1912. Schleswig-Holstein Zeitschr. Obst. -u. Gartenb. 1912: 3.
———. 1924. Handbuch der Gemüsebau. Berlin.
———. 1935. Deut. Landw. Presse 64: 588.
Behla, R. 1898. Centralbl. Bakt. Parasitk. 24: 829.
———. 1899. Zeitschr. Hyg. 32: 133.
———. 1903. Die Pflanzenparasitäre Ursache des Krebses und die Krebsprophylaxe. Berlin.
Beyer, T. 1925. Erf. im Obst. -u. Gartenb. 1925: 96.
Blumberg. 1922. Kleintierz. u. Gartenb. 17: 100.
Blunck, H. 1929. Gartenbauw. 1: 154.
Böhner, K. 1922. Fränkischer Kurier, Nürnberg 8. no. 111.
Bottner. 1916. Prakt. Ratg. Obst. -u. Gartenb. 32: 50.
Bremer, H. 1923. Landw. Jahrb. 59: 227.
———. 1924a. Nachrichtenbl. Deut. Pflanzenschutzd. 4: 16. 1924b. *Ibid.* 4: 73.
———. 1924c. Landw. Jahrb. 59: 673.
———. 1928. Erfurt Führer Obst. -u. Gartenb. 28: 341.
Brick, C. 1912. Jahrb. Hamburg. Wiss. Anst. 30.
———. 1913. Stat. f. Pflanzensch. z. Hamburg 15: 1.
Bromme, H. 1926. Obst. -u. Gartenb. 72: 358.
———. 1928. Raiffeisenbote, Braunschweig 1928: 103.
Bruck, C. 1912. Jahrb. Hamburg. Wiss. Anst. 30.
———. 1913. Stat. Pflanzensch. z. Hamburg 15: 1.
Bruck, W. F. 1907. Sammlung Göschen. Leipzig.
Burkhardt, F. 1915. Flugbl. no. 19. Abt. Pflanzenkr. Kais. Wilh. Inst. Landw. Bromberg.
Caspary, R. 1873. Schr. Physik.-Ockon. Gesell. Königsberg 1873: 109.
———. 1877. Gard. Chron. 3: 118.
Dankler. 1919. Der Gartenfreund 1919: 100.
Deutelmoser, E. 1926. Obst. u. Gemüsebau 72: 291.
Eggemeyer. 1920. Prakt. Ratg. Obst. -u. Gartenb. 35: 264.
Esmarch, F. 1924. Die Kranke Pflanze 1: 169. 1925, *Ibid.* 2: 207.
Feinberg, L. 1901. Ber. Deut. Bot. Ges. 19: 533.
———. 1902a. Dent. Med. Wochenschr. 28: 43.
———. 1902b. Berlin Klin. Wochenschr. 39: 572.
———. 1902c. Das Gewebe und die Ursache der Krebsgeschwulste. Berlin.
Felsberg, L. 1916. Prakt. Ratg. Obst. -u. Gartenb. 31: 172.
Flachs, F. 1930. Prakt. Bl. Pflanzenbau u. Pflanzensch. 8: 250.
———, and M. Kronberger. 1930. *Ibid.* 8: 74–80.
Fleishman, C. 1927. Gartenwelt 31: 781.
Frank, A. B. 1896. Die Pilzparasitären Krankheiten der Pflanze 2: 14.
Fruwirth. 1918. Handb. Landwirtsch. Pflanzensuch. 2.
———. 1920. Algemeine Zuchtungslehre der Landwirtsschaftlichen Kulturpflanzen. 5 ed. Berlin.
F. S. 1920. Deut. Gartenbauzeitg. 22: 66.
Gleisberg, W. 1920. Deut. Landw. Presse 1920: 705.
———. 1922a. Nachrbl. Deut. Pflanzenschd. 2: 89. 1922b, *Ibid.* 3: 10. 1923b, *Ibid.* 4: 10.
———. 1926. Obst. -u. Gemüsebau 72: 28.
Gretschel, F. 1916. Prakt. Ratg. Obst. -u. Gartenb. 31: 172.
Gross-Schlachters. 1916. *Ibid.* 31: 198.
Habernall. 1919. Der Gartenbau 12: 6.
Hayunga, J. 1909. Mitt. Deut. Landw. Gesell. 1909: 677.
———. 1911. Prakt. Ratg. Obst. -u. Gartenbau 11: 100.
———. 1912. Der Handelsgärt. 14: 173.
———. 1919. Mitt. Deut. Landw. Gesell. 1919: 52.
Hellmann, A. 1926. Erfurt Führer Obst. -u. Gartenb. 27: 94.
Herpers, H. 1913. Gartenwelt 17: 674.
———. 1923. Prakt. Ratg. Obst. -u. Gartenb., Beil. Prakt. Landw. no. 2.

———. 1924. Erfurt Führer Obst. -u. Gartenb. 25: 49. 1925a, *Ibid.* 50: 39.
———. 1925b. Gartenwelt 29: 706.
Hertel, F. 1926. Obst. -u. Gemüsebau. 72: 67.
Heyder. 1911. Oldenburgisch. Landw.-Blatt 1911: 65.
Hiltner, L. 1908. Ber. Tätigkeit Kgl. Agric. -Bot. Anst. München in Jahre 1907. 1908: 98–99.
———, and G. Korff. 1916. Prakt. Bl. Pflanzenb. u. Pflanzensch. 1916, h. 3: 25.
Hofferichter, K. 1926. Gartenflora 75: 262.
Hoffman, W. 1932. Ratschl. f. Haus, Garten, Feld. 7: 162.
Hollenbach, O. 1911. Gartenwelt 15: 8.
Hollrung. 1923. Die Mittel zur Bekämpfung der Pflanzenkrankheiten. Berlin.
Honig, F. 1931. Gartenbauwiss 5: 116.
———. 1932. Nachr. über Schädlingsbekämpf. 7: 22.
Honigmann. 1926. Pflanzenbau, Halbmschr. f. Saatwesen Anbau u Pflege d. Kulturpflanz. 2: 290.
Höstermann, G. 1909. Ber. Kgl. Gartenlehranst. Dahlem b. Steglitz 1908/9: 124. 1921, *Ibid.* 1920/21: 100.
———, and M. Noack. 1923. Lehrbuch der Pilzparasitären Pflanzenkrankheiten. Berlin.
Hsf. 1914. Mitt. Deut. Landw. Gesell. 29: 12.
Hurrle, A. 1916. Prakt. Ratg. Obst. -u. Gartenbau 31: 69.
Kalchschmid, W. 1941. Deut. Landw. Presse 68: 185.
Kappen. 1922. Mitt. Deut. Landw. Gesell. 37: 660.
Kellerman. 1903. Prakt. Bl. Pflanzenb. u. Pflanzensch. 9: 103.
Kimppel, K. H. 1922a. Prov. Sächs. Monatschr. Obst. -Wein-Gartenb. 23: 78. 1922b, *Ibid.* 23: 172.
Kindshoven, J. 1924. Mitt. Deut. Landw. Gesell. 39: 259. 1928, *Ibid.* 43: 522.
Kirschner, L. 1906. Die Krankheiten und Beschädigungen unserer landwirtschaftlichen Kulturpflanzen p. 371. Stuttgart.
———. 1927. Atlas d. Krankh. u. Beschäd. Landw. Kulturpf. 2nd ed. Stuttgart.
Klebahn, H. 1912. Grunzüge der algemeinen Phytopathologie. Berlin.
Klemm, M. 1938. Deut. Landw. Presse 65: 239.
Knorr, L. 1920. Prakt. Ratg. Obst. -u. Gartenb. 35: 344.
K. M. 1919. Erfurt Führer Obst. -u. Gartenbau. 21.
Koblischek. 1929. Mitt. Deut. Landw. Gesell. 1929: 250.
Köck, G. 1911. Centralbl. Landw. 1911: 15.
Köhne. 1928. Nachr. über. Schädlungsbekampf. 3: 61.
Korff, G., and K. Böning. 1927. Prakt. Bl. Pflanzenb u. Pflanzensch 5: 192.
Kreuzpointer, J. 1922a. Wegweiss. Obst. -u. Gartenb. 30: 131.
———. 1922b. Deut. Gemüsebau-Zeit. 10: 263.
———. 1923. Lehrm. Gart. Kleintierk. 21: 68.
———. 1929. Prakt. Bl. Pflanzenb. u. Pflanzensch 7: 96.
———. 1931. Ernährung der Pflanze 27: 172.
Krüger, R. 1920. Erfurter Führer Obst. -u. Gartenbau 21: 112. 1927, *Ibid.* 28: 31.
Kühn, J. 1858. Die Krankheit der Kulturgewasche, pp. 252, 253.
Küpke, W. 1933. Gartenwelt 37: 182.
———. 1935. Nachr. Schädl. Bekamp. 10: 16.
Küster, E. 1911. Die Gallen der Pflanzen. Leipzig.
Langenbeck. 1904. Deut. Landw. Presse 1904, no. 68.
Laubert, R. 1905. Prakt. Bl. Pflanzenb. u. Pflanzensch. 3: 73.
Leines, W. 1926. Nachrlb. Schädlingsbek. 1: 179.
Linder, H. 1911. Prakt. Ratg. Obst. -u. Gartenb. 26: 138.
Löwenthal, W. 1905. Zeit. Krebsf. 3: 16.
Ludwigs, K. 1925. Mitt. Deut. Landw. Ges. 40: 314.
Magnus, P. 1893. Abh. Gesell. Isis 1893: 8.
———. 1903. Abh. Nat. Gesell. Nürnberg 16.
Meyer, B. 1888. Landw. Jahrb. 17: 915.
Miller, P. 1923. Wegweiser im Obst. -u. Gartenbau 1923: 67.
Mothes, P. 1925. Deut. Obst. u. Gemüsebauzeit 71: 389.
Naumann, A. 1912. Der Handelsgärt. 14: 104.
———. 1913. Sitzb. Abh. Kgl. Gesell. "Flora" Bot. Gartenb. 17: 62.
Neger, F. W. 1917. Sächs. Landw. Zeitschr. 65: 145.
———. 1918. Illustr. Schles. Monatschr. Obst.-Gemüse. -u. Gartenb. 7: 66.
N. N. 1853a. Hamburger Garten -u. Blumenzeit. 1853: 433, 514.
———. 1853b. Zeitschr. Landw. Ver. Rheinprov. 1853: 226.
———. 1893. Erfurt. Illustr. Gartenzeit. 1893: 58.
Nötzel. 1922. Mitt. Gart. -Obst. -u. Weinbau 21: 64.
Panek, P. 1935. Deut. Gart. 50: 269.
Pape, H. 1925. Pflanzenbau 2: 172.
Pettera, A. 1917. Zeit. Gärtner u. Gartenfr. 13: 36.
Ponkler. 1896. Prakt. Ratg. Obst. -u. Gartenb. 1896: 408.
Popp, M. 1919a. Oldenburg Landw. Bl. no. 38: 553.
———. 1919b. Illustr. Landw. Zeit. no. 87/88: 439.
———. 1921. Landw. Jahrb. 55: 549.
Poter, G. 1917. Prakt. Ratg. Obst. -u. Gartenb. 22: 177.
Prowazek, S. 1902. Österr. Bot. Zeitschr. 52: 213.
———. 1905. Arb. Kais. Gesund. 22: 396.
Rabbas, P. 1930. Ratschlage f. Haus, Garten, Feld 5: 54.
Rath, L. 1933. Obst. -u. Gemüsebau 79: 63.
Rattke, R. 1921. Der Schles. Kleingartenfr. 1921: 290.
Rau, E. 1925. Blatt. f. d. d. Deut. Hausfrau no. 18.
Regel, E. 1879. Gartenflora 28: 170.
Remy, T., and G. Schneider. 1911. Zeitschr. Pflanzenkr. 21: 39.
———, and E. Lüstner. 1911. Veröff. Landw. Kammer. Rheinprov. no. 3: 24.

Riehm, E. 1913. Mitt. Biol. Reichanst. f. Land -u. Forstw. 24.
———. 1925a. Zeitschr. Agnew. Chem. 38: (1032).
———. 1925b. Deut. Obst. -u. Gemüsebau-Zeit. 71: 193.
Roger, A. L. 1912a. Erfürt Führer Obst. -u. Gartenbau 1912: 285. 1912b. *Ibid.* 1912: 185.
Roth, B. 1923. Wegweiser Obst. -u. Gartenb. 1923: 67.
Rump, L. 1925. Der Westdeutsch. Landw. no. 9, 3: 2–4.
Sattler. 1921. Deut. Obstbauzeit. 67: 281.
Schaffnit, E. 1922. *Ibid.* 68: 211.
———, and K. Meyer-Hermann. 1930. Phytopath. Zeitschr. 2: 99.
Schlodder. 1923. Lehrm. Gart. Kleintierh. 21: 393.
Schlumberger, O. 1914. Deut. Landw. Presse 41: 910.
Seelhoff, R. 1912. Prakt. Ratg. Obst. -u. Gartenb. 27: 157.
Seifert, H. 1916. *Ibid.* 31: 42.
Sommer, H. 1922a. Suddeutsch. Gartner-Zeit. Mannheim no. 4.
———. 1922b. Deut. Obstbauzeit. 68: 43.
———. 1922c. Erfurt Führer Obst. -u. Gartenb. 1922: 67.
———. 1922d. Gartenwelt 26: 70.
Soraur, P. 1874. Handbuch der Pflanzenkrankheiten. Berlin.
Straube. 1922a. Gesunder Viehstand. Stuck 23.
———. 1922b. Kleintierz. Gartenb. 41: 100.
Strohmeyer, A. 1913. Die Gartenw. 17: 267.
Tessenow, M. 1922. Möller's Deut. Gärtner-Zeit. 37: 91.
———. 1926a. Gartenwelt 30: 301.
———. 1926b. Obst. -u. Gemüsebau 72: 310.
Trieschman. 1917. Landw. Wochenbl. Schles-Holstein 67: 289.
Vielhauer. 1920. Georgine 13: 514.
Volmann, E. 1923. Erfurt Führer Obst. -u. Gartenb. 24: 183.
Vonwiller, P. 1918. Arch. Protistk. 38: 279.
Vouk, V. 1913. Die Naturw. 1: 81.
W. D. 1919. Illustr. Flora 1919: 27.
Wagner-Ettelbrück, J. P. 1908. Monatsb. Gesell. Luxemb. Naturfr. n. f. 1908: 292.
Wagner, J. P. 1909. Mitt. Deut. Landw. Gesell. 1909: 610.
Wähling, G. 1922a. Mitt. Garten-, Obst. -u. Weinb. 21: 7.
———. 1922b. Deut. Obstbau-Zeit. 1922. H. 3: 30.
———. 1924. Deut. Obst. -u. Gemüsebau-Zeit. 70: 178.
Weiss, E. 1918. Zeitschr. Pflanzenkr. 28: 116, 201.
Wisselingh, van C. 1898. Jahrb. Wiss. Bot. 31: 619.
Zopf, W. 1884. Enzykl. Naturw. 3: 129.

HOLLAND

Bos, R. 1902. Zeitschr. Pflanzenkr. 12: 344. 1904. *Ibid.* 14: 319.
———. 1917. Tidjschr. Plantenziek. 23: 34.
———, and J. G. Staes. 1898. Zeitschr. Pflanzenkr. 8: 350.
———, and J. G. Staes. 1901. Tijdschr. Plantenziek. 7: 1–202.
Pfeiffer and J. G. Staes. 1902. Zeitschr. Pflanzenkr. 12: 344.
Poeteren, van N. 1930. Tidjschr. Plantenziek. 35: 256.
Staes, J. G. 1900. *Ibid.* 6: 139.

HUNGARY

Bernátsky, J. 1915. A golyvásodás Plasmodiophorabetegség. Kertészet, 1919: 111.
Hegyi, D. 1904. Konyhakerti novenyeink legfontosabb betégsegei. Magyarovár 1904: 41.
Jablonowski, J. 1900. A káposztafélék gyökerének golyvásodásáról. A. kert. 1900: 93.
Moesz, G. 1926. Novenyvedelen 2: 4.
———. 1938. Ann. Hist. Nat. Mus. Nation. Hungarici 31: 63.
Schilberszky, K. 1901. A káposztagyökerek golyvasodása. Kertesz. Lap. 1901: 346.
———. 1906. A Kert. 7: 670.

INDIA

Butler, E. J. 1918. Fungi and diseases in plants. Calcutta.
———, and G. R. Bisby. 1931. The fungi of India. Calcutta.
McRae, W. 1928. Sci. Rept. Agric. Res. Inst. Pusa 1927/28: 56.
Sharangapani, S. G. 1930. Ann. Rept. Eco. Botanist Gov't Beng. 1929/30: 37.

IRELAND

Atkins, W. R. G. 1922. Proc. Roy. Dublin Soc. n. s. 16: 427.
Bailie, D. W., and A. E. Muskett. 1933. Jour. Min. Agric. North Ireland 4: 44.

ITALY

Briosi, G. 1904. Zeitschr. Pflanzenkr. 14: 213.
———, and F. Cavara. 1891. I funghi parassiti delle plante coltivate odutili. Fasc. VI. Pavia.
Perotti, R. 1940. Biologia vegetale applicata all 'agricoltura. III. Micologia-malattie parassitarie. Turin.
Petri, L. 1924. Congresso delle Cattedre Ambulanti di Agric. Italiane il 10 Maggio 1924.
———. 1936. Boll. Staz. Path. Veg. Roma 16: 1–25.
Pollacci, G. C. Rend. Acad. Linn. 5 ser. 20: 218.
———. 1911. Atti Inst. Bot. Univ. Pavia 2 ser. 14: 103. 1914a. *Ibid.* 15: 281. 1914b. *Ibid.* 15: 291.
Saccardo, P. A., and F. Cavara. 1900. Nuovo Giorn. Bot. Ital. n. s. 7: 272–310.
Voglino, P. 1907. Ann. R. Acad. d'Agric. 18: 42. 1908. *Ibid.* 50: 247.

JAPAN

Ikeno. 1929. (See Honig. Gartenbauwiss. 5: 119.)

JUGOSLAVIA

Jaap, O. 1916. Ann. Mycol. 14: 1–44.

LITHUANIA AND ESTONIA

Vilkaitis, V. 1933. Istaigos Darbai, Kaunas 1933: 7–55.

Elenkin, A. A., and A. S. Bondarzew. 1913. Jahrb. Pflanzenkr. (Russian) 1913: 218.

NEW ZEALAND

Anonymous. 1927. N. Z. Jour. Sci. Tech. 9: 44.

Gibbs, J. G. 1931a. N. Z. Jour. Agric. 42: 1. 1931b, *Ibid.* 42: 193.

———. 1931c. N. Z. Jour. Sci. Tech. 13: 104.

———. 1932a. N. Z. Jour. Agric. 44: 28. 1932b, *Ibid.* 44: 273.

———. 1932c. N. Z. Jour. Sci. Tech. 14: 145. 1934, *Ibid.* 16: 159.

———. 1935. N. Z. Jour. Agric. 50: 294.

———. 1939. N. Z. Jour. Sci. Tech. 20A: 409.

Neill, J. C. 1933. N. Z. Jour. Agric. 46: 207. 1935, *Ibid.* 50: 232.

Tennent, R. B. 1925. *Ibid.* 30: 259. 1930, *Ibid.* 41: 318. 1933, *Ibid.* 47: 297.

NORWAY

Brunchorst, J. 1887. Bergen's Mus. Aarberet. for 1886: 227.

Jorstad, I. 1922. Rept. Min. Agric. 1922.

———. 1923. Norsk. Havetid 39: 126.

Schoyen, W. M. 1895. Zeitschr. Pflanzenkr. 5: 173. 1898, *Ibid.* 8: 211. 1899, *Ibid.* 9: 302.

PALESTINE

Rayes, T. 1938. Palestine Jour. Bot. J. ser. 1: 143.

POLAND

Siemaszko, W. 1925a. Zentralbl. Bakt. Abt. II. 66: 116.

———. 1925b. Diseases and pests of plants. Warsaw 1: 40.

Sitensky. 1898. Zeitschr. Pflanzenkr. 8: 148.

Svec, F. 1922. Ochr. Rost. 2: 59. 1923: *Ibid.* 3: 18.

PORTUGAL, AZORES

Almeida, d'J. V. 1902. Zeitschr. Pflanzenkr. 12: 349.

ROUMANIA

Moez, G. 1930. (See Honig, 1931. Gartenbauw. 5: 120.)

RUSSIA

Bondarzew, A. S. 1903. Zeitschr. Pflanzenkr. 13: 219.

———. 1906. Acta Horti Petrop. 26: 1–22.

———. 1909. Boljezni Rast. 1909: 109.

———. 1927. Morbi Plant. 16: 1–17.

Brezhnev, I. E. 1934. Trav. Soc. Nat. St. Petersburg 63: 83.

———. 1937a. Trudy Leningr. Obsch. Estestv. Otd. Bot. 66: 188. 1937b, *Ibid.* 66: 296.

Diakonoff, H. J. 1910. Zeitschr. Pflanzenkr. 20: 463.

Elenkin, A. A., and A. S. Bondarzew. 1913. Jahrb. Pflanzenkr. (Russ.) 1913: 218.

Favorsky, W. 1910a. Mem. Soc. Nat. Kieff 20: 149.

———. 1910b. Zapiski Kiews Absc. Estedvoisp. 20.

Fedorintschik, N. S. 1935a. Summ. Sci. Res. WK. Inst. Pl. Protect. Leningr. 5: 61–66. 1935b, *Ibid.* 5: 87–89.

Fedotova, T. 1928. Mater. Mikol. i. Fitopat. 7(1): 155.

———. 1930. Phytopath. Zeitschr. 1: 195.

———. 1933a. Bull. Pl. Protect. II. ser. Phytopath. no. 3: 3. 1933b, *Ibid.* no. 3: 51.

Henckel, A. G. 1921. Wseross. Entomo-Phytopath. Siesda. Bull. III. n. s. 6: 20.

———. 1923. Bull. Inst. Rech. Biol. Perme II. fasc. 2.

Iwanoff, K. S. 1900. Zeitschr. Pflanzenkr. 10: 97.

Jaczewski, A. 1893. Bull. Soc. Mycol. France 9: 212.

———. 1908. Zeitschr. Pflanzenkr. 18: 33. 1911, *Ibid.* 21: 283.

Katterfeld, B. O. 1923. Morbi Plant. Script. Sec. Phytopath. Hort. Bot. Princ. Ross. 12: 11.

Kleimenov, P. D. 1912. Rept. Phytopath. Res. 1912 made by K. E. Nurashkinsky and P. D. Kleimenov, Moscow Govt. Zemstvo. 2nd paper, pp. 26–37.

Ludwig, F. 1910. Zeitschr. Pflanzenkr. 20: 84.

———. 1915. XI. Phytopath. Ber. Biol. Zentralstelle. Furstentumer Reuss a. L. und Reuss j. L. über das Jahre 1915. Greiz. 1916. XII. *Ibid.* 1916. 1917, XIII. *Ibid.* 1917.

Morozov, B. 1926. La Defense des Plantes 2: 592.

Naumov, N. A. 1925. Morbi Plant. 14: 49.

———. 1927. La Defense des Plantes 4: 320.

———. 1928. Morbi Plant. 17: 51.

———. 1933. Bull. Pl. Protect., ser. II. Phytopath. no. 3: 32.

Nawaschin, S. 1899. Flora 86: 404.

———. 1924. C. R. Acad. Sci. Russ. 1924: 173.

Newodowsky, G. 1910. Chosiastwo, Kiew 5: 990.

Podwyssozki. W. 1900. Centralbl. Bakt. Parasitk. Infekt. abt. I. 27: 97.

———. 1903. Zeitschr. Klin. Med. 47: 199.

Polyakoff, I. M. 1939. Bull. Pl. Protect. 1939: 60.

Potebnia, A. 1915. Parasitische Pilze des Gouv. Charkoff und angegrenzte Gebiete 1: 56–68.

Rochlin, E. 1933a. Phytopath. Zeitschr. 5: 381.

———. 1933b. Bull. Pl. Protect., ser. II. Phytopath. no. 3: 8–31.

Speschnew, N. N. 1896. Arb. Bot. Gart. Tiflis 1: 65. 1897, *Ibid.* 2: 199.

———. 1899. Zeitschr. Pflanzenkr. 9: 356.

Ssacharoff, M. 1916. Arb. Phytopath. Sta. Inst. f. Landw. Moscow. 1. 1916.

Troitzky, N. N. 1921. Proc. and All-Russ. Ento. Phytopath. Meet. Petrograd 1921: 165.

Vladimirskaya, N. N. 1930. Morbi Plant. 19: 22.

Woronin, M. 1874. Protocoll Bot. Sect. St. Petersburg Nat. Ges., Mar. 5, 1874.

———. 1875. Bot. Zeit. 33: 337.
———. 1877. Arb. St. Petersburg Nat. Gesell. 8: 169.
———. 1878. Jahrb. wiss. Bot. 11: 518.
———. 1880. Bot. Zeit. 38: 54.

SCOTLAND

Abbay, J. 1831. Trans. Highland Agric. Soc. Scotland n. s. 2: 238.
Anderson, A. 1855. *Ibid.* 3 ser. 6: 118.
Anonymous. 1923. North Scot. Coll. Agric. Bull. 29. 1931, *Ibid.* 1930 31.
Bird. 1859. Trans. Highland Agr. Soc. Scotland 8: 307.
Birnie, M. 1831. *Ibid.* n. s. 2: 241.
Farquharson, J. 1831a. *Ibid.* n. s. 233. 1831b. *Ibid.* n. s. 2: 338.
Findlay, W. M. 1931. Scot. Jour. Agric. 14: 173.
Fowlie, A. T. 1913. North Scot. Coll. Agric. Exp. Leaf. 38: 102.
Hendrick, J. 1923. Trans. Highl. Agric. Soc. Scotland 35: 54. 1932, *Ibid.* 44: 52.
Hunter, W. B. 1857. *Ibid.* (3) 7: 347.
MacDonald, J. A. 1935. Scot. Jour. Agric. 18: 164.
O'Brien, D. G., and R. W. G. Dennis. 1936. *Ibid.* 19: 4–18.
Pardy, A. 1913. North Scot. Coll. Agric. Exp. Leaf. 25: 52.
Sanderson. 1859. Trans. Highl. Agric. Soc. Scotland. 3 ser. 9: 37.
Wright, R. P. 1911. Rept. West Scot. Agric. Coll. 1911.

SPAIN

De Andres, C. G. 1929. Bol. Pav. Veg. y. Ent. Agric. 4: 98.
Rangel, E. 1914. Pragas dos pomares e das hortas. Lavo 17: 308.

SWEDEN

Anonymous. 1923. Kunzel. Landthr. Acad. Handl. och. Tidskr. 62: 357.
Ericksson, J. 1907. Zeitschr. Pflanzenkr. 17: 276.
———. 1888. Fungi parasitici Scandinavici exs. Fasc. 6.
———. 1912. Fungoid diseases of agricultural plants. London.
———. 1913. Die Pflanzenkr. Landw. Kulturpflanz. Leipzig.
———. 1926. Die Pflanzenkr. Landw. Kulturgewächse. Stuttgart.
Hammarlund, C. 1915. Medd. 106. Centralanst. f. j. Försök. p. Jordbruk.
Hedlund, T. 1910. Zeitschr. Pflanzenkr. 20: 143.
Hertzman, N. 1934. Tidskr. f. Landman 18: 11–13. 1935, *Ibid.* 19: 216.
Lindfors, T. 1921a. Kgl. Landtb. Akad. Hand. Tidskr. 63: 267.
———. 1924b. Medd. no. 262. Centralanst. f. Forsök. p. jordbr. Bot. avd. no. 32. 1925. *Ibid.* Medd. no. 282.
Olsson, P. A. 1939a. Sveriges Utsädesf. Tidskr. 49: 1. 1939b. *Ibid.* 49: 471. 1940, *Ibid.* 50: 287.
Tedin, O. 1932. Nord. Jordbr. 14: 324.

SWITZERLAND

Anonymous. 1907. Schweiz. Lands. Zeitschr. 35: 515.
———. 1922. Schweiz. Zeitschr. Obst. -u. Weinb. 31: 120.
Doppler, J. 1926. Schweiz. Obst. -u. Gartenb. Zeit. 1926: 300.
Fischer, E. 1923. Ber. Schweiz. Bot. Gesell. 32: 41.
Heinzelmann, F. 1882. Schweiz. Landw. Zeit. 10: 526.
Müller-Thurgau, H., and A. Osterwalder. 1919. Landw. Jahrb. Schweiz. 33: 1. 1923, *Ibid.* 37: 153. 1924, *Ibid.* 38: 613.
Osterwalder, A. 1922. Schweiz. Obst. -u. Gartenb. Zeit. 1922: 205.
———. 1929. Landw. Jahrb. Schweiz. 43: 785.
Schinz, H. 1923. Ber. Schweiz. Bot. Gesell. 32: 12.
Schmidt, A. 1919. Landw. Jahrb. Schweiz. 33: 513.
Weber, W. 1926. Schweiz. Obst. -u. Gartenb. Zeit. 1926: 301.

TASMANIA

Nichols, H. M. 1922. Tasmania Agric. Stock Dept. Rept. 1920/21.

UNION OF SOUTH AFRICA

Doidge, E. M. 1924. Bot. Surv. S. Africa, Mem. no. 6: 1–56.
Pole-Evans, J. B. 1913. Agric. Jour. S. Africa 6: 93.

U. S. A.

Anonymous. 1910. Amer. Agric. 31, no. 14.
———. 1922. Penn. Agric. Exp. Sta. Bull. 170.
———. 1924. Pl. Dis. Rept. Suppl. 34: 168, 178, 183.
———. 1925. Ann. Rept. Penn. Agr. Exp. Sta. 1925: 14.
———. 1926. Pl. Dis. Rept. Suppl. 45: 16, 53, 80.
———. 1931. Conn. Agr. Exp. Sta. Bull. 337: 451.
———. 1937. New Jersey Agr. Exp. Sta. Ann. Rept. 58: 80.
———. 1938. 15th Ann. Rept. Rhode Island State Coll. Agric. Exp. Sta.
———. 1939. Ann. Rept. Rhode Island State Coll. Exp. Sta. 51: 1–64.
Atkinson, G. F. 1889a. Alabama Exp. Sta. Bull. 9.
———. 1889b. Ann. Rept. South Carolina Exp. Sta. 1: 15–18.
Bailey, F. D. 1912. Oregon Agr. Coll. Exp. Sta. Crop. Pest. Hort. Rept. 1: 273.
Clupp, C. 1917. Cornell Agr. Exp. Sta. Bull. 387.
———. 1925. Manual of vegetable-garden diseases. New York.
———. 1928. Phytopath. 18: 30.
Clayton, E. E. 1926. New York Agr. Exp. Sta. Bull. 537.

Clinton, G. P. 1903. Rept. Conn. Agr. Exp. Sta. 1903: 310, 367.
Cook, M. T. 1923. Sci. n. s. 57: 6.
Coons, G. H. 1922. Pl. Dis. Bull. U. S. Dept. Agr. Suppl. 22: 357.
Crosby, C. R., and C. Chupp. 1934. Cornell Univ. Ext. Bull. 278.
Cunningham, G. C. 1912. Phytopath. 2: 138.
———. 1914. Vermont Agr. Exp. Sta. Bull. 185.
Davis, W. H. 1925. Mycologia 17: 160.
Duggar, B. M. 1909. Fungous diseases of plants. New York.
Edison, H. A. 1937. Pl. Dis. Rept. Suppl. 103.
———, P. R. Miller, and J. I. Wood. 1935. *Ibid.* Suppl. 90: 72.
———, and J. I. Wood. 1936. *Ibid.* Suppl. 96: 195–200.
Eyeleshymer, A. C. 1891. Jour. Mycol. 7: 79.
Gaylord. 1903. Zeitschr. Krebsforsch. 1: 93.
Gilbert, W. W., and C. H. Popenoe. 1924. U. S. Dept. Agr. Farm. Bull. 1371.
Gilman, A. C., and W. A. Archer. 1929. Iowa Jour. Sci. 3: 299.
Glover, W. O., and H. Glasgow. 1924a. Phytopath. 14: 25.
———, and ———. 1924b. New York Agr. Exp. Sta. Bull. 513.
Haenseler, C. M., and T. R. Moyer. 1937. Soil Sci. 43: 133.
Halsted, B. D. New Jersey Agric. Coll. Exp. Sta. Bull. 98.
———. 1890. Gard. and Forest. 3: 541.
———. 1891. New Jersey Agr. Exp. Sta. Rept. 1891: 242. 1893. *Ibid.* 1893: 332.
———. 1894a. Bull. Torrey Bot. Club 21: 76.
———. 1894b. New Jersey Agr. Coll. Exp. Sta. Rept. 1894: 278. 1895, *Ibid.* 1895: 250. 1896, *Ibid.* 1896: 290. 1897a, *Ibid.* 1897: 265. 1897b, *Ibid.* 1897: 287.
———. 1898a. Amer. Gard. 19.
———. 1898b. New Jersey Agr. Coll. Exp. Sta. Rept. 1898: 292. 1899, *Ibid.* 1899: 354. 1901, *Ibid.* 1901: 388. 1902, *Ibid.* 1902: 398.
Harshberger, J. W. 1917. A text-book of mycology and plant pathology, Philadelphia.
Harter, L. L., and L. R. Jones. 1923. U. S. Dept. Agr. Farm Bull. 1351.
Hawks, W. 1898. Rept. Agr. Exps. Cornwall County Council 1898: 27.
Heald, F. D. 1933. Manual of plant diseases. 2nd ed. New York.
Henderson, P. 1867. Gardening for profit. New York.
———. 1879. Cult. and Country Gentleman 44: 807.
Hulst, G. G. 1888. New Jersey Agr. Exp. Sta. Bull. 50: 14.
Humphrey, J. E. 1892. Massachusetts Agr. Exp. Sta. 9th Ann. Rept. 1891: 230.
Humphrey, H. B., and J. I. Wood. Pl. Dis. Rept. Suppl. 86: 52.
Jones, L. R. 1901. Vermont Agr. Exp. Sta. Bull. 66.
———. 1924. Am. Jour. Bot. 11: 601.
Jones, P. M. 1928. Arch. Protist. 62: 313.
Kadow, K. J. 1937. Pl. Dis. Rept. 21: 405.
Kunkel, L. O. 1918. Jour. Agr. Res. 14: 543.
Larsen, R. H. 1934. *Ibid.* 49: 607.
———, and J. C. Walker. 1934. *Ibid.* 48: 749.
Levine, I., and M. Levine. 1922a. Jour. Cancer Res. 7: 163. 1922b, *Ibid.* 7: 174.
Lutman, B. F. 1913. Vermont Agr. Exp. Sta. Bull. 175.
MacLeod, D. J., and J. L. Howatt. 1934. Amer. Potato Jour. 11, 3: 60.
Maneval, W. E. 1937. Univ. Missouri Studies 12: 3.
Martin, W. H. 1928. 48th Ann. Rept. New Jersey Agr. Exp. Sta. pp. 205–238.
———. 1933. *Ibid.* 53rd and 54th Ann. Repts. pp. 57–66.
Middleton, J. T., and M. W. Gardner. 1941. Pl. Dis. Rept. 25: 94.
Milburn, B., and E. A. Bessey. 1915. Fungoid diseases of farm and garden crops.
Moldenke, H. N. 1934. The observed flora of Watchung, N. J.
Montieth, J. 1924. Jour. Agr. Res. 28: 549.
Murphy, P. A. 1927. Jour. Dept. Lands and Agr. 27: 12.
Nance, N. W. 1941. Pl. Dis. Rept. Suppl. 128.
Owen, C. E. 1928. Principles Plant Pathology. New York.
Palmer, R. G. 1941. Phytopath. 31: 18.
Pryor, D. E. 1940. Jour. Agr. Res. 61: 119.
Reed, H. S. 1911a. Phytopath. 1: 159.
———. 1911b. Virginia Agr. Exp. Sta. Bull. 191.
Russel, H. L., and F. B. Morrison. 1922. Wisconsin Agr. Exp. Sta. Bull. 339.
Schwarze, C. A. 1917. New Jersey Exp. Sta. Bull. 313.
Slingerhand, M. V. 1894. New York Agr. Exp. Sta. Entom. Div. Bull. 74: 481.
Smith, W. G. 1894. Diseases of field and garden crops. Pp. 94–104.
Smith, R. E., and E. H. Smith. 1911. California Agr. Exp. Sta. Bull. 218.
Stevens, F. L. 1925. Plant disease fungi. New York.
Stewart, F. C. 1895. New York Agr. Exp. Sta. Ann. Rept. 14: 519.
Tabenhaus, J. J. 1920. Diseases of greenhouse crops and their control. New York.
Tillinghast, I. F. 1879. Cult. and Country Gentleman 44: 807.
Tompkins, C. M., and P. A. Ark. 1939. Pl. Dis. Rept. 23: 4.
Vaughan, R. E. 1924. *Ibid.* 34: 192.
———, and F. L. Wellman. 1926. Wisconsin Agr. Exp. Sta. Circ. 200.
Walker, J. C. 1934. U. S. Dept. Agr. Farm Bull. 1139.
———. 1935. Jour. Agr. Res. 51: 183.
———. 1936. Phytopath. 26: 112.
———. 1937. Rept. Wisc. Agr. Exp. Sta. Bull. 438.
———. 1939. Jour. Agr. Res. 59: 815.
———. 1941. Botanical Rev. 7: 458.

Walker, J. C., and R. H. Larsen. 1934. Wisconsin Agr. Exp. Sta. Bull. 428.
———, K. P. Link, and S. Morell. 1937. Wisc. Agr. Exp. Sta. Bull. 435.
Wellman, F. L. 1930. U. S. Dept. Agr. Tech. Bull. 784.
Wilson, J. D. 1934. Ohio Agr. Exp. Sta. Bimonthly Bull. 19: 58.

WALES

Davies, D. W., M. Griffith, and G. Evans. 1928. Welsh Jour. Agr. 4: 295.
Preston, N. C. 1928. *Ibid.* 4: 280.
———. 1931. Jour. Min. Agr. 38: 272.
Whitehead, T. 1922. *Ibid.* 29: 362.
———. 1925. Welsh Jour. Agr. 1: 176. 1935, *Ibid.* 11: 228. 1936, *Ibid.* 12: 183. 1940, *Ibid.* 16: 99.

POWDERY SCAB OF POTATOES

Powdery scab of potatoes is now almost world wide in distribution. Lagerheim's discovery of its presence in Ecuador suggests that it may be endemic to South America, since that continent is generally regarded as the native home of the potato. In that event powdery scab may be of greater antiquity than is generally supposed. It has doubtless attained its extensive distribution by the shipment and importation of infected tubers. From South America it may have been imported to Europe and then back to North America and other parts of the world. Powdery scab was first reported from Germany by Wallroth and Martius in 1842, but it had doubtless been known by potato growers for many years before that time. Shortly afterwards, it was described from England by Berkeley ('46) and later from Wales, Scotland, Norway and Ireland. The first record of its occurrence in North America was made in 1913 on potatoes in Quebec, and in the same year it was also found in Maine and other states. This disease is known by a variety of names throughout the world. In Germany it is described as Knollenbrand, Kartoffelgrind, Kartoffelgnatz, Schorfkrankheit, and Kartoffelschorf. In England, Scotland, Wales, Ireland and the U. S. A., it goes by the names of potato canker, corky end, corky scab, spongy scab, *Spongospora* scab, and potato tumor, although powdery scab is the term most commonly used. The names, potato canker and tumor are employed when the lesions and tumors are unusually deep and conspicuous.

As a destructive disease of potatoes, powdery scab is of secondary importance compared with late blight, virus, *Fusarium* wilt and rot, and common scab. In relatively dry and warm regions the damage caused may be so slight as to go unnoticed, while in other places with high precipitation and low temperatures the losses may be quite serious, particularly if the disease is of the cankerous type and is followed by powdery scab dry rot in storage. In England, Wales (Pethybridge, '24), New Zealand (Anony., '27), Peru (Abbott, '31), and Russia (Dorojkin, '36), destruction of 30 to 50 per cent and more of the crop has been reported in years of heavy rainfall and low temperature. Likewise, Melhus, *et al.*, found that 30 to 73 per cent of infected tubers may be destroyed by dry rot in storage or rendered useless for table or planting. Such losses, however, appear to be exceptional, but the disease is nevertheless of sufficient importance to warrant the establishment of strict quarantine and tuber inspection and certification laws by most countries throughout the world.

Predisposing Factors

The occurrence of powdery scab and incidence of infection are dependent on climatic conditions. Heavy rainfall, fairly low temperatures, damp, poorly-drained and water-logged soils favor infection and development of the disease. Melhus, *et al.*, observed that periods of rainfall, followed by cool, damp, cloudy weather during the growing season are highly essential to the development of the disease, and these observations were subsequently confirmed by Ramsey ('18) from greenhouse experiments. He found 83 per cent infection in pots of potatoes grown at 57°–60° F. under moist conditions, while no infection occurred in pots at 76°–80° F. and in relatively dry soil. Wild ('29), on the other hand, found no clear correlation between incidence of scab and the prevalence of any particular climatic conditions in Switzerland. Koltermann's ('31), Phillips' ('32), and Naumov's ('36) observations on the disease in Germany and Russia confirm those of Melhus, Ramsey, and others in America. Naumov found powdery scab to be more prevalent in soils with 60–90 per cent moisture content and with pH values from 4.7 to 5.9 than in soils with 10 per cent moisture and high pH values.

That unfavorable climate is an effective barrier to the spread of the disease is evident from experiments conducted by Melhus, *et al.*, which involved planting of heavily infected tubers in fifteen different regions along the Atlantic Coast from Massachusetts to Florida. All of these plantings yielded clean crops. These results are supported by the observations of Shepherd ('35), Nattras ('38), and Littlejohn ('39) that heavily infected imported tubers planted in Mauritius and Cyprus give healthy crops and that *S. subterranea* does not remain viable under prevailing climatic conditions on those islands.

Hydrogen ion concentration apparently does not influence the incidence of infection, since *S. subterranea* appears to tolerate both alkaline and acid reactions. Wild, Phillips, and Naumov found that infection may readily occur in soils with pH values ranging from 4.7 to 7.6. Furthermore, the incidence of infection is not affected by the carbonate or hexosan content of the soil, according to Wild.

The physical character of the soil, however, is an important factor. A close correlation between cer-

tain soil types and the degree of infection was observed by Melhus, *et al.*, in Maine, and they were accordingly able to predict the extent of development of the disease from the type of soil and its drainage. Wherever the Washburn silt-loam type of soil occurred infection was unusually heavy. Wild, likewise found that powdery scab flourishes in Switzerland in soils with a large pore space, high humus and methylpentosan content, coarse texture, and high water-holding capacity.

Symptoms

Powdery scab may manifest itself as shallow, scabby lesions or deep eroded cankers on the tubers, and galls or warts on the roots and stems. These phases of the disease may be followed by powdery scab dry rot after the tubers have been harvested and stored. The first evidence of infection on the tubers is the appearance of faint brownish-purple spots of pinhead size, which doubtless indicate the point of entry of the parasite. Each spot is usually surrounded by a circular translucent, 1 to 2 mm., area which apparently marks the distance to which the plasmodium has spread beneath the epidermis, according to Kunkel ('15). In the course of 6 to 8 days the areas may increase to ½ cm. in diameter, lose their brownish color, and protrude as a metaplastic, jelly-like mass of proliferating host cells and fungus spores. According to Horne ('12) these protrusions may be so prominent that they look like cushions or wart-like excrescences. The diseased areas gradually die, leaving shallow, crateriform depressions filled with a fine powdery mass of spore balls (Pl. 10, fig. 1). These are the so-called powdery scab symptoms of the disease which may be readily mistaken for those of the common scab.

Further development of the disease on the tubers depends to a great extent on the relative amount of moisture in the soil or in the storage bins after the potatoes have been harvested. If the infected tubers are growing in fairly dry soil, wound cork is rapidly formed under and around the lesions, so that the diseased areas are delimited. With abundant moisture and in poorly drained soil, however, the parasite may continue its depredations. As a result the lesions become deeper, larger and sometimes coalesce to form extensive eroded cavities or cankers as much as ½ inch in depth. This is one of the most severe types of the disease and is referred to as the canker-ous stage (fig. 2). This type appears to be common in Ireland, England, and Europe, but is not very prevalent in Maine and Canada. Melhus, *et al.*, attributed the latter to the shorter growing period of the potato in the northern regions of North America. In addition to causing shallow lesions and deep cankers *S. subterranea* may also lead to the formation of tuberous outgrowths and extensive warts on the tubers with the result that the latter are often misshapen and deformed, according to Horne ('12). These outgrowths are apparently formed in the same manner as the galls on the roots and stems, although Horne did not describe their development. They may be more or less uniformly infected and covered with scabs and bear a superficial resemblance to the tumors caused by *Synchytrium endobioticum*.

The galls on the roots and stolons of potatoes and other related species vary in size from minute tubercles to balls as large as garden peas (fig. 3). They usually precede tuber infection and may be present in abundance before there is any indication of lesions on the tubers, but their presence does not appear to have any great injurious effect on the growth of the host. These galls are simple in structure and consist primarily of enlarged and frequently divided undifferentiated cells, so that they are typically kataplasmic in structure. The causal organism is confined largely to the phloem and meristematic tissues, as in the case of club root of crucifers. Amoebae may be found occasionally in the xylem, but they do not occur in great numbers or cause distortion of the vessels. The presence of the parasite in the phloem stimulates the cells to enlarge and divide, and this hyperplastic growth often pushes the xylem out of its normal position.

Powdery scab dry rot usually sets in after infected tubers have been in storage for some time, and in some cases is abetted by numerous other fungi. This rot was first described by Melhus ('14) in North America, but it has been found subsequently on potatoes collected in Ireland, Holland, Chile, and other countries. It is accelerated by poor storage conditions, but even in good storage as much as 30 to 73 per cent of the tubers may be partly or wholly decayed and rendered useless for seed or table use, according to Melhus, *et al.* Although tubers may be often totally decayed, powdery scab dry rot is usually less severe and occurs in localized spots, 1 to 10 cms. in diameter. These areas may be only slight depressions in the superficial layers or extend to the center of the tubers. The extent of injury, however, depends to some extent on the time of harvesting, degree of infection, storage conditions, and the stage of development of the parasite when the tubers are stored. Dry rot may accordingly exhibit various types of symptoms. Desiccation or loss of water from the open lesions is a common occurrence when tubers are placed in warm dry storage and results in discoloration of the affected areas, wrinkling, shrinkage, and marked loss in weight. However, this type of dry rot is retarded as storage temperatures drop with the advent of the winter season. Another type of dry rot is caused by secondary infection and invasion of tissue around the old pustules by the plasmodium of *S. subterranea*. If moisture and temperature are favorable, the resting spores in old lesions may germinate and give rise to plasmodia which invade and kill the surrounding healthy cells. The plasmodium usually feeds on the tissue immediately beneath the epidermis, but occasionally it may be found at depths of 6 to 8 mm. in the tuber. In such extreme cases of penetration the

symptoms produced may resemble those of the cankerous stage in the field.

The open lesions may also be invaded by wound parasites of the genera *Phoma, Fusarium, Rhizoctonia, Papulospora*, etc., and this initiates the most destructive type of powdery scab dry rot. *Phoma tuberosa*, according to Melhus, *et al.*, is commonly associated with the early stages of rot and produces brownish to gray lesions in the bottom of the old pustules. As these lesions progress they become more sunken, darker and often hard and bony. At later stages the lesions may vary from 2 to 5 cms. in diameter and extend to a depth of 2 to 4 cms.

At this point it may be noted that Shapovalov ('23) contended that the skin-spot disease of tubers, which had been attributed to several causal organisms, including *Oospora pustulans*, is an early stage of powdery scab, but this was immediately refuted by Millard and Burr ('23). They reported that the former disease is caused solely by *O. pustulans* and is in no way related to powdery scab. Powdery scab is coextensive with late blight, caused by *Phytophthora infestans*, and both diseases are favored by the same climatic conditions. The latter disease may often be greatly increased by tuber and root infection by *S. subterranea*, according to Beregovoy ('39).

Cellular Relations Between Host and Pathogen

Spongospora subterranea has a marked effect on the host cells. Young infected cells as well as adjacent healthy ones are stimulated to divide by the presence of the parasite. The repeated division of healthy cells results in the formation of a new periderm around the regions of infection. When this periderm is invaded further cell divisions follow, which lead to the development of a second wound periderm, according to Wild ('29). Kunkel found a marked difference in reaction between the young growing cells of tubers and mature cells in the tissues around the old lesions. The former are not killed by invasion of the parasite but are stimulated to expand and divide. The latter, on the other hand, are quickly killed, and their contents are partly or wholly consumed. The increase in cell multiplication noted above is usually accompanied by cell enlargement. According to Kunkel, the latter process may begin while the plasmodium is still in the intercellular spaces and before it has entered the cells (fig. 17). This reaction suggests that the plasmodium may secrete a stimulating substance which precedes its invasion of the cells.

Infected cells may become 5 to 10 times their normal size, but enlargement is not equal in all directions. The expanding cells generally elongate outward towards the surface of the tuber, which finally results in the lifting and rupturing of the epidermis and the formation of cushion-like excrescences. In galls on the roots of *Solanum warcewiczii* and *L. esculentum* the infected cells occur in groups (fig. 16), according to Melhus, *et al.*, like the "Krankheitsherde" described by Nawaschin for club root of crucifers. These groups originate by continual division of one or more infected cells whereby the amoebae and young plasmodia are passively distributed. The nuclei of infected cells may divide mitotically and possibly amitotically also, as in the case of *Triglochinis* cells parasitized by *Tetramyxa Triglochinis*. When normal mitosis occurs, a cell plate is formed between the daughter nuclei (fig. 14), but in cases of amitosis the giant cells become multinucleate and later divide into numerous smaller cells, according to Kunkel. However, it is not obvious from his description whether these latter divisions occur by cell plate formation or cleavage after which walls are laid down. The host nucleus may be enveloped by or embedded in the plasmodium and become greatly enlarged, lobed, and distorted (fig. 18, 19, 26). Several nucleoli may frequently develop, while the chromatin strands become abnormal in appearance or disappear entirely. The nuclei are usually destroyed before the parasite is mature, but in exceptional cases it may remain intact until after the spore balls have been formed and lie between them.

The presence of *S. subterranea* apparently also stimulates an undue production of starch in and around infected cells. At least the starch appears to be more abundant in the regions of infection in the potato and tomato. In *S. warscewiczii*, however, Melhus, *et al.*, found that numerous infected cells may be found which are totally lacking in starch. The starch grains usually do not disappear entirely until after the spore balls are mature, but it is not certain that they are consumed directly by the parasite. Osborn claimed that the plasmodium feeds on starch, but Melhus, *et al.*, pointed out that if this were true an abundant supply would not always be present. Kunkel also reported that the starch grains are only slightly changed by the parasite and may remain after the cytoplasm and nuclei have been destroyed. Other workers, however, have claimed that the supply of starch diminishes as the parasite matures. Wild found that starch disappears below the diseased areas, being utilized in the process of cell division, or for nutrition of the parasite. It may also be noted here that infection with *S. subterranea* reduces the pH value of tubers from 5.70 to 4.35, according to Robertson and Smith ('31).

The physical relations between the protoplasts of host and pathogen appears to be close and intimate in light of Kunkel's and Melhus' observations. No antagonism is exhibited, and the two blend into each other in such a way that it is often impossible to determine clearly where one ends and the other begins. In fixed and stained preparations, on the other hand, the parasite stains more intensely with Congo red and Orange G than the host protoplasm (Massee and Kunkel). In the initial developmental stages there seems to be a marked attraction between the host nucleus and the amoebae, according to Melhus, *et al.* As is shown in figures 15 and 16 the latter may be crowded around the nucleus, which suggests that the

nutritional conditions are more favorable in that region of the cell.

The walls of the infected host cells are also markedly changed by the parasite. As the pseudopods of the plasmodium push down between the cells, the walls become swollen, gelatinous and wavy. These walls have a greater affinity for Orange G than those of healthy cells, according to Kunkel's and Wild's observations, which indicates that they have undergone a change in composition. The middle lamella is also usually dissolved by the action of the plasmodium.

Control

The shipment and importation of infected tubers appear to be the primary means of dispersal of powdery scab from one region or country to another. Most countries have accordingly enacted legislation against the importation of diseased potatoes and established inspection and certification bureaus within their boundaries to insure planting of healthy tubers. Locally, the disease may be transferred from one field to another by fertilizing with contaminated manure, by farm implements, contaminated sacks, and soil on the shoes of laborers. Sanitary practices must accordingly guard against dispersal by such means. Since fungus spores will survive passage through the digestive tract, infected tubers and parings should be boiled or sterilized before feeding to hogs and other animals to avoid contaminated manure. Other sanitary measures involve selection of disease-free tubers for planting and the avoidance of contaminated land.

Inasmuch as the spores of *S. subterranea* may remain viable in the soil for 3 to 5 years or longer (Melhus, *et al.*), crop rotation, fallowing, or pasturing the land are essential in regions where the disease is abundant and destructive. In such regions the potato crop may be largely destroyed if rotation is neglected (Pethybridge, '26). Dorojkin ('36) thus advocated compulsory crop rotation of no less than 3 years in Russia, but it is apparent that a longer period may be necessary to starve out the parasite. The rotation period obviously depends to some extent on climatic conditions, and the character of the soil. In Scotland, for instance, a rotation of 6 to 10 years or longer has been recommended for loamy soil in regions where high rainfall and low tempartures normally occur during the potato growing season.

Eradication of wild hosts is of doubtful value at present because *S. subterranea* has a comparatively limited host range and very little is known about its occurrence outside of the potato. Since the fungus develops on tubers only after they are partially mature early harvesting may sometimes be effectual in avoiding the disease, provided infection does not occur early in the season. However, it is not practicable because no marked above-ground symptoms of infection occur, which would indicate whether or not the tubers are infected.

Liming the soil as practiced in the control of club root of crucifers stimulates instead of inhibiting the development of powdery scab. Massee ('10, '15) recommended dressing the land with quicklime in the spring when the spores germinate in the soil, but Pethybridge, Horne ('12), and others found that lime increases the amount of diseased tubers. Melhus, *et al.*, likewise noted that lime at the rate of 3,000 lbs. per acre increased infection 13.2 per cent in one case but reduced it in another. These variations, however, were probably due to differences in soil types in the two test blocks. Phillips ('32) advised the application of lime to loosen the soil and make it more porous and thereby increase drainage. Since damp, water-logged soil favors the development of the parasite proper drainage is essential.

Other control measures involve seed tuber disinfection before planting, soil disinfection with chemicals and fungicides, and the use of resistant potato varieties. As to tuber sterilization various disinfectants have been advocated and used. Johnson ('08) found that soaking infected tubers for 18 to 24 hours in 2 per cent Bordeaux mixture, 1½ hours in corrosive sublimate, or 2 hours in a weak formalin solution is effective in killing the spores of *S. subterranea*. Subsequent workers have confirmed these results to some degree. Pethybridge ('13) in particular observed that seed tubers treated with weak solutions of formalin, copper sulphate, Burgundy mixture, or rolled in flowers of sulphur checked the disease to a marked degree. Tubers soaked in 1 per cent copper sulphate for 3 hours yielded no diseased offspring, while those rolled in sulphur gave only 1.03 per cent infection. Melhus, *et al.*, however, found that tuber treatment with 2 pts. of formalin per 30 gals. water at 46 to 50° C. for 5 minutes, or mercuric chloride, 4 ozs. to 15 gals. water at 44 to 45° C. for 5 minutes, gave better results than the usual long cold treatments with either of these substances. Rolling wet tubers in sulphur or soaking them in 5 per cent atomic sulphur for 1½ hours was less effective than treatment with formaldehyde and mercuric chloride. While these disinfectants reduce infection considerably, the results obtained are not to be regarded as absolute, according to Melhus, *et al.*, since other factors such as variations in soil moisture and texture, drainage, and temperature have a marked effect on the results. Later workers, including Abbott ('28), Dorojkin ('34, '36) and Rovdo ('36), have reported similar beneficial results from the use of corrosive sublimate, formalin, and mercuric chloride on infected seed tubers. Dorojkin found that soaking tubers 20 to 30 minutes in a .2 per cent solution of meranin, a liquid organic mercury preparation containing less mercury than mercuric chloride, gave excellent control in Russia.

Disinfection of contaminated fields by the application of sulphur at the rate of 300 to 900 lbs. per acre has been reported by Melhus, *et al.*, Cotton ('22), Abbott, and Böning and Wallner ('38) to reduce the incidence of infection considerably. Melhus and his collaborators reported that better results

may be secured with broadcast sulphur than when it is applied in drills. Cotton ('22) found that the addition of 600 lbs. per acre reduced the incidence of infection from 54 to 7.5 per cent, while Böning and Wallner ('38) reported that the incorporation of sulphur at the rate of 400 klg. per hectare with ordinary fertilizers diminished infection of the Parnassia variety from 24 to 17 per cent.

The addition of certain fertilizer ingredients to the soil has also been reported to reduce infection. Pethybridge noted very early that the application of superphosphate and sulphate of potash reduced the number of diseased tubers, and later this aspect of control was investigated more thoroughly by Melhus and his collaborators in Maine. Each of fifteen plots of soil of varying composition and texture were fertilized separately with sodium nitrate, old horse manure, new horse manure, phosphoric acid, ammonium sulphate and phosphoric acid, ammonium sulphate and potassium chloride respectively and tested against 7 controls treated with commercial fertilizers at the rate of 1,500 lbs. per acre and 7 untreated controls. These plots were then seeded with Green Mountain variety tubers which had been disinfected with the usual strength of mercuric chloride. All of the fertilizer ingredients tested alone reduced infection 5 to 12 per cent below that of the controls. Ammonium sulphate and acid phosphate gave nearly the same yields as the fertilized checks and diminished infection 7.6 per cent, while potassium chloride yielded the least infection, which may have been partly due to the prolonged growing season in this case.

So far no completely resistant and immune potato varieties have been found or developed. Although reports of partial to complete varietal resistance have often been made, it is not certain whether the reported resistance is due to inherent immunity or to the fact that the plants escaped infection. Soil composition and texture as well as the number of spores present doubtless vary considerably, and it is not improbable that these factors are often the cause of variations in infection. This is probably true in the case of Melhus' experiments conducted in 1915 in Maine. Melhus and his co-workers found four named varieties (Eldorado, Farys, Woiltmann, and Senator) and seven seedlings the tubers of which were free of the disease, while others showed very slight to severe infection. However, inasmuch as the control variety, Green Mountain, also showed wide fluctuations in degree of infection, Melhus, *et al.*, concluded that the variations in varietal response were partly due to the fact that some of the varieties escaped infection. They further believed that the nature of varietal resistance probably relates to the ability to form cork cambium. Gomolyako ('30) reported that Svitez, Deodora, Pirola, Rubia, Parnassia, Gavronec k and Jubel were least affected by powdery scab in Russia, but he was not certain that they are consistently resistant. Naumov likewise reported these varieties to be partially resistant, while only one, Rose of Milet, remained free of infection in 1935. Dorojkin ('36) found no completely resistant commercial varieties but regarded Jubel, Cobbler, and Parnassia as weakly susceptible. However, eight varieties of *Solanum* from South America proved to be immune, as well as nine hybrids developed by the Pan Soviet Institute of Plant Breeding. Berogovoy ('39) reported that none of the varieties tested at the Kief quarantine laboratory were completely resistant, although the degree of infection was quite low. The variety Woiltmann showed the highest degree of resistance and was recommended for sub-sandy soil.

It is obvious from these reports that varieties such as Parnassia, Woiltmann, Svitez, Jubel, Pirola, and Cobbler possess some degree of resistance to powdery scab. The use of these varieties in connection with other control measures such as proper soil drainage, tuber sterilization, soil disinfection, etc., will doubtless do much to alleviate infection with powdery scab where it is particularly abundant and destructive.

Geographical Distribution of Powdery Scab and Bibliography of Literature

ALGERIA

Anony. 1923. Bull. Agric. Algerie-Maroc. 2nd ser. 29: 69.

Cujoutantis, A. 1924. Rev. Path. Veget. Entom. Agric. 11: 60.

ARMENIA

Rovdo, A. S. 1936. White Russ. Acad. Sci. Inst. Biol. Sci. Minsk 1936: 39.

AUSTRALIA

Anony. 1927. Jour. Dept. Agric. Victoria 25: 613. 1936, *Ibid.* 34: 464. 1938, *Ibid.* 36: 301.

Bald, J. G. 1941. Pamphl. Com. Sci. Ind. Res. Australia 106.

Darnell-Smith, G. P. Ann. Rept. Dept. Agric. New South Wales. 1920–'21: 27.

Norwood, R. B. 1933. Queensland Agric. Jour. 40: 382.

Noble, R. J. 1924. Agric. Gaz. New South Wales 35: 883.

AUSTRIA

Hecke, L. 1923. Wiener Landw. Zeit. 73: 273, 281.

Janchen, E. 1921. Oesterr. Zeitschr. f. Kartoffelbau 1: 3.

Köck, G. 1922. Wiener Landw. Zeit. 72: 82.

———. 1927. Oesterr. Zeitschr. f. Kartoffelbau 1927, no. 3.

Wahl, B. 1924. Zeitschr. Landw. Versuchsw. Deut-Oesterr. 1924: 1–8.

CANADA AND MARITIME PROVINCES

Dickson, B. T. 1922. 14th Ann. Rept. Quebec. Soc. Protect. Plants, p. 67.

Eastham, J. W. 1914. Canada Agric. Exp. Farms Circ. no. 5: 7.
———. 1922. 16th Ann. Rept. Dept. Agric. British Columbia 1921: 64: 69.
Gorham, R. P. 1914. Dept. Agric. New Brunswick Hort. Div. Leaf. III.
Güssow, H. T. 1913. Phytopath. 3: 18.
Henry, A. W. 1934. Circ. Coll. Agric. Alberta, no. 15.
Hurst, R. R. 1926. Rept. Dominion Botanist, Div. Bot. Canada Dept. Agric. 1925: 20. 1929, *Ibid.* 1928: 165.
Ledingham, G. A. 1934. Nature 133: 534. 1935, *Ibid.* 135: 394.
Partridge, G. 1923. Agric. Gaz. Canada 10: 121.
Sanford, G. B. 1924. Alberta Agric. Coll. Bull. no. 5.
Tice, C. 1922. Scient. Agric. Canada 2: 249.
Tucker, J. 1921. Canada Dept. Agr. Pamph. no. 129. 1927, *Ibid.* Pamph. no. 84.

COLOMBIA

Franco, R. M. 1938. Agric. Bogota 10: 344.

CZECHOSLOVAKIA

Anony. 1930. Ochr. Rost. 10: 1.
Blattný, C. 1935. Rec. Inst. Rech. Agron. Rép. tchécosl. 137: 21.

DENMARK AND FAROE ISLANDS

Ferdinandsen, C. 1923. Tidsskr. f. Landøkonomi.
Gram, E., and S. Rostrup. 1925. Tidsskr. f. Planteavl. 31: 353.
———, and M. Thomsen. 1927. *Ibid.* 33: 84.
Rostrup, E. 1905. *Ibid.* 12: 352.
Weber, A. 1922. Tomatsygdomme. Copenhagen.
Winge, O. 1913. Ark. f. Bot. 12, no. 9: 26.

ENGLAND AND WALES

Anony. 1909. Jour. Bd. Agric. London 15: 749.
Berkeley, M. J. 1846. Jour. Hort. Soc. London 1: 33.
———. 1850. Ann. Mag. Nat. Hist. 2nd Ser. 5: 464.
Cotton, A. D. 1922. Min. Agric. Miscl. Pub. 38.
Cooke, M. C. 1903. Jour. Roy. Hort. Soc. 27: 801.
Cook, W. R. I. 1933a. Arch. Protistenk. 80: 179.
———. 1933b. Glamorgan County Hist. Nat. Hist. 1: 213.
Horne, A. S. 1912. Jour. Roy. Hort. Soc. 37: 362.
———. 1930. Ann. Bot. 44: 199.
Massee, G. 1908. Jour. Bd. Agric. England 15: 594.
———. 1909. Proc. Linn. Soc. London 1909: 6.
———. 1910. Diseases of cultivated plants and trees. 1st ed. New York; 2nd ed. 1915.
Millard, W. A., and S. Burr. 1923. Gard. Chron. 72: 255.
Ministry Agric. and Fish. 1934. Bull. 79.
Osborn, T. G. B. 1911. Ann. Bot. 25: 271, 327.
Pethybridge, G. H. 1913. Jour. Roy. Hort. Soc. London 38: 524.
———. 1926. Min. Agric. Miscl. Pub. no. 52.
———. 1927. Nat. Farmer's Union Year Book 1927: 162.
Potter, M. C. 1908. Jour. New Castle Farmer's Club 1908.
Preston, N. C. 1928. Rept. Advis. Dept. Harper Adams Agric. Coll. Newport Salop. 3: 4.
Robertson, I. M., and A. M. Smith. 1931. Biochem. Jour. 25: 763.
Schwartz, E. J. 1914. Ann. Bot. 28: 227.

FRANCE

Chauzit, J. 1923. Prog. Agric. et Vitic. 40: 63.

GERMANY

Appel, O. 1918. Landw. Hefte 35: 1.
Behrens, J. 1915. Jahrb. Deut. Landw. Ges. 30: 48.
Bartling, E. Versammelung Deut. Nat. und Aerzte zu Braunschweig im September 1841. Vieweg und Sohn, 1842.
Böning, K., and F. Wallner. 1938. Prakt. Bl. Pflanzenb. 15: 268.
Föcke, E. 1846. Die Krankheit der Kartoffeln in Jahre 1845. Bremen.
Frank, E., and C. Krieger. 1896. Zeitschr. f. Spiritusind. 1896.
Fulmek, L., and A. Stift. 1917. Centralbl. Bakt. Parasitenk. II. 47: 545. 1920a, *Ibid.* 51: 97. 1920b, *Ibid.* 51: 315. 1920c, *Ibid.* 52: 81.
Koltermann, A. 1931. Fortschr. d. Landw. 6: 292.
Korff, E., and K. Böning. 1927. Prakt. Bl. Pflanzenb. u. Pflanzensch. 5: 192.
Martius, C. F. P. 1842a. Die Kartoffelepidemie. München.
———. 1842b. C. R. Acad. Sci. Paris 15: 314.
Phillips, W. 1932. Die Kranke Pflanze 9: 111.
Schlumberger, O. 1915. Deut. Landw. Presse 41: 369.
———. 1927. Mitt. Deut. Landw.-Gesell. 42: 637. 1933, *Ibid.* 48: 317.
Schneider, G., O. Schlumberger, and K. Snell. 1930. Mitt. a. d. Biol. Reichsanst. 38: 84.
Stift, A. 1912. Centralbl. Bakt. Parasitenk. II. 33: 447. 1916, *Ibid.* 45: 305.
Stranak, F. 1918. *Ibid.* 48: 520.
Wallroth, F. W. 1842a. Linnaea 16: 332.
———. 1842b. Beitr. zur Bot. 1: 118.
Wehmer, C. 1896. Centralbl. Bakt. Parasitenk II. 2: 261.
Wollenweber, H. W. 1926. Mitt. Gesell. Vorrats. 2: 32.

HAWAII

Carpenter, C. W. 1920. Hawaii Agric. Exp. Sta. Bull. 45: 1–42.
Lyon, H. L. 1936. Rept. Hawaii Sugar Exp. Sta. 1935: 26.

HOLLAND

Quanjer, H. M. 1916. Med. R. H. L.-, T.-en B. School. Wageningen 9: 94.
Van Poeteren, N. 1926. Versl. en Meded. Plantenz. Dienst Wageningen 44.
Ziekten van Cardappelknollen. 1909. Meded. Phytopath. Dienst te Wageningen no. 9.

INDIA

Galloway, L. D. 1936. Sci. Rept. Res. Inst. Pusa 1934–1935: 120.
Maun, H. H., *et al.* 1921. Dept. Agric. Bombay Bull. 102.

IRELAND

Johnson, T. 1906. Jahresb. Verein. Vererts. Angew. Bot. 4.
———. 1907. Econ. Proc. Roy. Dublin Soc. 1: 345. 1908. *Ibid.* 1: 453.
———. 1909. Scient. Proc. Roy. Dublin Soc. n. s. 12: 165.
Pethybridge, G. H. 1909. Irish Nat. 18: 118.
———. 1910. Jour. Dept. Agric. Tech. Instr. Ireland 10: 241. 1911. *Ibid.* 11: 417. 1912. *Ibid.* 12: 334. 1913. *Ibid.* 13: 160. 1918. *Ibid.* 18: 410.

ITALY

Catoni, G. 1931. Trento. Fitographia Edit. Mutilati Invol.
Perotti, R. 1940. Biologia vegetale applicata all' agricoltura. III. Micologia-malatti parassitarie. Turin.

KENYA, AFRICA

Gillett, S., J. McDonald, and T. J. Anderson. 1931. Kenya Dept. Agric. Bull. No. 10 of 1931.
McDonald, J. 1928. Ann. Rept. Dept. Agric. Kenya 1928: 225.

MADAGASCAR

Bouriquet. 1938. Int. Bull. Pl. Protect. 12: 191.

MALTA

Borg, P. 1927. Append. F. Repts. Depts. Malta 1925–1926.

MAURITIUS

Shepherd, E. F. S. 1935. Rept. Dept. Agric. Mauritius 1931: 19.

MOROCCO

Bouhelier, R. 1936. Fruits Primeurs 6: 213.

NEW ZEALAND

Anony. 1927. N. Z. Jour. Sci. Tech. 9: 44.
Blair, I. D. 1937. Bull. Canterbury Agric. Coll. Lincoln 94.
Cockayne, A. H. 1921. N. Z. Jour. Agric. 21: 169.

NORWAY

Branchorst, J. 1887. Bergens Mus. Aarsberet. 1886: 225.
Jorstad, I. 1932. Landbruksd. Beretning Tellegg C.
Lunden, A. P. 1938. Meld. Norge Landbr. Hoisk. 18: 183.

PERU

Abbott, E. V. 1928. Estac. Exp. Agric. Soc. Nac. Agrar. Lima Circ. 7.
———. 1929. Phytopath. 19: 645. 1931. *Ibid.* 21: 1064.
Lagerheim, G. 1891. Jour. Mycol. 7: 103.
Patouillard, N., and G. Lagerheim. 1891. Bull. Soc. Mycol. France 7: 158.

POLAND

Siemaszko, W. 1929. Centralbl. Bakt. Parasitenk II. 78: 113.

PORTUGAL

Gonzales, de Andres, C. 1930. Junta Admin. Servicios. Agric. Divul. no. 3: 1–46.

RHODESIA

Hopkins, J. C. F. 1940. Rhod. Agr. Jour. 37: 411.

SCOTLAND

Anony. 1927. Bd. Agric. Scotland Miscl. Pub. no. 3.
Cuthbertson, D. C. 1925. Jour. Roy. Hort. Soc. 1: 21.

SOUTH AFRICA

Doidge, E. M. 1924. Bot. Survey S. Africa Mem. no. 6: 1–56.
Pole-Evans, J. B. 1910. Transvaal Agric. Jour. 8: 162.
———. 1910. Transvaal Dept. Agric. Farm Bull. 110.

SWEDEN

Henning, E. 1915. Tradgarder. 1915. no. 3.
———. 1922. Kungl. Landb.-Akad. Handl. Tidskr. 1922: 26.
———, and T. Lindfors. 1921. De viktigare potatissjukdomarna. 1921.

SWITZERLAND

Anony. 1920. Schweiz. Landw. Zeitschr. 1920: 7.
Wild, N. 1929. Phytopath. Zeitschr. 1: 367.

TASMANIA

Darnell-Smith, G. P. 1922. Ann. Rept. Dept. Agric. New South Wales 1920–21. 1922.

U. S. A.

Anony. 1924. Pl. Disease Rept. Suppl. 34: 168, 178, 183.
Bailey, F. D. 1915. Sci. n. s. 42: 424.
Barrus, M. F., and C. Chupp. 1926. Cornell Exp. Sta. Bull. 135.
Brigham, E. S. 1914. Vermont Dept. Agric. Bull. 18: 2.
Clinton, G. P. 1915. Rept. Conn. Agric. Exp. Sta. Bull. 1915: 463.
Cook, M. T., and G. W. Martin. 1914. New Jersey Agric. Exp. Sta. Circ. 33: 3.
Haskell, R. J., and J. I. Wood. 1927. Pl. Dis. Reporter Suppl. 54: 209.

Kunkel, L. O. 1915. Jour. Agric. Res. 4: 265.
Link, G. K. K., and G. B. Ramsey. U. S. Dept. Agric. Misc. Pub. 98.
Lutman, B. F., and G. C. Cunningham. 1914. Vermont Agric. Exp. Sta. Bull. 184.
Lyman, G. R., and J. T. Rogers. 1915. Sci. n. s. 42: 940.
McCubbin, W. A., R. E. Hartman, and K. M. Lauer. 1926. Penn. Dept. Agric. Bull. 420.
Melhus, I. E. 1913. Sci. n. s. 38: 132.
———. 1914. U. S. Dept. Agric. Bull. 82.
———, J. Rosenbaum, and E. S. Schultz. 1916. Jour. Agric. Res. 7: 213.
Morse, W. J. 1913. Sci. n. s. 38: 61.
———. 1914. Maine Agric. Exp. Station Bull. 227.
Orton, W. A. 1913. U. S. Dept. Agric. Farmer's Bull. 544.
Ramsey, G. B. 1918. Phytopath. 8: 29.
Sands, H. C., and G. G. Atwood. 1914. New York State Dept. Agric. Circ. 111.
Shapovalov, M. 1923. Jour. Agric. Res. 23: 285.
———, and G. K. K. Link. 1924. U. S. Dept. Agric. Farmer's Bull. 1367.
Shear, C. L. 1914. Phytopath. 4: 36.

U. S. S. R.

Beregovoy, P. 1939. Pl. Protect. Leningrad 1939: 163.
Djelaloff, R. 1933. Azerbaijan Agric. Inst. Pamph. 1933.
Dorojkin, N. D. 1934. Crop. Protect. Moscow 1934: 11.
———. 1936. White Russ. Acad. Sci. Inst. Biol. Sci. Minsk 1936: 5.
Gomolyako, N. J. 1930. Morbi Plant. 19: 79.
Khrobrykh, N. D. 1938. Summ. Sci. Res. Inst. Pl. Protect for the year 1936. 1938: 27.
Kiyanovski, P. M. 1936. White Russ. Acad. Sci. Inst. Biol. Sci. Minsk 1936: 39.
Mercklin, E. 1856. Bull. Soc. Nat. Moscow 29 pt. 2: 301.
Naumov, N. A. 1936. Summ. Sci. Res. WK. Inst. Pl. Protect. Leningrad 1935: 520.
Roydo, A. S. 1936. White Russia Acad. Sci. Inst. Biol. Sci. Minsk 1936: 87.
Rybakova, S., and H. Nedoshivina. 1936. *Ibid.* 1936: 67.
Speschnew, N. N. 1897. Arb. Tiflis Bot. Gart. Lief. 11–199.

SPECIES INDEX

Anisomyxa, 68
Plantaginis, 70
Clathrosorus, 54
Campanulae, 56
Cystospora, 76
Batata, 76
Ligniera, 58
Isoetes, 62
Junci, 60
pilorum, 62
radicalis, 60
vascularum, 63
verrucosa, 62
Membranosorus, 52
Heterantherae, 52
Molliardia, 37
Triglochinis, 37
Octomyxa, 40
Ichlae, 10
Ostenfeldiella, 22, 32
Diplantherae, 32
Peltomyces, 76
hyalinus, 76
Blatellae, 76
Forficulae, 76
Plasmodiophora, 22
Alni, 34
Brassicae, 22
bicaudata, 33
californiae, 34
Diplantherae, 32
Elaeagni, 34
Fici-repentis, 32
Halophilae, 32
Humuli, 35
Orchidis, 35
Solani, 36
tabaci, 35
tomati, 35
vascularum, 35
Vitis, 34
Polymyxa, 63
graminis, 64
Pyrrhosorus, 71
marinus, 71
Rhizomyxa, 64
hypogeae, 64
Sorodiscus, 46
Callitrichis, 47
Heterantherae, 52
karlingii, 50
radicicolus, 18
Sorolpidium, 66
Betae, 66
Sorosphaera, 41
radicalis, 43
Veronicae, 42
Spongospora, 54
Campanulae, 58
Cotulae, 58
scabies, 57
Solani, 57
subterranea, 57
subterranea var. *radicicola*, 57
subterranea var. *tubericola*, 57
Sporomyxa, 74
Scauri, 74
Tenebronis, 75
Tetramyxa, 37
Elaeagni, 38
parasitica, 37
Triglochinis, 37
Trematophlyctis, 70
Leptodesmiae, 70

SUBJECT INDEX

Ichlya, 10
Acrasiae, 57
Acrocystis, 77
Actinomyces, 77
Agropyron, 64
Agrostis, 66
Aira, 66
Akaryote stage, 2, 10, 11, 12, 20, 81
Alisma, 60
Allomyces, 10
Alnus, 31, 91
Alternation of generations, 13, 82, 84
Amaurochaete, 84
Ammonium sulphate, 115, 133
Amoeba, 8, 9, 79
Amoebosporus, 35
Amylophagus, 88, 89, 90
Anagallis, 66
Aphanomyces, 10
Aphelidiopsis, 88, 90
Apodachlya, 10
Archimycetes, 78
Arcella, 11, 79
Asterocystis, 67, 78
Bacteria, 26
Badhamia, 84
Bartsia, 66
Battatis, 76
Basic fertilizers, 115
Bellis, 60
Beta, 62, 66
Binuclearity hypothesis, 2, 11
Biological races, 26
Biscutella, 66
Blatellae, 76
Blepharoplast, 2, 80, 81
Bodo, 90
Borrago, 66
Bordeaux mixture, 109, 110
Brassican, 109
Briza, 66
Bromus, 62
Calendula, 66
Callitriche, 47, 48, 60
Campanula, 58, 66
Cancer, 28
Capillitia, 2, 81
Capsella, 66
Carbolineum, 105
Carbolic acid, 109
Catabrosa, 43
Caryotropa, 8
Centrosomes, 9, 11
Cerastium, 60
Ceratiomyxa, 18, 80
Ceratium, 34
Ceresan, 109
Chara, 50
Chenopodium, 62
Cheshunt brown, 105, 110
Chloropicrin, 105
Chromidia, 2, 10, 11
Chromidia hypothesis, 2, 11
Chromosome numbers, 14
Chrysanthemum, 60
Chytridiales, 78
Cleavage, 14
Clubicide, 105, 110
Club root, 93
Comatrichia, 80
Copper oxide, 110
Copper carbonate, 110
Copper sulphate, 110, 132
Corrosive sublimate, 105, 132
Cotula, 58
Cresol, 109

Crop rotation, 117, 132
Cruciform divisions, 2, 8
Cucurbita, 91
Cystoclonium, 71
Cystosorus, 2
Degree of infection, 99-103
Delphinium 66
Dictyaethalium, 80
Dictyostelium, 57, 80
Didymium, 80, 88
Dinaria, 66
Diplanthera, 32
Disease control, 104, 132
Double anchor stage, 2, 8
Economic losses, 93, 129
Elaeagnus, 34, 38
Equatorial ring stage, 7, 8, 9, 10
Erigeron, 66
Erionema, 84
Erysibe, 57
Euglena, 8
Eucarpy, 2
Fedia, 66
Festuca, 62
Ficus, 32
Flagellata, 78
Folosan, 109
Forficula, 76
Formalin, 105, 132
Frankia, 34
Frankiella, 34
Fuelgen's nuclear reaction, 9
Fuligo, 80, 88
Fusarium, 129
Garland stage, 12
Germisan, 109
Glucocochlearin, 120
Gluconasturtin, 120
Glucotrapaeolin, 120
Gymnococcus, 78, 79, 88, 90
Gymnococcaceae, 78, 88
Gynandropsis, 48
Gypsum, 115
Halophila, 32
Haplomonoecious, 3, 82
Haplophenotypic, 82
Haplosynoecious, 3, 82
Helianthus, 91
Heterocont, 3
Heteranthera, 52, 53
Heterothallic, 3, 84
H-ion concentration, 97, 98, 129
Hippophae, 34
Holocarpy, 3
Homothallic, 3, 84
Hordeum, 64
Hosts, 99-103
Humulus, 35
Hydromyxaceae, 88
Hyperplasy, 3
Hypertrophy, 3
Hypoplasy, 3
Idiochromatin, 3, 6
Immunity, 118, 133
Iris, 60
Isocont, 3, 71
Isoetes, 63
Isogamy, 3, 81
Isomorphic, 3, 81, 84
Juncus, 59, 60, 66
Karyogamy, 3, 15, 16, 18, 82
Karyosome, 5, 8
Kataplasmic, 94, 130
"Krankheitsherde," 55, 94, 131
Lagenidium, 63
Lamium, 66
Leptodesmia, 70, 71
Leptomyxa, 35, 77
Leptothrix, 34
Liming, 110-115
Lotus, 66
Lycogola, 80
Lycopersicon, 57
Magnesium carbonate, 115
Medicago, 66
Meiosis, 12, 16, 83, 84
Mentha, 60
Mercuric chloride, 105, 106, 109, 132
Meront, 3, 14
Methyl green, 105
Monadineae, 78, 88
Monas, 88
Monilia, 43
Mustard oil, 109, 120
Mycetozoidia, 88
Myxoidea, 79
Myxomycetes, 78, 79, 82, 83
Myxosporidia, 90
Myxochytridiales, 78
Myzocytium, 63, 66
Naegleria, 36
Nematodes, 96
Newton's gentian violet method, 9
Nuclear dimorphism, 4
Olpidiaceae, 85
Olpidium, 23, 66, 96
Olpidiopsis, 85
Oospora, 131
Papulospora, 131
Parachlorobenzine, 109
Petroleum, 109
Phaseolus, 91
Phoma, 131
Physarella, 80, 88
Physarium, 80
Phytomyxa, 78, 79
Phytomyxaceae, 2, 78, 79
Phytomyxini, 2, 78, 79
Phytophthora, 131
Planocyte, 3, 80
Plantago, 60, 70
Plasmodiocarp, 3, 80
Plasmodium, 3, 80, 81
Plasmogamy, 3, 15, 16, 18, 82
Poa, 43, 60, 62, 66
Polygonum, 60
Polysphondylium, 57
Potassium permanganate, 109
Potomogeton, 37, 60
Powdery scab, 129
Powdery scab dry rot, 129, 130
Preplasmodium, 25
Pringsheimella, 85
Promitosis, 3, 4, 10, 11
Protomitosis, 3, 5
Proteomyxa, 35, 79, 88
Protista, 78
Protomyces, 57
Protomyxidea, 79
Protozoa, 79, 88
Pseudocommis, 34
Pseudospora, 79
Pseudosporopsis, 88
Quarantine, 129, 133
Radium, 110
Ranunculus, 60, 66
Resistance, 118, 119, 120, 133
Reticularia, 84
Rhizoctonia, 131
Rhizopoda, 90
Rhizosporium, 57
Rozella, 85
Rozellopsis, 86
Rumex, 64
Ruppia, 37
Saccharum, 35
Sappinia, 76
Saprolegnia, 40
Sarcodina, 88
Saltpeter, 116
Saturn stage, 3
Scaurus, 74
Schinzia, 34
Schizont, 3, 14
Schizogony, 3, 14, 15, 90
Schizozooites, 90
Sclerotia, 25, 29
Scolochloa, 64
Secale, 64
Segetan, 109
Semesan, 109
Septolpidium, 63
Serumsporidium, 79
Setaria, 66
Sexuality, 15, 81, 82
Sileue, 66
Sinablin, 120
Sinigrin, 120
Sodium carbonate, 110
Sodium chloride, 110
Soil drainage, 98, 117, 132
Soil rot, 77
Soil moisture, 98
Soil pox, 77

Solanum, 36, 57, 91, 134
Solibar, 105
Sorocarp, 3
Sorosporium, 42, 57
Sorus, 1, 3
Soot, 109
Sporangiosorus, 1, 3
Spore germination, 97, 129
Spore longevity, 104, 132
Sporocyst, 3, 90
Sporogametes, 25
Sporogenesis, 10
Sporont, 3, 14, 15
Sporozoa, 79, 90
Stellaria, 66
Stemonitis, 80, 84
Sulcan, 109
Sulgine, 109
Sulphur, 105, 132, 133
Symbiosis, 28, 38
Synchytrium, 71, 78, 130
Synchytriaceae, 70, 78, 85
Synkaryon, 3
Temperature effects, 98, 129
Tenebrio, 76
Tillantin B, 110
Trichia, 18, 80
Trifolium, 66
Triglochin, 37, 66
Triticum, 64
Trophochromatin, 6
Trypanosoma, 8
Tuberculina, 42, 57
Tylogonus, 2
Ulmus, 57
Uspulun, 105, 109
Vampyrella, 79
Vaucheria, 86
Veronica, 42, 60, 62
Vitis, 34
Woronina, 10, 78, 85, 86, 91
Woroninaceae, 85, 86, 91
X-ray, 110
Zannichella, 37
Zoocyst, 3, 90
Zosteria, 33

AUTHOR INDEX

Abbay, 93, 110, 111, 127
Abbey, 35
Abbott, 129, 132, 135
Abe, 81, 82
Adam, 93, 121
Adams, 77
Albert, 111, 120
Alexieff, 4, 9
Almeida, 126
Anderson, 93, 110, 117, 127
Andreucci, 32
Appel, 26, 94, 95, 106, 110, 115
Archer, 134
Arker, 106, 111, 116, 118, 123
Atkins, 97, 99, 125
Atkinson, 96, 127
Atwood, 136
Aue, 123
Bailey, 127, 135
Bailie, 109, 125
Bald, 133
Bärner, 36
Barnes, 2, 79
Barre, 77
Barrett, 18, 13, 58, 59, 66
Barrus, 135
Bartling, 58, 134
Baudys, 121
Bayer, 123
Beaumont, 97, 110, 119, 121
Becker, 111, 123
Behla, 30, 123
Behrens, 34
Belar, 91
Benehley, 105
Bennett, 111, 121
Beregovy, 131, 133, 136
Berkeley, 57, 121, 129, 134
Bernatsky, 125
Bessey, 79, 80, 81
Beyer, 96, 111, 123
Biffin, 121
Bird, 127
Birne, 93, 127
Bjorkenheim, 34
Blair, 135
Blattny, 57, 134
Blomfield, 4, 10
Blumberg, 123
Blunek, 97, 105, 110, 111, 116, 118, 123
Böhner, 93, 111, 123
Böning, 93, 132, 133, 134
Bondarzew, 126
Borg, 135
Borzi, 1, 64, 66
Bos, 34, 98, 110, 135
Bottner, 123
Bouhlier, 135
Bouriquet, 135
Braun, 106
Brehmer, 36
Bremer, 96, 97, 98, 105, 109, 110, 123
Brezhnew, 115, 119, 126
Brick, 106, 110, 123
Brigham, 135
Briosi, 34, 125
Brönnle, 106, 123
Brown, 109, 111, 121
Bruck, 123
Brunchorst, 2, 34, 54, 57, 93, 135
Bubac, 121
Buchman, 93, 121
Burk, 20, 22, 42, 47
Burkhardt, 99, 106, 109, 111, 117, 123
Burr, 131, 134
Busse, 34
Cadman, 1, 79, 80, 82, 84
Calkins, 8, 79
Carpenter, 134
Carricklee, 111, 112, 121
Carruthers, 96, 121
Casali, 34
Caspary, 94, 123
Catoni, 135

Cavara, 34
Cavers, 78
Chardon, 35
Chatton, 4
Chauzit, 134
Chodat, 34
Christensen, 97, 121
Chupp, 16, 23, 24, 28, 96, 98, 105, 110, 112
Cienkowski, 1, 88
Clark, 121
Clayley, 82
Clayton, 105, 109, 127
Clinton, 128, 135
Cockayne, 135
Cöic, 104
Collinge, 121
Cook, M. T., 35, 63, 94, 128, 135
Cook, W. R. I., 1, 10, 16, 18, 22, 37, 42, 48, 50, 54, 59, 67, 81, 85, 86, 91, 134
Cooke, 34, 121, 134
Coons, 128
Cotner, 80
Couch, 1, 4, 18, 40, 81, 85
Crosby, 128
Cotton, 132, 134
Cuboni, 34
Cujoutantis, 133
Cunningham, 94, 97, 99, 110, 115, 128
Curtis, 93, 121
Cuthbertson, 135
Dankler, 106, 123
Darnell-Smith, 105, 110, 133, 135
Davies, 129
Davis, 119, 128
De Andres, 117, 127
DeBary, 78, 80
Debray, 34
De Bruyne, 88, 90
Delage, 2, 79
Deutelmoser, 116, 123
Diakonoff, 110, 126
Dickson, 133
Diedicke, 42
Djelaloff, 136
Doflein, 14, 79, 91
Dodge, 78
Doidge, 48, 127, 135
Donald, 42
Doppler, 127
Dorojkin, 129, 132, 133, 136
Ducomet, 34
Dufrenoy, 121
Duggar, 77, 128
Eastham, 134
Edison, 128
Edson, 93
Eggemeyer, 106, 109, 123
Elenkin, 126
Elliott, 76, 77
Ellis, 93, 121
Ericksson, 96, 115
Esmarch, 3, 24, 104, 109, 123
Evans, 119, 129
Eycleshymer, 1, 24, 93, 95, 117, 128
Farquharson, 93, 127
Farsky, 116, 121
Favorsky, 23, 80, 96, 126
Fedetova, 28, 126
Fedorintschik, 1, 10, 16, 23, 24, 81, 96, 104, 117
Feinberg, 30, 123
Feldman, 32, 33
Felsberg, 123
Ferdinandsen, 5, 32, 54, 57, 58
Findlay, 119, 127
Fischer, 78, 85, 127
Fitzpatrick, 2, 3, 66, 70, 77, 79
Flachs, 106, 116, 123
Fleishman, 123
Föcke, 134
Fowlie, 127
Franco, 134
Frank, 34, 123, 134
Fron, 62
Fruwirth, 123
Fulmek, 134
Gaillat, 62
Galloway, 134
Gäumann, 78
Gay, 121
Gaylord, 23, 24, 30, 128
Georgeson, 93, 110
Gibbs, 26, 97, 104, 106, 112, 115, 116, 117, 118
Gilbert, 80, 82, 128
Gilchrist, 110, 121
Gillett, 135
Gilman, 128
Glasgow, 106, 110, 128
Gleisberg, 26, 93, 96, 105, 111, 123
Gloyer, 106, 110, 128
Goebel, 2, 37
Gomolyako, 133, 136
Gonzales, 135
Gorini, 32
Gorham, 134
Gram, 121, 134
Gravis, 35
Gretschel, 123
Grevillius, 42
Griffith, 119, 129
Griffon, 103
Grigon, 103
Gross-Schlaeters, 123
Güssow, 119, 121, 134
Guyot, 62, 66, 67
Gwynne-Vaughan, 2, 79
Habernall, 123
Haddon, 37
Haensler, 111, 112, 128
Hall, 99, 121
Halstead, 1, 77, 93, 99, 110, 128
Hammarlund, 106, 109, 127
Hanley, 105
Harder, 97
Harper, 82, 92

Harshberger, 128
Harter, 77, 110, 119, 128
Hartman, 136
Hartog, 79
Haskell, 93, 135
Hawk, 111, 128
Hayunga, 98, 116, 123
Heald, 128
Hecke, 133
Hedlund, 127
Hegyi, 125
Heinzelmann, 117, 127
Hellman, 123
Henekel, 80, 126
Henderson, 93, 110, 128
Hendrick, 119, 127
Hennings, 42, 99, 121, 135
Henry, 134
Herouard, 79
Herpers, 93, 107, 111, 123
Hertel, 105, 109, 110, 124
Hertwig, 79
Hertzman, 127
Herviaux, 104
Heyder, 124
Hildebrand, 62, 63
Hiltner, 97, 107, 115, 116, 121
Hissinger, 37
Hockey, 119, 121
Hofferichter, 105, 124
Hoffman, 105, 124
Hollenbach, 124
Hollrung, 107, 110
Holmes-Smith, 105, 106, 121
Hopkins, 135
Honig, 23, 26, 94, 95, 96, 97, 98, 99, 104, 106, 116, 124
Hönigmann, 124
Hooker, 120
Horne, 2, 9, 13, 16, 18, 56, 81, 91, 129, 134
Höstermann, 26, 107, 111, 119, 124
Howard, 80, 84
Hulst, 128
Humphrey, 98, 128
Hunter, 110, 111, 127
Hurrle, 124
Hurst, 134
Ikeno, 125
Iwanoff, 126
Jablonowski, 125
Jahn, 1, 78, 80, 81, 82, 83, 84
Jaap, 34, 126
Jaczewski, 126
Jamlainen, 97, 99, 105, 118, 119
Janchen, 133
Janson, 98, 110
Johnson, 56, 132, 135
Jones, L. R., 104, 110, 112, 119, 128
Jones, P. M., 1, 4, 10, 15, 24, 26, 35, 80, 128
Jorgenson, 105, 109
Jorstad, 23, 93, 105, 110, 117, 135
Juel 71
Kadow, 128
Kambly, 82, 92
Kalchschmid, 124
Kappen, 124
Kareltschikoff, 47
Karling, 50, 60, 63, 85, 86
Karsten, 121
Katterfeld, 99, 110, 116, 119, 126
Keissler, 34
Kellermann, 124
Khrobyrkh, 57, 136
Kimppel, 124
Kindshoven, 99, 105, 106, 116, 124
Kirk, 35
Kirschner, 107, 111, 117, 124
Kiyanovski, 136
Klebahn, 124
Klebs, 88, 92
Kleimenov, 126
Klein, 88, 92
Klemm, 124
Kniep, 78, 84
Koblischek, 107, 115, 124
Köck, 104, 117, 124, 133
Köhne, 107, 121
Koltermann, 129, 134
Korff, 115, 124, 134
Kornauth, 120
Knorr, 124
Kränzlin, 84
Kreuzpointer, 110, 111, 124
Krieger, 134
Kronberger, 106, 116
Kruger, 124
Kudo, 79, 92
Kühn, 93, 124
Kupke, 105, 109, 124
Kunkel, 1, 18, 23, 54, 57, 80, 95, 96, 128, 129
Küster, 94, 124
Lagerheim, 42, 43, 46, 57, 135
Langenbeck, 124
Lankester, 79, 88
Larsen, 23, 96, 98, 111, 115, 128
Laubert, 93, 94, 95, 104, 111, 117
Lauer, 136
Ledingham, 1, 10, 18, 23, 56, 63, 81
Leger, 74, 76
Leines, 107, 124
Leitner, 40
Levine, 30, 95, 128
Linder, 104
Lindfors, 97, 107, 109, 110, 115, 127
Link, 35, 120, 136
Lister, 82
Littlejohn, 129
Loew, 108, 109, 112
Lohwag, 34
Lotsy, 78, 92
Lowenthal, 30, 124
Ludwig, 126
Ludwigs, 97, 104, 108, 117

Lunden, 135
Lundegårdh, 91
Lüstner, 93, 108
Lutman, 8, 15, 24, 95, 128, 136
Lyman, 136
Lyon, 134
MacDonald, 135
MacLeod, 26, 108, 110, 119, 121, 128
Maire, 2, 8, 18, 34, 37, 58, 62, 78
Magnus, 99, 124
Mangin, 116
Mano, 91
Marchand, 103
Marcell, 120
Marchionatto, 120
Maneval, 128
Manns, 77
Marshall, 121
Martin, 93, 97, 110, 112, 128, 135
Martius, 129, 134
Massee, 1, 18, 34, 35, 54, 57, 132, 134
Matz, 35
Mathieu-Sanson, 95, 111, 116
Maublanc, 103
Maun, 135
McAlpine, 111, 116
McCubbin, 136
McLarty, 121
McLennan, 35
McRostie, 121
McRue, 125
Merkenschlager, 108
Melhus, 57, 129, 130, 131, 133, 136
Mercklin, 136
Meyer, 97, 124
Middleton, 128
Milburn, 121, 128, 131
Millard, 134
Miller, 93, 116, 124
Milovidov, 5, 11, 12, 15, 121
Minden, 66
Minchen, 79, 92
Moeller, 34
Moesz, 36, 125
Moldenke, 128
Molliard, 37, 38
Montieth, 98, 110, 117, 128
Moritz, 34, 126
Morozov, 126
Morse, 136
Mortensen, 121
Motte, 26, 97, 105, 112, 116, 117
Mothes, 110, 124
Moyer, 111, 112
Müller, 96, 109
Müller-Thurgau, 95, 96, 108
Murphy, 111, 115, 116, 128
Muskett, 109
Nägler, 4, 9, 91
Nance, 128
Nattras, 129
Naumann, 104, 115, 116, 124
Naumov, 28, 96, 97, 110, 129, 133, 136
Nawaschin, 1, 4, 10, 14, 24
Nedoshivinia, 57
Neger, 104, 111, 115, 124
Neill, 126
Nemec, 1, 66, 67, 68, 70
Newodowsky, 126
Nichols, 35, 127
Nicholson, 85, 86
Nicoloff, 95, 121
Nielsen, 114, 117, 121
Noak, 111, 115
Noble, 120, 133
Noel, 121
Nötzel, 124
Norwood, 133
O'Brien, 127
Oger, 123
Ogilvie, 105, 108, 121
Olive, 82
Olsson, 118, 119, 127
Orton, 136
Osborn, 1, 5, 10, 12, 18, 56, 81, 134
Ostenfeld, 47, 52
Osterwalder, 95, 96, 108, 109, 116
Owen, 128
Palm, 18, 22, 42, 47, 62
Palmer, 108, 109, 128
Panek, 124
Pape, 94, 124
Pardy, 127
Partridge, 134
Pascher, 78, 92
Passy, 123
Patouillard, 70, 71, 135
Pavillard, 14, 16, 78, 92
Perotti, 125, 135
Petch, 121
Petersen, 52
Pethybridge, 129, 132, 133, 134, 135
Petri, 110, 125
Pettera, 117, 124
Pfeiffer, 125
Phillips, 129, 132, 134
Pichler, 121
Pinoy, 26, 28, 84
Podwyssozki, 30, 126
Poetern, 125, 134
Pollacci, 32, 125
Pole-Evans, 127, 135
Polyakoff, 104, 126
Ponkler, 116, 124
Poole, 77
Pope, 119
Popp, 105, 108, 110, 111, 115, 124
Potebnia, 126
Poter, 124
Potter, 104, 134
Potts, 103, 104, 110
Preston, 105, 108, 110, 114, 129, 134
Prillieux, 34, 123
Price, 77

Prowazek, 5, 12, 15, 24, 30, 80, 124
Pryor, 117, 119, 120, 128
Quanjer, 134
Rabbas, 105, 108, 110, 124
Rabenhorst, 57
Rainio, 121
Ramsey, 129, 136
Rangel, 127
Rath, 124
Rattke, 124
Rau, 124
Ravaz, 34
Ravn, 94, 97, 98, 110, 114, 116, 118, 121
Rawlins, 67
Rayes, 126
Read, 98
Reed, 95, 128
Regel, 124
Reichenow, 79
Reitschel, 76
Remy, 93, 124
Renard, 93, 114, 123
Rhumbler, 79, 92
Riehm, 108, 109, 110, 124
Robertson, 134
Rochlin, 23, 24, 96, 97, 118, 119, 120, 126
Roger, 115, 125, 136
Rosanoff, 47
Rosen, 82, 92
Rosenbaum, 57, 136
Rosenfeld, 48
Roth, 125
Rostrup, 42, 93, 134
Roydo, 132, 133, 136
Roze, 34
Rump, 125
Russell, 121, 128
Rybakova, 57, 136
Saccardo, 67, 77, 125
Sanderson, 127
Sands, 136
Sanford, 134
Sattler, 124
Sauvageau, 34
Schaffnit, 97, 108, 115
Schaudin, 11
Scherffel, 88, 90, 92
Schibata, 34
Schilberszky, 125
Schinz, 127
Schlodder, 125
Schlumberger, 94, 95, 105, 125, 134
Schmid, 115
Schmidt, 116, 121, 127
Schneider, 134
Schoyen, 126
Schroeter, 1, 20, 34, 41, 78
Schultz, 57, 136
Schünemann, 83, 84, 92
Schwartz, 1, 10, 13, 37, 42, 59, 60, 78, 137
Schwarze, 128
Seeloff, 116, 125
Seifert, 125
Setchell, 37
Selfensperger, 110, 115
Sharangapani, 125
Shapovalov, 131, 136
Sharples, 57
Shear, 136
Shephard, 129, 135
Shewell-Cooper, 114
Siemazko, 110, 117, 126, 135
Sinoto, 80, 92
Sitensky, 110, 121, 126
Skupienski, 1, 18, 82, 84
Slingerhand, 93, 128
Smieton, 105, 109
Smith, 26, 78, 80, 96, 128
Snell, 134
Sommer, 105, 125
Sommerville, 110
Sorauer, 93, 125
Speschnew, 126, 136
Ssacharoff, 97, 99, 118, 119, 126
Staes, 125
Staniland, 97, 110
Stefanow, 95, 121
Stephens, 121
Stevens, 78, 128
Stewart, 111, 128
Stift, 121, 134
Stosch, 80, 84
Strasburger, 82, 92
Stranak, 134
Straube, 116, 125
Strohmeyer, 125
Stubbs, 111, 117
Svec, 121, 126
Sydow, 34
Tabenhaus, 76, 77, 128
Tahara, 91, 92
Takase, 12, 13, 38
Taliaferro, 36
Tedin, 119, 127
Tennent, 110, 114, 119, 126
Terby, 1, 8, 9, 12, 81
Tessenow, 110, 111, 125
Tice, 134
Tillinghast, 128
Tison, 2, 8, 18, 34, 37, 58, 62, 78, 85
Tischler, 91
Tokunaga, 86, 92
Tompkins, 128
Townsend, 77
Trieschman, 99, 104, 108, 125
Troitzky, 126
Trotter, 42, 46
Truscott, 57
Tubeuf, 2, 78
Tucker, 134
Uzel, 121
Vanderyst, 93, 95, 98, 121
Vaughan, 110, 128
Vercier, 123

Viala, 34
Vielhauer, 108, 125
Vilkaitis, 97, 108, 109, 114, 126
Vincent, 104, 123
Voelcker, 110, 111, 121
Vladimirskaya, 108, 109, 110, 114, 126
Vogel, 114, 116
Voglino, 125
Vogt, 108
Vohman, 125
Vonwiller, 125
Vouk, 28, 80, 84, 125
Wager, 91
Wagner, 116
Wagner-Ettelbruck, 125
Wahl, 133
Wähling, 95, 108, 125
Walker, 26, 96, 98, 111, 114, 115, 119, 120, 128
Waldheim, 57
Wallner, 132, 133
Wallroth, 57, 58, 134
Webb, 8, 9, 13
Weber, 57, 127, 134
Weimer, 77
Weiss, 110, 125
Wellman, 97, 98, 105, 110, 111, 114, 117, 128
Wenyon, 14
Werham, 4, 13, 18, 52, 54
Werner, 37
Werth, 26, 94
Wescheider, 123
Wettstein, 79, 92
Whiffen, 10, 18, 24, 40, 82
Whitehead, 98, 110, 115, 116, 119, 120, 129
Wilcox, 77
Wild, 56, 129, 135
Wildemann, 66
Wilson, 1, 80, 84, 108, 110, 114, 128
Winge, 5, 10, 13, 22, 32, 46, 54, 58, 67, 71, 78, 85, 134
Winter, 42
Wisselingh, 125
Wood, 93, 116, 135
Woodman, 105, 121
Wollenweber, 134
Woronin, 1, 22, 78, 93, 95, 96, 99, 109, 126
Wright, 127
Yendo, 12, 38
Yuasa, 80, 92
Zopf, 2, 78, 79, 85, 86, 88, 92, 125