THE UNIVERSITY
OF ILLINOIS
LIBRARY
570.5
ILL
v.15, cop.2

NATURAL HISTORY

JAN 31 1951

eturn this book on or before the
atest Date stamped below. A
harge is made on all overdue
ooks.
University of Illinois Library

JUL 28 1952		
AUG 4 1952		
APR 9 1953		
APR 17 1953		
DEC 7 1954		
Oct. 9, 1969		

L161—H41

ILLINOIS BIOLOGICAL MONOGRAPHS

VOLUME XV

PUBLISHED BY THE UNIVERSITY OF ILLINOIS

URBANA, ILLINOIS

EDITORIAL COMMITTEE

JOHN THEODORE BUCHHOLZ
FRED WILBUR TANNER
HARLEY JONES VAN CLEAVE

ILL
v.15 cop.2

TABLE OF CONTENTS

No. 1. Experimental Studies on *Echinostoma revolutum* (Froelich), a Fluke from Birds and Mammals. By PAUL CHESTER BEAVER.

No. 2. A Generic Classification of the Nearctic Sawflies (Hymenoptera, Symphyta). By HERBERT H. ROSS.

No. 3. Studies on the Biology of the Crayfish *Cambarus propinquus* Girard. By WILLIAM CARL VAN DEVENTER.

No. 4. Taxonomic Studies on the Mouth Parts of Larval Anura. By RAY JANNEY NICHOLS.

Digitized by the Internet Archive
in 2011 with funding from
University of Illinois Urbana-Champaign

http://www.archive.org/details/experimentalstud15beav

ILLINOIS BIOLOGICAL MONOGRAPHS

Vol. XV No. 1

Published by the University of Illinois
Under the Auspices of the Graduate School
Urbana, Illinois
1937

EDITORIAL COMMITTEE

JOHN THEODORE BUCHHOLZ
FRED WILBUR TANNER
CHARLES ZELENY

1000—5-37—9900

UNIVERSITY
OF ILLINOIS
PRESS

EXPERIMENTAL STUDIES ON ECHINOSTOMA REVOLUTUM (FROELICH) A FLUKE FROM BIRDS AND MAMMALS

WITH THREE PLATES

BY

PAUL CHESTER BEAVER

CONTRIBUTION FROM THE ZOOLOGICAL LABORATORY OF THE
UNIVERSITY OF ILLINOIS
No. 491

ILL
v.15
cop.2

CONTENTS

Introduction 7
Life History and Morphology 8
Historical 8
Larval Stages 10
Miracidium 10
Redia 12
Cercaria 13
Adult 18
Morphology and Size Variation 18
Morphological Variation Induced by the Host Species . 27
Hosts and Geographical Distribution 29
Immunity Phenomena 33
Discussion of Related Species and Synonyms 39
Protocol of Experiments 56
On the Determination of Methods, Purity of Stocks, and Hosts 56
On the Transfer of Adult Worms from Host to Host . . 62
Conclusions 64
Summary 67
Tables 67
Charts 78
Plates 84
Bibliography 90
Index 95

ACKNOWLEDGMENT

The preliminary part of this work was done under the direction of Professor Henry B. Ward. It was concluded under Dr. L. J. Thomas, and many valuable suggestions were received from Dr. David H. Thompson. Whatever success has been attained is in large part accredited to them. I am indebted to Dr. Ward, Dr. W. E. Swales, Dr. N. Ishii, Dr. E. L. Miller, and Dr. E. W. Price for numerous records and valuable assistance gained from the study of specimens received from them as loans or gifts.

INTRODUCTION

Studies on larval trematodes and life history researches have progressed rapidly during the last few years and it should now be possible to take advantage of the accumulated facts in investigations of less familiar related problems, particularly the still very perplexing problems regarding the reproductive processes of the larval stages. In connection with studies on another species a rough idea regarding the life history of an echinostome species was obtained, and it was my original plan to study some of the less familiar phenomena including the reproductive processes within the snail host. There is at present much disagreement as to the general type or types of reproduction in these stages; parthenogenesis, polyembryony, and various other types of asexual and sexual reproduction each being proposed by various authors. Where studies have been made with special attention to reproductive phenomena the method of approach has been entirely cytological and the results contradictory. It was thought that an analysis of the variation in pedigreed offspring might serve as a favorable approach, and it appeared that the crown of spines and the cuticular spination of an echinostome would provide ideal material for a study of variation if the worms could be cultured in large numbers; and since certain laboratory animals were easily infected, an investigation of the above problems was attempted.

In order to take advantage of the researches already completed a final determination of the species was soon attempted. This proved to be a very considerable problem in itself, however, for so much contradiction and confusion was found that it was not until after most of the studies were completed that a final decision was reached regarding the species studied. Long and tedious studies were necessary in order to elucidate certain points regarding even the morphology of the very common adult worm. Most of my earlier experiments were designed to discover suitable methods for culturing each of the stages in the life cycle and a number of hosts were used with more or less success. It was noticed that the worms from some hosts differed slightly from those of others and two more problems were added by this observation. It was necessary first to prove absolutely that only one species of parasite was being studied, and secondly to determine the amount and nature of the variation due to the type of host. During the experimentation to exclude the possibility of mixed infections certain immunity phenomena were encountered. Only a very preliminary sort of study of these phenomena was possible, but some of the data obtained are interesting enough to report.

The whole of the present report is in many respects only preliminary. The volume of data that had accumulated seemed to require analysis, however, before further studies were made.

Special methods employed are described in the various sections where the reader is most likely to be interested in them. Brief introductions and discussions are also added to the various sections where they are most pertinent. Some repetition is held to be preferred to profuse cross reference and is therefore allowed for clarity.

LIFE HISTORY AND MORPHOLOGY

Historical

It is probable that the first experimental work on the life history of *Echinostoma revolutum* was done by Pagenstecher in 1857. Although he did not give a description of the encysted metacercaria or give sufficient data on the morphology of the adult to exclude the possibility of misdetermination, it seems fairly certain that he was working with *Echinostoma revolutum (D. echinatum)*. At any rate his was the first demonstration of the so-called life history of an echinostome. He found that echinostome cysts which were present in great numbers in *Paludina* sp. when fed to young ducks would develop into an echinostome species which he identified as *Distoma echinatum*. He also found cysts in *Lymnaea stagnalis* and *Planorbis corneus* which he considered identical with those from Paludina. Some of the other important observations which he made are that unhealthy or starved ducks do not hold their parasites and that the worms mature in about two weeks. He noticed too that although several thousands of cysts were fed to the hosts only about a hundred mature worms could be recovered from them. The gradual depopulation of the gut was explained on the basis of two factors: first, as the worms become mature the suckers become proportionally smaller and are thus less able to overcome the natural flushing of the gut, and secondly, the crown spines are gradually broken off until they are so reduced that the worms are crippled and eventually passed out. The latter is of minor importance if indeed it is a factor at all.

It has been observed many times that the cephalic or crown spines are lost under certain conditions such as rough treatment after poor fixation or death within the host so that fixation cannot be good, but it has not been demonstrated that these spines are shed by a living animal. Johnson (1920) reported that the cercariae have 43 spines while the adult has only 37, and stated that he believed this to be due to a natural shedding. A suggestion from Tubangui (1932b) to which I subscribe is that Johnson observed the spination of another species of cercaria (see page 15).

Ercolani (1881, 1882) reported experiments on this species but his identification was incorrect and his work of no importance here excepting that it has caused incorrect records to appear repeatedly in the literature. This work in which he reported experimental infection in ducks and mice and that of Generali (1881) in which he reported the same species from the duodenum of the dog were accepted by Railliet (1895) and Ercolani's figures were copied into his "Traite de Zoologie Medicale et Agricole." These figures with summaries of Ercolani's experiments have since appeared in English (Neumann-Macqueen, 1905), and thus have been fairly well distributed in Italian, French, German and English literature. This, of course, has a rather important bearing upon the present study because of the uncertainty attached to each of the reports of host and distribution records that might be based upon this or similar work. Wedl (1857) reported, described, and figured a species which he incorrectly identified as *D. echinatum.* Perhaps it was Wedl's description and figure that Ercolani used in identifying his material. They are somewhat similar.

Johnson (1920) was the first to make observations on all or nearly all stages in the life cycle of an echinostome. He was supposedly working on *Echinostoma revolutum.* Whether or not this is true some of his observations are of interest here, for he again demonstrated the method of infection and described some of the developmental stages. He took forty-five eggs from adult worms which he found in a duck, *Marila marila,* and when the miracidia hatched (in about three weeks) he exposed laboratory raised *Physa occidentalis* to them. He learned by subsequent studies that the miracidia develop directly into mother rediae, which after about three months produce a generation of daughter rediae, and these in turn produce the cercariae. As only forty-five eggs were collected it was not possible to infect many snails and consequently the studies on laboratory infections were not extensive. The morphology of the rediae and cercariae was studied on material collected in the field. Metacercariae were found encysted in *Physa occidentalis, Lymnaea traski,* and planarians, but he (Johnson, 1920) was unsuccessful in an attempt to recover them from laboratory exposed tadpoles of *Hyla regilla* Baird and Girard and *Notophthalamus torosus* (Rathke). By feeding metacercariae in *Physa occidentalis* he produced an infection in mongrel ducklings from which he recovered eight "full grown" and sixty-five less mature adult worms.

Vevers (1923) fed a young duck some cysts of a form which he identified as *Cercaria echinata* and after 12 days recovered fifteen immature specimens of *Echinostoma revolutum.* In the same year Miki (1923) found the tadpoles of *Rana esculenta* heavily infected with echinostome cysts and upon feeding these to rats and mice recovered worms identified

as *E. cinetorchis (= E. revolutum)*. The following year Lutz (1924) reported studies on this species. He obtained two infections in the duck *(Cairina moschata)* by feeding it *Physa rivalis* which carried a laboratory infection of metacercariae. The cercariae also came from *Physa rivalis*. More recently Tubangui (1932b) reported observations on both the larval stages and adult worms from experimentally infected pigeons; and Fallis (1934) has infected goslings by feeding them infected tadpoles.

The various stages in the life cycle can now be fairly well described and many of the details regarding the general biology of the species summarized from the researches of others. The present studies were made primarily as a basis for irrevocable identification, and the successful culturing of cercariae and adult worms. For that reason the observations were more or less confined to these stages. They will be discussed in the following order: miracidium, mother-redia, daughter-redia, metacercariae (agamodistome), and adult. The egg is considered a character of the adult worm.

Larval Stages

Miracidium

Eggs collected in feces, washed, and allowed to stand at room temperature hatch in from 18 days to one month. Active miracidia may be seen within the egg much earlier, however, as the flame cells and cilia appear several days before the embryo assumes the elongate shape of the fully developed individual. At about 6 days the young miracidium is nearly spherical, is ciliated, and has two flame cells in the center of the body. In diameter it is about one-half the length of the egg. During the next few days it elongates, dark granules appear in the region where eye spots are to be formed, cuticular plates become evident, and the right flame cell becomes more anterior in position. After 10 or 12 days eye spots are clearly defined, the body is as long as or slightly longer than the egg, and it has, with the exception of size, the appearance of the hatched miracidium (Fig. 11).

The following description of the fully developed miracidium is based on individuals incubated from numerous hosts and from several stocks. Very little variation was observed in the characters described. By inverting a glass funnel in a beaker containing the washed feces, miracidia were concentrated in the neck of the funnel and studied much the same as one would study an infusion of protozoa. As soon as the miracidia hatch they swim upwards, thus collecting in the narrow neck at the surface. *Intra-vitam* stains were found to be useful in differentiating some structures, and others were more clearly shown in stained permanent mounts. Many of the larvae were studied while still within the egg.

More often than not the larva is facing the operculum of the egg. The body is longer than the egg but much less in diameter. This necessitates a flexure which is at the posterior end and is in most cases dorsal, leaving the entire ventral side in contact with the shell (Fig. 11). The space not occupied by the larva is filled with two or three large clear globules which appear to be attached to the miracidium between the first and second row of plates.

The body is covered with large unicellular ciliated plates arranged in alternate fashion in four circumferential rows (Figs. 7, 11). The cilia are nearly uniform in length (10-14 μ) over the entire body. The four rows contain, respectively, from anterior to posterior 6, 6, 4, and 2 plates. When stains are used a large elongate nucleus is clearly seen along the posterior border of each plate.

The eye spots are clearly defined as dark crescents along the mesial edge of the lobes of the brain. A few scattered granules are also sometimes present along the posterior border. There is one flame cell in the anterior part of the body at the level of the second row of the cuticular plates, and a posterior one at the level of the third row. The anterior one is on the right side and ventral, the posterior one left and dorsal. The somewhat coiled excretory ducts lead independently to the lateral excretory pores at the triple junction of the posterior plates and those just anterior to them. The "germ balls" lie loose in the posterior two-thirds of the body and are variable in arrangement and number. Between the proboscis or protrusible papilla and the eye spots is a sac-like structure which in other species has been called an intestine. Lateral and ventral to this structure are masses of cells which by their staining reaction seem to be glandular.

The only echinostome miracidium sufficiently described to allow comparison is that of *Echinoparyphium recurvatum* which was recently described by Rasín (1933). Both forms have the 6-6-4-2 arrangement of the cuticular plates, two flame cells whose ducts empty between the third and last rows of plates, eye spots in similar position, and, in general, a very striking similarity is apparent. There is, however, one difference according to the descriptions given. In *E. revolutum* the posterior plates appear to be dorsal and ventral while in *Echinop. recurvatum* they are lateral, one on either side of the body.

After hatching, the miracidia swim rapidly and almost constantly until they find a host or finally die. Eighteen hours is the longest free-swimming period observed but under natural conditions it may be somewhat longer. The exact method of penetration was not determined, but they were often observed spinning vigorously with the proboscis-like anterior end undergoing a rapid succession of expulsions and withdrawals

while in contact with the body of the snail. None of the individuals observed were successful in breaking through.

Upon entering the snail, which may be almost any pulmonate, the miracidia metamorphose either directly into a mother-redia or into a sporocyst. Johnson (1920) believes that the sporocyst stage is omitted, but Rasín's studies on the closely related species *Echinop. recurvatum* and Mathias' (1925) observations upon *Hypoderaeum conoideum* indicate that the sporocyst does occur.

In the present study sporocysts were not observed, but no special effort was made to discover them. Snails were infected by the following method: the feces from infected hosts were washed repeatedly by decantation and the coarse material strained out, thus concentrating the eggs and minimizing the bacterial growth. The sediment containing the eggs was incubated at room temperature in 500 cc beakers or similar containers, and the water changed as often as was necessary to maintain freshness. As the miracidia began to hatch they came to the surface where they could be seen by looking through the glass container at right angles to a beam of light. When it was known that miracidia were present in the culture a few snails were placed in a coarse screen cage and partially immersed for a day or two.

Thus far this method has not been as successful as is desirable. Either because of improper conditions after exposure or because of too heavy infections, practically all the snails died before mature cercariae were produced. In the first of these experiments 96 snails were exposed and only 3 lived to produce cercariae; 3 others lived to the end of the experiment but were uninfected. As the snails died or were observed to be dying they were opened and examined for developmental stages. Only rediae were found in snails opened in less than six weeks after exposure, and mature cercariae did not develop until the ninth or tenth week. The snails were kept in a basement aquarium where the temperature ranged between 50 and 60° F, and were fed regularly on lettuce. Later attempts to infect snails were equally unsuccessful. Each snail was exposed to only 3 or 4 miracidia and kept in a green-house aquarium where the temperature was 80-110° F. Fewer snails died but none that lived were ever found infected.

Redia

The rediae of *E. revolutum* are so variable in size and shape that description of them is difficult. For that reason and for the reason that there is a very close similarity between all of the species of the genus none of the descriptions made thus far differentiate it from that of closely related species. My own observations have not been detailed enough to

describe this stage adequately even when added to and compared with other descriptions. Johnson's description (1920) of it is rather detailed but must be accepted with the reservation that it was made from studies on questionable material.

In one instance mother-rediae were observed in a snail that had been exposed to miracidia 62 days previously. These mother-rediae are large and elongate, measuring about 2 mm in length and 0.3 mm in width when extended. The pharynx is about 0.1 mm in diameter, which is relatively larger than that of the largest daughter-rediae. The collar, which in the daughter-rediae is fairly conspicuous, especially in the younger ones, is barely distinguishable in the old mother-rediae and likewise the posterior projections are not pronounced. Eighteen posterior and 9 anterior flame cells were observed.

The very young daughter-rediae are colorless but gradually accumulate an orange-colored pigment as they become older, so that the largest ones are always somewhat colored. This seems to be characteristic of all echinostome rediae. The pharynx and gut change in relative size as the rediae increase in total length. In small individuals they appear very large, and in the largest ones very small and inconspicuous. For example, a specimen which measures 0.28 mm in length has a pharynx which is 35 μ wide and in one 0.85 mm long it is 40 μ in diameter. Both specimens are about equally extended. In these same specimens the gut measures respectively 0.15 and 0.30 mm in length. A birth pore which seems to be dorsal can be seen on some specimens, and a collar is evident on all contracted ones. The pigment makes flame cell studies in the larger ones difficult, and in the smaller ones the flame cells are themselves so small that they are difficult to study. Although 36 flame cells were not seen in any one specimen it seems probable that there are four groups of them, 2 posterior ones near the lateral projections and 2 about half-way to the anterior end near the distal end of the gut, and that each of these groups contains 9 flame cells. Johnson (1920) described them in two groups, 17 in the anterior and 18 in the posterior.

Cercaria

The cercariae of the genus Echinostoma exhibit but few specific characters by which they may be distinguished. Tubangui (1932b) in working with the two species *Echinostoma revolutum* and *Euparyphium (Echinostoma?) murinum,* found that he could not distinguish one from the other except by counting the spines on the cephalic crown. In some forms it is difficult to study the cephalic spination, and accuracy in any case is not easy to attain. It naturally follows that any of the earlier descriptions which omitted the accurate notation of number and distribu-

tion of the spines must be considered as doubtful species save where other circumstances such as feeding experiments confirm the identifications.

The several researches on the life histories of echinostome species have demonstrated a relatively wide range of hosts for all of them, and the practice of using the host as a diagnostic feature must be abandoned in favor of more detailed morphological descriptions. For purposes of identification there seems to be no morphological feature in the echinostome species as reliable and serviceable as the cephalic spination. Some of the cercariae are alleged to be spineless, but none of these spineless "echinostomes," the "Agilis" group of Sewell (1922, p. 106), have, to my knowledge, been definitely shown to develop spines in later stages. The nature of the cephalic spines is perhaps no more characteristic of the various species than any one of a number of other features, but most of the other characters are either obscure or variable with the conditions under which they are studied. After long and tedious studies on the excretory system and the so-called "flame-cell formula" or "pattern" of this and other species, I am inclined to agree with Wesenberg-Lund (1934, p. 8) that while it is an admirable ambition it is also a practical failure, and I must confess along with him and Tubangui (1932b) that I cannot with absolute certainty determine the number of flame cells in this species. The measurements and figures were for the most part made on fixed specimens, since there is less variability due to methods of fixation than to the amount of pressure exerted on the living specimen. Three different methods were used in studying the spination: (1) living specimens were compressed and stained with the usual *intra-vitam* stains; (2) fixed specimens were cleared in glycerine to which a small amount of methylene blue had been added; (3) fixed specimens were stained in the usual manner and cleared in oil of wintergreen to which a few crystals of picric acid had been added. All of these methods are good but the second and third are best. For studying the cephalic spines I have used the second method more than either of the other two, and the third method is best for studying the cuticular spination.

The living cercariae are active swimmers and good creepers on a smooth surface. With the use of the two suckers they are able to creep fairly rapidly in the measuring-worm fashion, and even on relatively large tadpoles they sometimes find their way into the cloaca in a few minutes. They remain active and infective for 10-12 hours, then sink to the bottom and die after 20-24 hours. The swimming movements seem to be random. Numerous experiments were devised to determine whether they are attracted by any of the secondary intermediate hosts, but no action of this kind was discovered. However, after they have come into contact with the body of the host they immediately begin the creeping

movements which seem to be in the majority of cases in the direction of an entrance. This is particularly true on tadpoles. It was noticed that they very rapidly find their way into the small opening of the cloaca, and after a number of observations under magnification it was found that they usually creep downward and posteriorly until they reach the mid-ventral line of the tail, then turn anteriorly to follow this line into the cloaca. Fig. 5 indicates the usual course followed.

The following measurements were taken on a typical fixed specimen: length of body 0.323 mm, width 0.095 mm; length of tail 0.45 mm; oral sucker 41 by 46 μ; diameter of acetabulum 58 μ, pharynx 17 by 21 μ. The acetabulum is in the posterior third of the body (Fig. 12). The crown-spines over the head are identical in number and distribution with those of the adult. Although there is less difference in the relative size of the various spines there is a noticeable difference that corresponds with the condition found in the adult worms (cf. Figs. 1, 2, 3, with Fig. 9 and Text-figs. 1-5, p. 24). Riech (1927) found the same condition in *Echinoparyphium aconiatum*. Johnson (1920), on the other hand, reported a greater number in the cercaria, 43 instead of 37, and Iwata (1933, p. 1) implies the opposite in the statement that the head crown usually has 37 spines but that the younger ones have only 35. Figs. 1, 2, and 3 are camera lucida drawings of the crown seen from three views. The important features of this region are that there are 5 spines on the lappets, 6 unalternating lateral spines, and 15 dorsal ones in 2 alternating rows. The spines of the oral row are very slightly smaller than those of the aboral.

The cuticular spines are very inconspicuous, but when the specimens are lightly stained with methylene blue and observed without pressure or stained in picric acid after clearing they are distinctly evident over the whole of the ventral surface and almost all of the dorsal. They are most evident between the two suckers.

The oral sucker and pharynx are joined by a fairly long prepharynx which is rather indistinct. The esophagus and crura are also indistinct in unstained specimens. Associated with the oral sucker is a group of ducts which, as Brown (1926) and Wesenberg-Lund (1934) have already noted for *Cercaria echinata* (?), are outlets to a group of gland cells which are situated lateral to the esophagus (Fig. 12). There are 12 of these ducts, but whether a single gland cell is attached to each could not be determined. These cells and ducts are arranged in two groups. An anterior one in contact with and lateral to the esophagus empties through the three pairs of ducts which pass dorsally over the oral sucker and terminate at its dorsal lip, and a second group is more posterior. The three pairs of ducts from the posterior group are less easily demonstrated,

but by using only a small amount of neutral red they are shown to be more coiled and much smaller. In fact, they do not look exactly like ducts, being somewhat too irregular (Fig. 6). In about half of the toto mounts of the adult worms these larval structures are still noticeable as 6 minute papilla-like structures along the anterior border of the oral sucker (Fig. 9).

The excretory bladder is divided into a small anterior and a larger posterior compartment, both contractile. A single duct runs posteriorly into the tail and bifurcates abruptly into a right and left tributary with an opening (?) to the outside in the region of the 40th-50th circular muscle band, which is about 0.1 mm from the base of the tail. The two main excretory trunks pass anteriorly, lateral and dorsal to the acetabulum, where they enlarge somewhat and are filled with numerous (40-80) refractive granules to about the level of the pharynx. Here they narrow down and as they reach the level of the oral sucker turn back to form a complete loop and extend almost to the posterior end of the body where they again turn anteriad and extend to the region of the oral sucker. Fine vibratile patches are numerous in the canal that extends from anterior to posterior and are sometimes confused with flame cells. The total number of flame cells is probably 36 although it is possible that some were not found. The region between the two suckers is opaque and no flame cells were found in that particular portion of the body. The flame cells are arranged in 6 groups of three each, making a formula 2 $[(3 + 3) + (3 + 3) + (3 + 3)]$. Fig. 12 shows the distribution of the units found. Sewell (1922) has used the excretory system as one of the characters for separating the echinostome cercariae into the three groups "Echinatoides," "Coronata," and "Echinata." On the basis of Johnson's (1920) description of the cercaria of *Echinostoma revolutum* he placed it in the "Coronata" instead of the "Echinata" group, although he had originally put it in the latter and named the group after it. Sewell was correct in making *Cercaria Echinostomi-revoluti* a member of his "Echinata" group, however, for the cercaria on which the present work was done seems to fit this group in all respects excepting the tail. The presence of a tail fin is one of the diagnostic characters of the "Echinatoides," but Sewell does not definitely state that this character excludes a form from the "Echinata" group. The tail of the cercaria of *E. revolutum* has a distinct fin membrane, but as it is dorsal and vertical it is not seen in all preparations. At the tip of the tail the circular muscle bands do not continue further than the tail fin membrane, which leaves the tip proportionately more delicate than the rest of the tail (Figs. 4, 8, 10).

Discussion.—It is impossible to say how many times this species of cercaria has been described or how many times other species have been

mistaken for it. The original description is accredited to von Siebold (1837). It was not possible to examine von Siebold's description, and many of the older descriptions have not been studied for the reason they are too incomplete to be of use in this work.

In addition to the descriptions of the cercaria of *E. revolutum* by Johnson (1920), Tsuchimochi (1924), and Tubangui (1932b), there are in the newer literature descriptions of eleven 37-spined cercariae. Some of these are readily distinguished from *Cercaria Echinostomi-revoluti* by the character of the cephalic spines and the absence of a tail fin membrane, but the others do not allow a satisfactory systematic treatment. Lühe (1909), Brown (1926), and Wesenberg-Lund (1934) have described *Cercaria echinata,* which is generally accepted as the larva of *Echinostoma revolutum,* as having no tail fin membrane, and as having four corner spines larger than all the other cephalic spines. These descriptions agree throughout with the cercaria of *Echinoparyphium aconiatum* Dietz as described by Riech (1927) and Dubois (1928). While it is not certain that they are all identical, it is certain that they cannot be distinguished from each other and are all very different from the cercaria of *Echinostoma revolutum. Cercaria limbifera* Seifert, 1926, as originally described has 37 spines but differs from the cercaria of *E. revolutum* in size, arrangement of the spines, and character of the tail. According to the measurements given, it is a much larger form. The tail has both a dorsal and a ventral fin fold, and the 6 lateral spines of the collar are illustrated as being placed in two alternating rows continuous with the dorsal ones, making the arrangement entirely different from that of the cercaria of *E. revolutum.* The redia of Seifert's form has a much longer gut than the average mature redia of this species. Brown (1931) described a form which he considered identical with Seifert's *C. limbifera.* There is at least one very important contradiction in the observations of the two researchers, however. Brown's cercaria has only 35 spines, and the lateral ones are unalternating, as they are in the cercaria of *E. revolutum.* Since they both have the peculiar tail fin over the posterior two-thirds of the tail, both are larger than any of the others, and the rediae are both of the long-gut type, it is probable that Brown has corrected Seifert's observations regarding the number and arrangement of the cephalic spines. Rees (1931, Text-fig. a) who experimented with *C. limbifera* has given the best illustration of its tail. Dubois (1928) described 12 new species of larval echinostomes from Neuchâtel, and four of them are very similar to the cercaria of *E. revolutum.* One of them is identical with it and the other three may also be. All four of these cercariae are 37-spined forms with the spination formula (5) + 13 + 1 + 13 + (5), according to Dubois's method of writing it. No figures of them are given, and the descriptions of the spines are much too

brief to be of use in separating the 4 species. The description of *C. helvetica* XXIV agrees throughout with the cercaria of *E. revolutum*. The rediae are also identical, and the two are doubtless identical species. The only difference in the two forms *Cercaria helvetica* XXII and XXIII and the cercaria of *E. revolutum* is the absence of a tail fin in the two former. *C. helvetica* XXII is a little larger than *C. helvetica* XXIII, but in other respects they are very similar if not identical. The data given for *C. helvetica* XXV hardly if at all constitute a description. The spination is given as identical with *C. helvetica* XXII, XXIII, and XXIV. A tail fin fold is present, and the gut of the redia reaches almost to the lateral appendages, being much longer than in any of the 3 other forms mentioned above. I am doubtful, however, if the length of the gut in the redia is reliable in separating species of the echinostome cercariae. *Cercaria trivolvis* Cort, 1914, was found in the same host from the same general region as the cercaria on which the present study was made (Cort, 1914, 1915). The cephalic spines were not described in detail and the tail fin membrane was not mentioned in the description, making it necessary to examine the type material in order to confirm the suspicion that the two are identical. Very excellent preparations were made available to me by Professor Henry B. Ward in whose collection the "type" specimens were deposited. A dorsal fin is present in *C. trivolvis,* and the spination as well as all other features are identical with my own specimens.

The relation of *C. Echinostomi-revoluti* (von Siebold, 1837) to the most similar forms may be summarized as follows: *Cercaria echinata* as described by Lühe (1909), Brown (1926), and Wesenberg-Lund (1934) all differ from *C. Echinostomi-revoluti* in cephalic spination, the absence of a dorsal fin on the tail, and the character of the rediae. They are all probably identical with *C. Echinoparyphii-aconiati* as described by Riech (1927) and Dubois (1928). *Cercaria limbifera* Seifert, 1926, differs in the tail fin, which extends along the posterior two-thirds of the tail and is both dorsal and ventral; and it probably has only 35 cephalic spines instead of 37. *Cercaria helvetica* XXII Dubois, 1928, differs from *C. Echinostomi-revoluti* only in the length of the gut in the redia. The cercaria of *Echinost. revolutum* described by Johnson (1920) is a 43-spined form that is in all probability a misdetermination. *Cercaria trivolvis* Cort, 1914, and *Cercaria helvetica* XXIV Dubois, 1928, are synonyms of *Cercaria echinostomi-revoluti* (von Siebold, 1837).

Adult

Morphology and Size Variation

Three hundred stained permanent mounts are used as a basis for this description. The worms vary from 0.3 to 21.4 mm in length, and,

except for an interval of about 4 mm between the lengths 13 and 17 mm, the specimens used are fairly evenly distributed in size and almost reach the maximum size given in previous descriptions. Leidy (1888) reported specimens up to 25 mm, and Barker and C. A. Beaver (1915) give 22-30 mm as the length for *Echinostoma coalitum,* which is identical with *E. revolutum.*

Echinostoma revolutum becomes mature, that is, it begins producing ova, when it attains a length of 4 to 7 mm, differing considerably in the various hosts. The range then for mature worms is from 4 to 30 mm in length. This size range is so great that a description of any one size is grossly inadequate for an accurate diagnosis of all other sizes, and a description by the ordinary methods is impractical when applied to worms having so great a range in size. This would not be true, however, if it were possible to give proportionate measurements applicable to all sizes which, as is shown below, cannot be done. Practically every feature has been measured and plotted; and in no instance is the proportion between two organs or structures a constant throughout the series.

The worms used were all killed by dropping them from warm tap water into Gilson's fixative heated to 65° C. This method gave fairly uniform results, leaving the worms well extended. Relaxing the worms by shaking was avoided because of the possibility of injury to the cephalic and cuticular spines. For very large worms it was necessary to raise the temperature to 68-70° C in order to get straight specimens. As soon as the specimens straightened out they were pipetted off and the fixation completed in warm solution. No flattened specimens were used, for if worms are even slightly flattened their body proportions are greatly altered.

The greatest width of the body is at the acetabulum in immature specimens, and in the region of the uterus in mature ones. Charts 1, 9, and 10 show the plotted measurements and the partially smoothed curve which expresses the ratio between body length and width. In the smallest specimens the length is three times the width, and it gradually changes to about ten times the width in the largest ones. In worms that have died without fixation the ratio of length to width is often as much as 15:1. In like manner the acetabulum changes from one-fifth to less than one-fifteenth of the body length (Charts 1, 11, and 12).

The position of the acetabulum and ovary is frequently used in describing this and other species. Their position in *E. revolutum* is shown for all sizes up to 20 mm on Charts 4, 13-16. In the smallest individuals the ovary lies within the posterior fifth of the body, but when a length of 5 mm is reached it becomes more anterior and remains near the middle of the body for all larger sizes, being slightly nearer the

anterior end in individuals over 10 mm in length. The distance from the acetabulum to the ovary or that part of the body occupied by the uterus, shifts from about one-fifth of the body length to one-fourth of it, varying considerably on either side of the line, but in no case is it as much as one-third or as small as one-sixth of the length of the body. The preacetabular region becomes less in proportionate length as the animal increases in size. In the smallest individuals, less than 1 mm in length, it is about one-half of the body length, and in the larger ones it is only one-sixth to one-eighth of it.

The relative size of the suckers and the cephalic collar is shown in Charts 2, 3, 17, and 18, where it may be seen that the acetabulum increases in size at a greater rate than does either the oral sucker or collar. The acetabulum and oral sucker are almost equal in the smallest individuals, but in larger worms the latter has a diameter of less than one-third of that of the acetabulum. The diameter of the collar is almost 2 times that of the acetabulum in small individuals and in larger ones it is about equal to it, while in the largest ones the collar is only three-fourths as large as the acetabulum.

The transverse diameter of the pharynx is usually only slightly less than its length. In all sizes of worms above 2 mm it is about equal in length to the diameter of the oral sucker, and in very small worms it is somewhat less, about two-thirds (Charts 19 and 20).

It is difficult to express the size of the testes and ovary because of their variability in shape and the consequent inadequacy of length and width measurements. An expression of the volume of each would of course be ideal but impracticable. The average diameter (half the sum of width and length) is used here as the most practical measurement of size (Chart 5). The posterior testis is slightly larger than the anterior one in most individuals but here the average of the two is used. After sexual maturity is reached the average diameter of the testes is in all sizes of worms about one-twentieth of the total length of the body, and the ovary about one-thirtieth. These proportions, of course, do not hold true for the youngest specimens, and there is a good deal of variability in all sizes.

None of the structures or regions measured and plotted can be described as having a constant proportion to any other part of the body in all sizes of worms, although after maturity is reached some of the ratios are fairly constant. The oral sucker is equal in diameter to the length of the pharynx; the average diameter of the ovary is about one-thirtieth of the body length; the average diameter of the testes is about one-twentieth of the body length; and the ovary is near the middle of the

body. The length of the region between the acetabulum and ovary is also fairly constant after the uterus is formed, regardless of the number of eggs it contains.

Body Spines.—In addition to changes in body proportions there are also structural changes caused by the growth and aging of the worms. The cuticular spination is an example of this, as illustrated on Charts 7 and 8. Cuticular spination also varies in the different hosts. In young individuals from all hosts the dorsal spines extend to the region of the acetabulum, and in the very young individuals, less than two days old, the whole of the dorsal side of the body bears minute spines. Ventrally they reach to the posterior end in specimens up to about 3 mm. With increasing age the dorsal rows diminish in number until they have entirely disappeared in some individuals from bird hosts. Ventrally the spination reduces much less in extent; and it also differs in the bird and mammal hosts. The posterior limit of the spines moves forward more slowly in mammals than in birds, sometimes reaching the ovary in bird hosts but rarely going anterior to the anterior testis in those from mammals. Anterior to the acetabulum the number of rows of ventral cuticular spines is apparently not much altered by age or host. Counts made on all sizes of worms vary between 28 and 42 rows from the genital pore to the anterior end, but there seems to be no correlation between high or low counts and the age or size of the worm. The mean is doubtless much nearer the higher number, probably 36-38. The counts cannot be very accurate since some of the posterior rows are usually folded under the acetabulum. The rows on the dorsal and ventral sides are joined laterally, and the spines of successive rows alternate, being arranged in oblique rows as well as transverse.

The size of the cuticular spines cannot be determined very accurately as it is rarely possible to get accurate measurements on both the length and width of the same spine. However, the length of the lateral spines and the width of the dorsal or ventral ones can be measured. Chart 7 gives the length of these spines in the tenth row measured at the lateral margin of the worms. They become progressively smaller in proportion to body length, and in worms from bird hosts they are relatively longer than in those from mammals. The relation of width to length of the spines also changes as the length of the worms increases. The width of the ventral spines plotted against the length of the lateral ones of the same row shows that the width changes from one-half to almost equal the length in large specimens from bird hosts. In those from mammal hosts the ratio of width to length is nearer 1:1.5 in the largest specimens.

A point that should be noted here is that this species is spiny on the ventral side of the body in all individuals, but the larger specimens, 10

mm in length or more, may be without dorsal cuticular spines. Several instances were observed in which the worms were entirely without spines when taken by autopsy that had been delayed so long that the worms were dead for several hours. This condition is not due to a loss of spines alone, however, but to a loss of the entire cuticula. In some cases portions of the cuticula may be seen adhering to the body in small patches. It should be added also that this character, the distribution of the cuticular spines, which has been used as an important character in both specific and generic diagnosis of echinostomes, should be as thoroughly investigated for other species, for just as it cannot be said that *E. revolutum* is either with or without dorsal cuticular spines without reservation, the same probably holds true for other species.

Collar Spines.—Except in cases where spines are lost or duplicated, or where accessory spines appear, there are 37 cephalic spines in all of the several hundreds of specimens examined. In other words, *E. revolutum* is a 37-spined species but does not in all cases bear exactly 37 spines. In well fixed worms the spines are not often lost but are sometimes broken in handling. In fact they are so well attached in the living worm that it is difficult to extract them at all. On the other hand, a worm that has been dead a few hours in the gut of its host easily loses both cephalic spines and cuticula and in some cases may be entirely without either and still appear fairly normal in other respects.

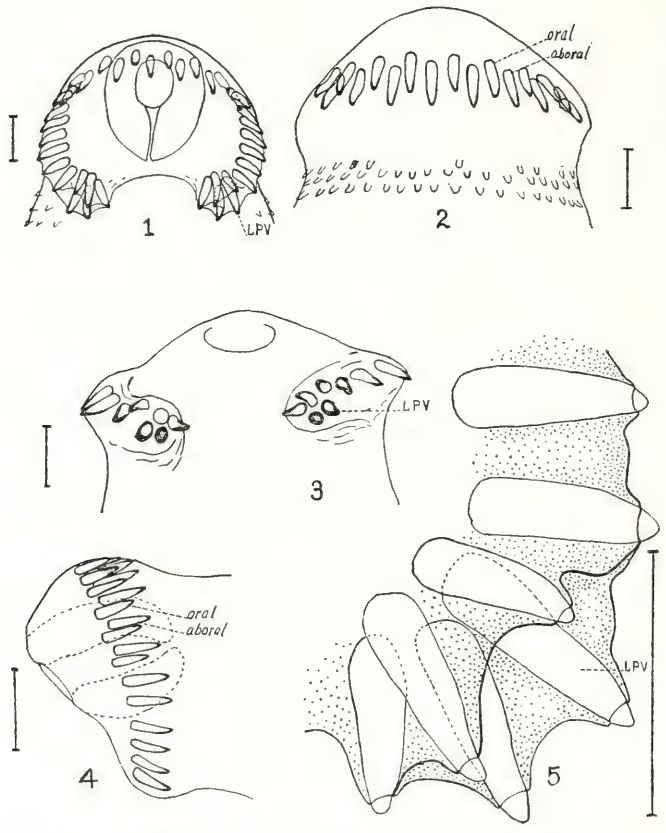
The cephalic spination of 220 worms was observed minutely and the condition of each spine carefully noted. Of this number, 173, or 79 per cent, were found to be entirely normal, i.e., they bear 37 typically placed spines, all of which are normal in size and shape. The other 21 per cent fall into 3 general types of abnormalities. (1) Spines may be missing either because of having been injured in life or improperly handled after fixation. Usually these spines can be counted, however, for a scar is often noticeable and even when a scar cannot be seen the vacant space is evident. Thirty-one of the 220 worms were abnormal in this respect. (2) A less common type is produced by accessory spines, which are most common in the larger individuals, being added where spines normally do not occur. They are smaller than the others in all but one case observed and are easily distinguished from them. In a few instances accessory spines were observed growing from the same position as a normal one, a condition described here as a "duplication." Accessory spines were observed in 17 of the 220 individuals. (3) A third type was observed in 11 of the 220. In these the position of the spines is normal but one or more of them are much smaller, being in some instances very minute. Their size and position suggest that they are being regenerated to take the place of spines that have been lost by injury.

Due to the three types of abnormalities described above, particularly the first, it is important to give special attention to the arrangement of the spines of the collar. Fortunately this character is almost invariable and for that reason is of greater value in diagnosis than spine number or size. Even in specimens that have died in the host and lost most of their spines an accurate determination may often be made if some region including one of the lappets has retained most of the original spination.

Although the grouping of spines for description is somewhat a matter of choice there is a more or less conventional terminology which has been adopted here to facilitate comparison. There are 3 general groups, 2 of which are paired: the 2 corner groups of 5 spines each, 2 lateral groups of 6 each, and a dorsal group containing 15 alternating spines (Text-fig. 1, p. 24). When the specimen is seen in side view or when the lappets are lifted, the position of the lateral and corner spines is seen most clearly (Text-figs. 3, 4). The arrangement of the spines within the various groups may be described as follows: 5 alternating corner spines, 3 of which are on the surface or oral, and 2 that are deeper or aboral; 6 laterals in a single row; and 15 alternating dorsals, 8 of which are anterior or oral to the other 7 which are aboral and continuous with the 6 laterals. The spines are symmetrically arranged; the saggital plane of the animal passes through the median aboral spine.

The relative length of the collar spines to that of the body changes very appreciably as the worm becomes larger (Chart 6). In 1-mm worms the lateral aboral corner spine (*LPV* in Text-figs. 1, 3, 5) varies slightly on either side of 28 μ, in 10-mm ones it is around 90 μ, and in the larger ones it is relatively much shorter, varying 15-20 μ on either side of 120. The relative length of each spine to each of the others usually is not changed by age. In some instances, however, the median dorsal spines become relatively smaller in the older worms, particularly in the oral row in bird hosts. However, in the relative size of the various spines there occurs a good deal of difference which cannot be correlated with age. The lateral aboral one is, in all cases observed, the largest of the 5 corner spines. The median aboral and the middle one of three orals are about equal, the oral one usually being the larger. The lateral oral corner spine is more often than not the smallest one on the collar, and the median oral is about the same size (Text-fig. 5). The most ventral one of the lateral spines is about equal in size to the middle oral corner one or a little smaller than it, and each of the other laterals is slightly larger in a dorsal progression (Text-fig. 4). The dorsal aborals are about equal although the median ones are usually slightly longer than the more lateral ones. The orals are each slightly smaller as a rule than the aboral ones nearest them (Text-figs. 1, 2, 4).

A number of counts and measurements were made on the cephalic spines of the cercaria and metacercaria in order to determine whether spination is the same in both larval and adult stages. There is apparently



TEXT-FIGS. 1-5.—Cephalic spines of the adult worm, showing the arrangement and relative size of the individual spines. All are camera lucida drawings. All scales are 100 μ.

1.—Ventral view. Notice that the oral spines of the dorsal side appear to be much shorter than the aborals because they are seen more from end view. Spine *LPV* is the lateral ventral aboral, the largest of the corner spines.

2.—Dorsal view. Aboral and oral spines almost equal in size when seen from the same angle.

3.—Ventral view of specimen killed with the lappets brushed up to show the position of the ventral aboral spines. Corner spines all seen from end view.

4.—Lateral view. Notice that the 6 unalternating lateral spines are continuous with the aboral dorsals. In this specimen the orals are considerably smaller than the aborals.

5.—Ventral view of the corner spines greatly enlarged.

no difference in the number of spines and only a slight difference in the relative sizes. On the whole there is less size difference among the various spines of a single individual in the larval stages than in the adult, and the number is more constant. Careful examination of 25 cercariae from each of 3 snail hosts, fixed and cleared in glycerine, gave the following data: 72 with 37 spines, typically arranged; 2 with 1 of the dorsal aborals missing; and 1 with a dorsal aboral spine duplicated—giving a count of 38 for this individual. Accessory spines may have been overlooked, as they would be very small if present.

Genital System.—The genital system is apparently very similar to that of all other species of the genus. The testes are usually regular in outline in unflattened fixed specimens but frequently are divided into 3-5 shallow lobes (Figs. 13, 14). In worms that have died in the host the inner parts of the testes pull away from the outer epithelium and give the appearance of a lobed testis, and worms that have been killed under pressure almost invariably have irregular testes. The vasa deferentia run into a large seminal vesicle which lies in the posterior part of the muscular cirrus sac. The anterior part of the sac is occupied by the pars prostatica, the voluminous prostate gland, and the well developed cirrus. The cirrus opens through a common pore with the metraterm, which runs along the right side of the cirrus sac in all specimens examined. Looss (1899, Figs. 5, 10) shows the uterus on the right side in one and on the left in the other figure. The position of the cirrus sac is usually anterior to the middle of the acetabulum and is often entirely anterior to it in the younger worms; in some cases it is entirely dorsal and reaches the posterior border of it (Fig. 14). The female system is also typical of the genus, having, instead of a seminal receptacle, a more or less voluminous receptaculum seminis uterinum. A fairly distinct Laurer's canal, a diffuse shell gland, vitellaria from acetabulum to the posterior end, and a small vitelline reservoir are also characteristic structures. The uterus is long, regardless of the age or maturity of the worm, and is coiled transversely into 7 or more loops. After it begins to fill with ova it usually does not have less than 9 loops, and more are added as the eggs increase in number. Barker (1916) has attempted to use the number of eggs in the uterus as a character on which to key the muskrat echinostomes. This is not a good character, however, for this species may have from none at all to over 6,000 eggs. The uterus in a 21-mm worm from a muskrat was teased out and the eggs counted. Each egg was counted as it was drawn into a pipette and in this manner 6,200 were counted and a hundred or more were probably left in the tissues. The number of eggs produced in 24 hours was also determined for several worms at different ages. In Transfer X (p. 64), a single immature worm

was taken from a rat and transferred to the cloaca of a pigeon. It matured and began producing eggs in 6 days, or when it was 18 days old. After it had been producing eggs for 7 days, a 24-hour fecal sample was taken, and by the dilution and direct count method the number of eggs was determined to be 4,100. The following day the count was 3,800 and the counts for the next 2 days were respectively 3,460 and 3,402. Similar records were taken on other transferred worms and the highest number for any one day was 4,600, which is more than 3 per minute. This count was made by the Rivas method (1932, p. 479), which was finally adopted as the most satisfactory for this type of work. Most of the worms produced 2,000-3,000 eggs daily to within a few days of death, and 700 per day is the least any worm produced. The 700 per day count was made on a worm 59 days old.

Egg size varies with the age of the worm and is possibly somewhat altered by the host. The younger worms produce eggs with a very great range in size, and the average length is much greater than in more mature ones. The range in size for the eggs of the worms from all hosts was found to be 91 to 145 μ by 66 to 83 μ. The largest were in the first positive stools of Cat 4 and the smallest were found in Muskrat 261, but those from some of the pigeons were almost as small as the ones from the muskrat. Usually the eggs from bird hosts are darker in color and appear to have a thicker shell but there are some exceptions to the rule, for those from muskrat hosts are usually almost as dark as the ones from the pigeon. Variability in the size and color is much too great to allow the use of this character in distinguishing *E. revolutum* from other closely related species.

The transfer experiments also gave some interesting data regarding the self-fertility of the species. On 10/24 Rat 26 was killed in order to get some immature worms for transfer. These worms were 12 days old, and so far as could be determined by means of microscopic examination of the living worms, all were sexually immature, having no sperm in either the seminal vesicle or receptaculum seminis uterinum. In fact the latter organ was not yet formed. One worm was transferred into each of 4 pigeons (T7, 8, 9, 10; see p. 64). Three of them gave positive stools on the fifth day after the transfer was made, and T9 lost its worm before it matured. About a week later 50-100 ova were collected from the feces of each pigeon and incubated in tap water. Normal miracidia developed in the eggs from each of the worms and the rate of development was the same as for those from hosts carrying an infection of several worms, so that cross-fertilization would be possible. The question as to whether trematodes are self- or cross-fertile is one that has received a good deal of attention. The only data to be found on it are occasional

reports of observations on worms with the cirrus inserted into either its own metraterm or into the genital pore of another individual (Looss, 1893). Where single individuals are found in the hosts it might be assumed that they were self-fertile if viable eggs were produced, but one could not be sure that a second individual had not been present earlier.

Morphological Variation Induced by the Host Species

In the preliminary experiments it was noticed that the worms from the various hosts had slightly different appearances, but these differences were in most cases so slight that they could not be described or figured. It became especially noticeable when worms from avian and mammalian hosts were compared, and since some confusion seems to be due to lack of understanding of this variability it was thought worthwhile to study it in more detail.

A fairly complete size series was obtained from both avian and mammalian hosts, and in order to compare all sizes of worms the most satisfactory method was found to be the graph system commonly used in biometric studies. Three graphs were used for the comparison of each character in most cases. One was used to show the number of individuals used and their measurements from the bird hosts and a similar one for those from the mammals. A third chart to show the partially smoothed curves was taken from the other two. This method indicates the range in all classes and the variability within the range; it gives the proportions of the worms at all sizes, and the differences in the worms from the two types of hosts. The significance of the differences can be judged directly from the plotted data. About fifty such graphs were prepared, but only twenty of the most illustrative have been included here.

Although there is a very marked difference in the size attained in the various hosts used in these experiments, *Echinostoma revolutum* apparently does not reach a definite size but continues to grow until it is expelled. The maximum size indicated in the charts does not express the absolute maximum size for the species but the actual size of the individuals studied. The curves were made by drawing straight lines through the points obtained by averaging the points within arbitrary limits of range. The worms from all hosts were fixed, stained, etc. in exactly the same manner in so far as it was possible. None were killed under pressure.

Various authors, notably Looss (1899), have suggested that much of the variation in this parasite may be due to the host species, but no one has to my knowledge attempted to measure or describe it.

One of the most conspicuous differences in worms from the two types of hosts is the greater reduction in the dorsal cuticular spination in the

worms from birds. While there is also a variation in those from each of the mammals and birds, the differences are less marked than when the bird- and mammal-reared worms are compared collectively (Chart 8). That the reduction in dorsal spines is not due to age alone is shown by the fact that it is not so easily correlated with spine reduction as is the body length. When the number of complete rows of dorsal cuticular spines plotted against the length of the worms is averaged and a curve drawn, a very pronounced contrast in the larger worms becomes apparent. For example, an 8-10 mm worm from a duck may be completely spineless on the dorsal side while on another of the same size from a rabbit or muskrat there may be 10-15 complete rows of spines. This is particularly important in the light of some of the definitions of the two genera *Echinostoma* and *Echinoparyphium*. Sprehn (1930, 1932) considers one of the differences in these genera to be the absence of dorsal cuticular spination in the former and the presence of it in the latter. Following this definition would necessitate putting the worms of this species into both genera.

The length of the cuticular spines was measured and plotted in Chart 7. Contrary to expectations, the spines are longer in comparison to body length in bird-hosts, and the difference is considerable. In the smaller worms the collar spines are longer in mammals, but in the larger worms the variability is so great that although they are probably also longer in mammals the data do not clearly indicate it.

Differences in body proportions are not very pronounced in most cases, but since there seems to be a significant variation in the same direction for all parts measured it is clear that some of the variation in shape in this species can be attributed to the different types of hosts. As a rule the worms develop more rapidly in birds (Table 7, p. 77), become mature earlier and at a smaller size, and live a much shorter period. The differences in proportion are shown in Charts 9-20. The worms from birds progress toward mature proportions more rapidly than those from mammals, making a rather pronounced difference in the younger individuals. There is less difference in those between 6 and 8 mm, but between 8 mm and the upper limits the differences become progressively greater, the worms from birds being generally wider and stouter than those from mammals. There is apparently not much difference in the size of the acetabulum (Charts 11, 12), but when the diameter of the oral sucker is plotted against that of the acetabulum a significant difference is observed (Chart 18). The larger worms from bird hosts have a relatively larger oral sucker. Similarly the collar is larger as compared with the acetabulum in the larger specimens from birds, and smaller in the shorter ones (Chart 17). Coincident with this variation is the difference in

growth rate in the two types of hosts mentioned above. It should be mentioned, however, that differences in body proportions are not great enough to make them readily discoverable by inspection. It is only when large numbers of individuals are compared biometrically that the differences become obvious. It is shown in Charts 9-14 that the variability within each group is sufficient to give much overlapping.

In one instance the variation due to the host was very conspicuous. The worms taken from the pig were barely mature after four weeks and the size of the various parts as well as the whole were very strikingly dissimilar to the average from the other hosts. The body is only 3.6-3.75 mm long and wider than normal. The acetabulum is large and the post-testicular region is much reduced. The testes and ovary are nearly equal in size and much too small, being slightly smaller than the pharynx. The spination on the collar is normal in number and distribution of spines but the length is greater than normal. The whole appearance of these worms is so different from the normal that they could easily have been described as a different species had their origin not been known.

It may be concluded that while there are very definite variations induced by the conditions within the various hosts, they are comparatively slight in most cases. However, they are conspicuous enough to account for some of the confusion in the literature regarding this species. It is particularly true of the cuticular spination and the total length attained, there being a range of from less than 4 to about 30 mm in the length of the worms producing eggs. The differences in proportion can be explained on the basis of differences in growth rate, but the differences in the character of the cuticular spination are as yet unexplained.

Hosts and Geographical Distribution

Echinostoma revolutum is one of the most cosmopolitan of trematode species, having been reported from every temperate region in the world with the exception of South Africa and inland China. It is also found near the equator but is apparently less common there. Tubangui (1932a, b) records it as fairly common in the Philippine Islands (Luzon), and Picard (1930) took an echinostome species from the pigeon in Java which is probably *E. revolutum*. The regions where it is known to occur are indicated in Tables 1, 2, and 3. Only the records that have been published since Kowalewski (1895) and Looss (1899) redescribed the species are used, and some of these reports are doubtful and have either been excluded or indicated as questionable.

This species seems to be as cosmopolitan in its choice of hosts as it is in geographical distribution. This lack of specificity in all of its parasitic stages is of course the most important reason for its wide

distribution. The experiments of Tsuchimochi (1924) and Anazawa (1929) in which they infected certain mammals suggested that this species might also occur in mammals in nature. My own experiments have confirmed those of Tsuchimochi and Anazawa and added many new experimental hosts; a number of new records of natural infections in mammals and birds have also been obtained. Tables 1, 2, and 3 give the hosts, distribution, and references for the incidence of the cercaria, metacercaria, and adult, respectively.

Cercaria Echinostomi-revoluti is known to occur in at least four genera and ten species of pulmonate snails. Although it may be found in several species of a particular region it seems to be decidedly more prevalent in a single species. In the vicinity of Urbana, I have searched for it in *Physa gyrina* for three years and have never found a single infection. Miller (1936), who made more extensive collections in the same region, has given but a single record of it from a stream where the incidence is particularly high in *Helisoma trivolvis*. The percentage of infection in *P. gyrina* in this region is probably much below 1 per cent while in the same region it is as high as 60 per cent in *Helisoma trivolvis* during the summer months.

The agamodistome may occur in any of a very great variety of hosts. In this study a number of attempts were made to determine the most "natural" hosts, and for this particular region it seems that they are *Physa gyrina, Helisoma trivolvis,* and *Rana pipiens,* named in the order of frequency of natural infection. *Helisoma trivolvis,* which also serves as the primary intermediate host, carries a rather high percentage of infection in nature, but not as high as it was at first thought; an erroneous idea regarding this is easily acquired if care is not taken to insure against laboratory infections of the snails examined. By carefully drying each snail as it was collected in the field and isolating it in the laboratory or by examining it immediately, I found that in the one region which received most of my attention Physa is probably the most natural host. Six adult *Rana pipiens,* 6 half-grown tadpoles of the same species, 4 medium-sized *Physa gyrina,* and 6 small *Helisoma trivolvis* were collected from the same bend of a stream, all on the same day, and examined for cysts of *E. revolutum.* Each cyst from the frogs and most of those from the snails were pressed under a coverslip and carefully examined. The results are shown in tabular form:

Host	*Number of Cysts*	*Number of hosts positive*
Adult *R. pipiens*	2 to 27	4 out of 6
Larval *R. pipiens*	6 to 41	4 out of 6
Physa gyrina	12 to 125	3 out of 3
Helisoma trivolvis	1 to 18	6 out of 6

When tadpoles and the two species of snails, all laboratory raised, were put into a small aquarium with active cercariae, the tadpoles seemed to get the largest share of the parasites and the Helisoma the least. It was found that it was necessary to govern carefully the infections in the tadpoles and Physa for they would become so heavily infected in a few hours that they would soon die. On the other hand the Helisoma very rarely died because of heavy infection with metacercariae. Heavily infected tadpoles became swollen and died with a sort of general edema. Lutz (1924) made the same observation on tadpoles infected with *Echinostoma nephrocystis (? = E. revolutum)*. The following experiments will serve to illustrate how readily the cercariae enter and encyst in tadpoles. A laboratory raised tadpole (*R. pipiens*) about 25 mm in length was put into a 200 cc container with 75 freshly emerged cercariae and left for about one hour. They were observed from time to time, and on one occasion 7 cercariae were seen creeping around within the transparent cloaca. The following day the tadpole was dissected under the binocular microscope and 53 cysts found in the kidneys and adjacent ducts, and 7 cysts were found in the gill chamber. Many other tadpoles were infected and used in the feeding experiments. A half-grown hairless rat (Rat 27), for example, was fed a 30 mm tadpole that had been in a finger bowl with an infected snail (C6-9) for about 2 days, and 431 worms were recovered from it. Two small Physa were also infected from snail C6-9, and fed to Pigeon 15 from which 63 worms were recovered (Tables 5 and 6). There can be no doubt that Tsuchimochi (1926) and Fallis (1934) are correct in their statements that *Echinostoma revolutum* will use either a snail or a tadpole, depending upon which is available. There is apparently very little difference in the two species of tadpoles used in the experiments; the larvae of *Rana catesbiana* were as easily infected as *Rana pipiens*. *Pseudosuccinea columella* and *Fossaria modicella* were also easily infected. It was also possible to get infections in several more unusual hosts. Five *Sphaerium* sp. and 3 *Pisidium* sp. were exposed to heavy infusions of cercariae for 8 hours and examined after 24 hours and 5 days, respectively. In one Sphaerium 14 cysts were found, but in the same individual 22 of the cercariae had lost their tails and were still creeping around under the mantle; another had 9 cysts and 18-20 unencysted cercariae; one had 6 creeping and none encysted, and the other two, which were smaller individuals, had none. Eight cysts were counted in a Pisidium five days after it had been exposed for eight hours. Two others of the same lot were negative and one had only two cysts. The cysts when present were in the flesh of the clam near the hinge in both species. Several species of scaly fish were exposed and examined but none were ever found positive. It is probable, however, that the catfishes are sometimes natural hosts, for the two

experiments with the black bullhead, *Ameiurus melas,* indicate that the cercariae react to it very much as they do to the tadpoles. A fish that was about 8 cm long was put in a shallow dish with 250-300 cercariae and left for eight hours. When it was dissected the next day 67 cysts were found deep in the substance of the kidneys, 4 in the left mesonephric duct near the kidney, and 9 in the same duct near the cloaca—a total of 80 cysts. They were all normal in appearance having well formed walls, and the host did not appear to be suffering ill effects. A second fish of the same species was left in a shallow dish in 1 liter of water with an infected snail for 4 days. On the last day it was somewhat bloated and very inactive and finally died. Several hundreds of cysts were found in the kidneys and associated ducts.

In the experimental work done thus far on *Echinostoma revolutum,* including the present study, it has been shown that 14 or 15 species of snails may serve as secondary intermediate hosts, and none of the other species of snails have been experimentally excluded as possible hosts. Johnson (1920) has made the only unsuccessful attempt to infect tadpoles. At least four species—*R. pipiens, R. catesbiana,* and the "tadpoles" of Tsuchimochi (1926) and Fallis (1934, *Bufo americana*)—are known to carry infections of this species. Miki (1923) found cysts in *R. esculenta,* which he fed to rats and mice and recovered *E. cinetorchis(= E. revolutum).* According to Ando and Ozaki (see Dollfus, 1925, p. 90) the metacercariae of *E. cinetorchis (= E. revolutum)* were found in *Rana nigromaculata, R. rugosa,* and *R. japonica,* which increases the number to seven or eight known amphibian hosts if *E. cinetorchis* is accepted as a synonym of *E. revolutum* (see p. 47). *Ameiurus melas,* which is shown by this study to be a probable natural host, is the first fish to be reported as a host for this species.

The adult worm apparently may be found in almost any bird or mammal (Table 3). Natural infections are, of course, limited to those animals that feed upon aquatic animals, but many others may be experimentally infected and act as "normal" hosts if not "natural" ones. In the present study an effort was made to discover the host which would serve as the most satisfactory one for routine culturing of this species, and incidentally the pig was infected to determine the effects produced by the larger host. These experiments are outlined in another section (p. 56) and summarized in Table 6. For culturing the worms rapidly the pigeon or guinea pig are the best adapted, but greater size is attained in the duck or rabbit. The rats and mice used were found to be undependable; and although the worms develop as well in the dog and cat, these hosts require more space and attention and were for that reason less often used. Although the worms develop slowly and attain a smaller size in the domestic pig, in view of its habits it may be expected to harbor natural infections of *E. revolutum.*

Immunity Phenomena

Although only a few experiments were designed specifically to test the types or degree of immunity developed in the various hosts used, the large number of infections and variety of hosts used has lead to certain new conclusions and confirmations which are being reported even though they must in most cases be repeated and extended before the results are accepted.

Table 5 and 6 (pp. 72-76) indicate the number of hosts used in the first series of experiments. The number of hosts from which no worms were recovered or were at least not known to be infected are also given here. In most of the hosts reported negative, it is certain that the worms remained in the gut for a few days and in some cases were lost only shortly before maturity was reached. Those hosts reported negative are, then, those which contained no worms when autopsied or did not produce stools which were found positive by fecal examination.

It may be seen from Table 6 that all attempts to infect cats, dogs, chickens, hogs, and guinea pigs were successful. Very few of the pigeons, ducks, or rabbits were negative, but the mice and rats could not be relied upon to hold the worms. This is particularly true of the albino and hooded rats, in which no infection was accomplished in 14 attempts. On the other hand, large mature worms were taken repeatedly from wild and hairless rats.

All of the rats were treated similarly. The diet after the experiments began was the same for all and they were fed in all cases as much food as they would eat. They were obtained from the "discards" at the Animal Breeding Laboratories at the University of Illinois. Unfortunately, the history of these rats was not obtained, as it was not thought to be sufficiently important at that time. Accurate data on the diet and the pedigree of each rat would, however, have made the experiments of greater value since these factors probably account for some of the "natural immunity" displayed. Ackert *et al* (1931a, b) and others have shown that vitamin deficiencies alter the natural course of infections, and since other researchers (Tsuchimochi, 1924) have reported infecting laboratory rats with *E. revolutum,* it seems even more likely that under different conditions the rats used in these experiments would have been more consistently positive. Tubangui (1932b) had similar experiences. He was using pigeons and albino (?) rats in his experiments and fed cysts of *E. revolutum* to both. While the pigeons were always positive the rats were in each case negative. His rats may also have been maintained on a diet which rendered them nonsusceptible.

The results obtained from the mice are not particularly significant in this connection as it was not possible to determine whether the wild

ones actually ate infected snails or not; and all of the positive mice either died in a few days after infection or were killed before the worms matured.

In regard to natural immunity it may be concluded that among birds it is practically nonexistent, and although the albino rats seem to demonstrate some natural immunity none of the other mammals have been experimentally excluded from the list of possible hosts for this species of echinostome.

A few experiments were carried out to determine whether or not an acquired immunity to reinfection could be demonstrated in pigeons. It was demonstrated that pigeons usually lost their infections in about three weeks. In one case (Pigeon 14) in which the infection was lost after 15 days (many eggs had been present in the feces for five days) and the pigeon reinfected two weeks later, only one very immature worm was recovered on the eighth day. An abundance of cysts (100-150) had been fed both times, and the diet was not changed. These results indicated that this pigeon had either acquired an immunity to reinfection or that the diet used over the long period of time was unsuitable for the parasite.

The following is a summary of the experiments which followed this observation.

Pigeon Im 1 had been used as T2 and T4 (p. 63). As T2 it had received *per anus* 5 worms that had lived 18 days in Rabbit 7, and it carried this infection for six days. As T4 it had likewise received 10 worms 14 days old from Pigeon 18. It carried this infection for 12 days. Transfer 2 was made on 10/3/33, and the other transfer was made just two days after all of the worms had been lost. On 11/2, or eight days after the last of the T4 worms had been lost, it was fed a Physa carrying an abundance of one-day-old C7-1 cysts (see p. 63). No ova were found in the feces, although careful daily examinations were made from 11/13 to 11/22. On 11/22 it was fed 35 of the C7-1 cysts that were twenty days old. These were teased out from the Physa and administered in a small capsule. Daily fecal examinations from 12/1 to 12/15 were made and no eggs were found. The host was killed on 12/15 to determine whether the worms were present but retarded in development. No worms were present.

Pigeon Im 2 had been used as 14 and T6. As 14 it had been fed C6-12 cysts on 10/15. Ova appeared in abundance on 10/15 but disappeared on the fifteenth day (10/20). As T6 it had received *per anus* on 10/24 a single mature worm that had been in Rat 24 for 39 days. This worm lived only six days in Pigeon Im 2, and produced ova in fair abundance until it was expelled on 10/30. On 11/2 it was fed two snails, each carrying about a hundred cysts that were 1-3 days old. No ova were discovered by daily fecal examinations from 11/12 to 11/23. On 12/6 it was again fed about 125 cysts that had been in the snail host for 48 hours. Eight days later it was killed and a single very immature worm only 2.5 mm in length was recovered. This is about one-half of the length normally attained in this time.

Pigeon Im 3 had been used as T3. As T3 it had received *per anus* on 10/8, ten worms that had matured in Dog 3 (23 days). It carried these for 24 days, or until 11/1. On 11/2 it was fed a snail with about a hundred cysts one day old.

Ova were first found on 11/12 or on the tenth day and continued to the sixteenth day. No ova were found on 11/19 or thereafter. It was fed two snails each carrying about 75 cysts two days old on 12/6. On 12/16 it was killed and three very immature worms (1.6 to 1.7 mm in length) were recovered.

Pigeon Im 4 had been used as T1. As T1 it had received *per anus* on 10/2, six mature worms from Chicken 5. It carried these worms until 10/28, or 26 days. On 11/2 it was given a snail with about a hundred cysts one day old. A few ova were found in the feces on 11/18. This was the sixteenth day after feeding, which is four to six days later than normal. The ova were present in very small numbers until 12/5, or the 33rd day. On 12/6 it was again fed about one hundred cysts in a Physa and killed nine days later. It was negative.

Pigeon Im 5 was used as T10. It had received *per anus* one immature worm from Rat 26 on 10/24. The worm was expelled on 11/22, or 28 days later. This worm had been in the rat host for twelve days. On 12/10 it was fed a snail with 78 cysts that had been encysted for two, three, and four days. It was positive by fecal examination on the seventeenth day and was killed the next day (12/28). Two small mature worms were recovered.

Pigeon Im 6 was used as T5. It received one mature worm from Pigeon 19 *per anus* on 10/18 and carried it until 11/28, or for forty days. This worm had been in Pigeon 19 for 21 days. On 12/10 it was fed a small Physa with about forty cysts two days old. It was positive by fecal examination on the twelfth day and subsequently until it was killed on 12/31, the 21st day. Eleven mature worms 8 to 8.8 mm in length were recovered.

Pigeon Im 7 was used as T7. It had been given *per anus* one immature worm from Rat 26 on 10/24. The worm was expelled on 11/14, or twenty days after the transfer was made. On 12/10 it was fed a snail with 22 cysts one day old. Ova were never discovered in the feces, and it was negative when killed on 12/27.

Pigeon Im 8 had been used as T9. It received *per anus* one immature worm from Rat 26 on 10/24. The worm was expelled before it matured, probably immediately. On 12/10 it was given a snail with 150-200 cysts. Ova were not found in the stools until the nineteenth day. It was killed the same day (12/29), and a single mature worm 6.95 mm long was recovered.

At the termination of the experiments the hosts had been in captivity, held in small cages 2′ x 2′ x 3′ for the following lengths of time: Im 1, 8 weeks; Im 3 and 4, 10 weeks; and Im 2, 5, 6, 7, and 8, about 12 weeks each. These experiments are summarized in Table 4.

There is a good deal of variation in the behavior of the eight reinfected pigeons. It seems to be generally true that the rate of development is decidedly slower in the second and third infections. The average time for the appearance of positive stools in first infections is ten to twelve days, but the time required in the second infection for Im 4 was 16 days; for Im 5 it was 17 days, and in Im 8 ova appeared in the stools on the nineteenth day. However, in the case of Im 3 eggs were found on the tenth day and in Im 6 on the twelfth, which would seem to contradict the conclusion that the pigeons acquire a sort of immunity that retards development. There is still a further contradiction offered by Im 8 in which the one worm administered *per anus* in the first infection

did not remain long enough to be really considered as an infection, and yet there is no essential difference between the second infection in it and that of Im 5, whose first infection lasted for 28 days. In both cases the rate of development was about equally retarded.

The number of worms present in the first infection is ruled out as a factor influencing the second one by the fact that Im 1, which had only a total of 17 worms for a period of 18 days, was refractory to both subsequent infections; while Im 3 had carried ten worms for 24 days and several more for a second period of 16 days, and although the rate of development was somewhat retarded in the third instance, an infection was accomplished.

There is some possibility that the age and number of the cysts fed are factors influencing the character of the infections, but they are probably minor ones, since in the first infections in these experiments and in the infections used in other experiments no noticeable difference appeared in cases where cysts ranging from one day to three weeks old were used. The slight metamorphosis that occurs after the cercaria encysts is accomplished very rapidly, and the metacercariae are infective almost immediately after encystment. In Experiment 11, for example (p. 61), Rat 27 was given cysts less than 36 hours old, and 431 worms were recovered from it.

Because of such marked uniformity in the first infections in all of the pigeons used, age immunity is considered to be of very little importance. The pigeons were taken without regard for age.

While the data may be interpreted as indicating some immunity reactions it seems more probable that the diet of the hosts and the conditions under which they were maintained in the laboratory more satisfactorily explain the phenomena observed. In the case of the first and single infections the pigeons were taken from a barn where they could choose from a variety of foods, exercise freely, and follow a normal daily program. When they were brought into the laboratory they were held only long enough to determine by fecal examination whether intestinal parasites were present and to get them to feed. The diet used was fairly uniform throughout the whole series of experiments, i.e., all the hosts were given the same foods in nearly equal proportions. The diet consisted of yellow corn, wheat, oats, and a little rice. Although pronounced haemorrhagic enteritis was produced in a few cases (in the experiments to determine the general nature of infection in the various hosts), none of the pigeons used in the immunity experiments ever lost weight noticeably but appeared to maintain excellent health throughout the length of the experiments, which lasted for three months in some cases. However, there was a decided change in the character of the

feces after a month or two in captivity, which indicates that there was a change in the general condition of the host. During the first few weeks the normal stool contained considerable greenish material regardless of the type of grain fed. A few weeks later it became a pale green and finally a greyish yellow with an occasional greenish tinge. This of course varied greatly with the various individuals and in the case of Im 6 the stools continued to be greenish during the whole period. It was also noticed that some of the pigeons were considerably more restless than others, thus introducing another variable, the amount of exercise. Im 6, which was the most restless of all, was the most normal with respect to the type of stools, and was the only one to carry what could be considered a normal infection after a long period of captivity.

I do not propose to attempt an explanation of these phenomena, but in passing it is interesting to note again that the diet consisted largely of yellow corn with some wheat, oats, and a little rice. The pigeons were never in direct sunlight after they were captured and were not allowed to exercise more than was possible in the small cages. The diet was not made up with any regard for vitamins and must certainly have been deficient in vitamins A and D, and as soon as the hosts' reserves were exhausted they would be deficient from that time on to the end of the experiments. Ackert *et al* (1931a) found that chickens maintained on a vitamin A free diet developed more and larger *Ascaridia lineata* than controls that were fed on balanced diets. This was interpreted as a result of lowered resistance induced by the vitamin deficiency. The diet used in those experiments differs considerably from that used in mine, and the hosts in Ackert's experiments were irradiated. In another series of experiments (Ackert *et al,* 1931b) similar results were obtained with vitamin B free diets. In the conclusions to the latter report it was suggested that there may be a direct effect of the diet upon the parasite rather than upon the resistance of the host. That the experiments of Ackert differ greatly from mine both with respect to experimental material and methods is, of course, recognized. However, since in both cases vitamin deficient diets were involved and since they led to diametrically opposed conclusions, it is worth while to point out again that instead of the resistance of the pigeons being lowered or the parasites being benefited by the diet and lack of sunlight or exercise, the opposite was generally true in my experiments. The parasites either did not develop at all or were decidedly retarded in their development.

The transfer experiments provide some interesting facts relative to the topic of immunities in that there seems to be a definite increase in the longevity of the worms by transferring them from one host to another. These experiments have been described and summarized in

Table 9 (p. 77). The average duration for heavy and light infections alike is about three weeks for the first infections in pigeons. In six of the ten transfers the worms lived 20 days or longer in the second host, and in Transfer 3 the one worm that was taken after it had lived its expected limit in the first host, lived an additional forty days in the second. In Transfer 4, in which the infection lasted only twelve days, the pigeon which served as the second host had already been used in Transfer 2, in which it had carried five worms for six days. This would seem to show that the pigeon used in Transfers 2 and 4 had more than the average resistance and should be compared with the others on that basis.

Since no other transfers such as those described here have been made, it is not possible to broaden the conclusions by comparisons. Unfortunately, they were few in number and for that reason merely indicate rather than decide the above question as to whether or not there is a sort of accumulative immunity developed which limits the length of the parasite's life.

In all the experiments which have a bearing upon the question of immunity a good many factors have not yet come under consideration. Taliaferro (1929, p. 247), in his extensive treatise on "Immunology of Parasitic Infections," points out that age as a factor independent of all others is often an important one and more especially in cases where the host is an unnatural one. Since there are so many hosts for *E. revolutum* and quite an extensive list of both birds and mammals from which it is repeatedly taken in nature, it is hardly possible to decide whether the pigeon should be considered a natural or incidental host. There have been numerous reports of natural infections of echinostomes in pigeons (Müller, 1897; Krause, 1925a; Bolle, 1925; Zunker, 1925; Heelsbergen, 1927; Sprehn, 1927, 1930; and Picard, 1930), and while most of them were either determined only to genus, identified as new species, or species closely related to *E. revolutum,* it is more than probable that in many cases the worms dealt with are identical with *E. revolutum.* This has been discussed at length in another section (p. 51). Notwithstanding these facts it must still be maintained that the pigeon is relatively incidental as a host for this species or any other trematode parasite whose occurrence in the pigeon's gut is dependent upon its having fed upon aquatic animals or vegetation. Furthermore, pigeons as a rule spend so little time around bodies of water that it is most certainly true that the snails from which the cercariae emerged to infect the second intermediate host, which the pigeons ate in order to become infected, probably were not in the first place infected by miracidia which hatched from eggs that were dropped by pigeons. If the pigeons may be considered to be

more or less incidental hosts, then they might also be expected to show some age immunity. In a few cases it was possible to determine that the pigeons used were scarcely more than "fledglings," but in most cases it could only be noted that they were relatively old or young. Some of the lack of uniformity may then be due to this difference in age although it is probably a relatively minor factor, since there was a very striking uniformity in the pigeons that were used soon after they were captured.

Taking all of these factors into account there seems to be some evidence that although the prolonged captivity and unsuitability of the diet produce more obvious reactions which tend to obscure the immunity phenomena, the pigeon does develop a slight immunity to *Echinostoma revolutum,* as indicated by the transfer and reinfection experiments.

Discussion of Related Species and Synonyms

In the newer literature there are at least eighteen species which according to the descriptions have about 37 cephalic spines. After having made a detailed study of the diagnostic characters in a complete series of worms from a variety of hosts, I had expected to be able to clear up some of the confusion regarding these species. I am doubtful, however, whether this single attempt will lessen the number of questions regarding synonymy, for while it does reduce several of the described species to synonymy it also shows that many others that might have been carried along in the literature as valid species must now be considered doubtful until they have been reinvestigated. This in itself would not be particularly noteworthy if in all cases the type specimens were available, but this is not so in many instances.

It was my original intention to restudy the descriptions of all closely related species and possible synonyms, but after having worked at length on the older literature I was convinced that it is indeed impossible to determine from the figures and descriptions of most of the earlier authors whether they had studied this or some other species. Furthermore, if I were able to reach a conclusion regarding them I would merely add another "opinion" to the already extensive list. There is even much doubt in my mind that the laws of priority have been correctly followed in the naming of *Echinostoma revolutum.* After examining Froelich's description (Froelich, 1802) and figures of his *"Fasciola revoluta"* (see Fig. 26), I am very much less convinced than Dietz (1910, pp. 293-296) that Froelich actually studied this species. At least I cannot agree with him that he was justified in changing the name *"echinatum,"* which had already been in use for over a century. Dietz's argument merely indicates that Froelich's worm could possibly have been the same as *E. echinatum* and does not prove that it was beyond reasonable doubt

identical. *"Echinostoma revolutum"* has been accepted now and used as the correct name for this species since 1910, and irrespective of opinion regarding priority and propriety I would not propose further change in the nomenclature for *Echinostoma revolutum*. *Echinostoma echinatum* (Zeder, 1803) is then listed as the first synonym of *Echinostoma revolutum*. Other synonyms which have been generally accepted, although they have not and cannot be demonstrated as such, are *Distomum oxycephalum* Rud. 1819, *Distomum dilatatum* Miram 1840, and *Distomum armatum* Molin 1850 (see Hassall, 1896). I have examined the original descriptions of each of these and agree that they could all be identical with *E. revolutum*.

Only two of the eighteen forms which have been described as having about 37 cephalic spines can be definitely distinguished from *Echinostoma revolutum*. Eight of them are questionable species and the other eight are probably synonyms. The two species which are easily distinguished are *Echinoparyphium aconiatum* Dietz 1909 and *Echinoparyphium contiguum* Barker and Bastron 1915. The very short uterus in these forms readily separates them from all members of the genus Echinostoma, and the relative size and arrangement of the cephalic spines further distinguishes them. It is doubtful whether they can be separated from each other, however. Studies were made on numerous specimens of *Echinoparyphium* sp. from the muskrat which is the host for *Echinop. contiguum*, including two specimens identified as *Echinop. contiguum* (accessioned in the collection of the B. A. I. of the U. S. D. A. as #30851). All of them have many more than 37 spines, having 45 in some and 47 in others.

I believe that it is unquestionably true that the number and above all the arrangement of the collar spines serve as the most reliable characters for diagnosis of closely allied species. Kowalewski (1895), Dietz (1910), Tubangui (1932a, b), Rasín (1933), and many others seem to hold the same opinion. On the other hand some have argued, notably Looss (1899), that there is too much variability in the spines to permit diagnosis on this basis. Looss, however, did not consider the arrangement of the spines in more detail than the alternating rows and grouping of the spines of the ventral lappets. Other characters which have been used are the extent of the vitellaria and the location of the worm in the host. The character of the vitellaria is too variable with age to be of value, and the position within the host although fairly constant for a species is altered very considerably by the number of worms present and the length of time elapsed between the host's death and the autopsy. The presence or absence of cuticular spines also depends upon the latter condition since dead worms frequently slough off the whole cuticula. The

stoutness of the body and general shape depends too largely upon the host, age, diet of the host(?), time of autopsy, and type of fixation to be of very great value in diagnosis. The most constant characters have already been described in another section (p. 18). As a result of the present study the following characters have been adopted as of diagnostic value: number of collar spines; relative size of the various spines; their arrangement in the $(2+3)+6+(8+7)+6+(3+2)$ fashion; the length of the uterus; and the relative size of the two suckers and the pharynx. In my opinion the host species is not to be regarded as important in this parasite.

In studying the descriptions of closely allied forms, the above characteristics have been used as a basis for conclusions regarding relationship. Other characteristics such as size of the body, eggs, and gonads have also been noted. The eight doubtful species mentioned above are those that have been described as having 37 spines, but the data given is insufficient to separate them from or identify them with *Echinostoma revolutum;* or, as in the case of *E. sudanense,* the description and the figures do not agree and one or the other seems identical with this species. In addition to *E. sudanense* Odhner, 1911, the doubtful species are: *E. acuticauda* Nicoll, 1914; *E. callawayensis* Barker and Noll, 1915; *E. erraticum* Lutz, 1924; *E. neglectum* Lutz, 1924; *E. microrchis* Lutz, 1924; and *E. echinocephalum* (Rud., 1819). These species of dubious standing and those that are shown by this study to be identical with *E. revolutum* will be discussed in detail.

Echinostomum sudanense, Odhner, 1911 (? = *E. revolutum*).—*Echinostomum sudanense,* described by Odhner (1911, pp. 116-117, pl. 3, figs. 12, 13), is based upon a single mature specimen from the rectum of a "Sattelstorch," *Ephippiorhynchus senegalensis,* and an immature specimen from the intestine of *Scopus umbretta* in North Africa. It is 6.5 mm long, cuticula spiny on the ventral side only, uterus long and the eggs numerous; length of pharynx equal to the diameter of the oral sucker; oral sucker about one-third the diameter of the acetabulum. All of these characters agree with specimens in my own collection taken from ducks. The collar spines are described as "31 schlanke und spitz Kragenstacheln sind vorhanden, von denen 4 in jeder der ventralen Ecken sitzen und die übrigen 23 in einer doppelten, ununterbrochenen Reihe angeordnet sind." His figure 13, which I have copied (Fig. 27), shows 33 instead of 31 spines and the arrangement is strikingly similar to that of *E. revolutum.* The corner spines, which Odhner described as being four, are distinctly five on the left side of the figure, and on the right side the most lateral of the three oral corner spines is shown over on the adjacent one. This specimen was flattened under a cover glass and the spines somewhat

displaced, but it still resembles *E. revolutum* so closely that I am of the opinion that some of the spines were lost and that *E. sudanense* is really *E. revolutum.* There are at least two points on the figure where spines seem to be absent (cf. Fig. 27, 9; and Text-figs. 1-5). It is possible that it is a distinct species, but since the description is based on a single mature flattened specimen and the corner spines are so nearly identical with *E. revolutum* as are also the others in the unalternating laterals and alternating dorsals, it should be considered a probable synonym until more information on it is obtained.

Echinostoma acuticauda Nicoll, 1914 (? = *E. revolutum*).—This species was described by Nicoll (1914, pp. 110-111, pl. 6, fig. 4) from the straw-necked ibis, *Carphibis spinicollis.* It is a 37-spined form which from the description alone cannot be differentiated from *E. revolutum.* Nicoll states that *E. revolutum* is "altogether a much broader and bulkier form." It has already been pointed out that *E. revolutum* is bulkier and stouter in ducks than in other hosts and that it is quite delicate and slender in some. As these specimens were taken from the ibis they might be expected to differ from the ones seen in the duck. The figure of Nicoll's species resembles very much specimens which I have taken from the guinea pig or specimens taken from any host after they have been dead for some time. It is probably not as similar to *E. revolutum* as the description indicates, but since the cephalic spines are not described it is not clearly a distinct species.

Echinostoma callawayensis Barker and Noll, 1915 (? = *E. revolutum*).—This species was described from a collection of parasites from the muskrat, *Ondatra zibethica,* from Nebraska. It is described as having 37-41 cephalic spines; 31-33 on the rim and 2-5 on each flap. This variability in number suggests at least one of the two following conditions: either the material was in such poor condition that the spines were lost on some of them, or more than a single species was used for the description. I have not been able to examine Barker's material, but the figures and description lead me to the conclusion that small *E. revolutum* and *Echinoparyphium* sp. were both used in describing *E. callawayensis.* There has been only one record of this species since it was described (Law and Kennedy, 1932). Unfortunately the cephalic spines are not included in the description, and no specimens could be procured. In the figure given by Barker the posterior end is very much flattened so that the testes appear to be very large and the post-testicular region much shorter than normal for *E. revolutum.* The whole of the body looks more like Echinoparyphium than Echinostoma. The figure given by Law and Kennedy (1932, fig. 10) is, on the other hand, distinctly an *Echinostoma* sp. If *E. callawayensis* is a valid species it must at present be separated

from *E. revolutum* on very unreliable characters, particularly since the description is made from compressed specimens. I am not able to distinguish this species from related forms. In his key to the parasites of the muskrat, Barker (1916, p. 183) separates it from the other species on the compactness of the uterus. Since this character depends upon the degree of maturity and extent of contraction it is not safe to use it for diagnosis. Law and Kennedy (1932, p. 19) do not make any statement regarding the distinguishing characters.

Echinostoma erraticum Lutz, 1924 (? = *E. revolutum*).—Lutz, (1924) described a number of new echinostomes from South America, several of which are described as having 37 cephalic spines, and the collar of spines is left entirely undescribed in one of them. Four of these species are probably synonyms of *E. revolutum*. *Echinostoma erraticum* is so named because of its showing so little choice in its hosts, which is very definitely a characteristic of *E. revolutum*. Lutz found that *Physa rivalis, Planorbis* sp., and *Spirulina* sp. serve as hosts for the metacercaria, and if these are fed to pigeons, *Gallinula galeata, Chamaepelia passerina,* or *Aramides cayennensis,* adult worms may be recovered from each of them. He also found natural infections in *Creciscus viridis, Nycticorax violaceus,* and *Crotophaga ani.* He found specimens up to 8 mm in length, and his figure shows no characters not found in *E. revolutum.* The only statement regarding the cephalic spines is that the number varies from 35 to 39, with 37 as the usual number. In this same paper Lutz reports researches upon *E. revolutum* but does not make any statement regarding the specific characters by which he recognizes each of the several 37-spined species. I cannot find any characters to distinguish it from other species. The redia is also similar to *E. revolutum,* having a short gut and posterior appendages. According to the description, the 37-spined cercaria is in every respect identical with *Cercaria Echinostomi revoluti.*

Echinostoma neglectum Lutz, 1924 (? = *E. revolutum*).—This species looks very much like *E. revolutum* in the figures, but the description does not include any of the important diagnostic features. It is stated (Lutz, 1924, p. 87) that it can be distinguished from *E. erraticum* by the shape of the testes. As the shapes figured for both species are common in *E. revolutum* it is doubtful if *E. neglectum* is a distinct species.

Echinostoma nephrocystis Lutz, 1924 (? = *E. revolutum*).—*Echinostoma nephrocystis* is a form with which Lutz was able to infect tadpoles. He fed these to *Aramides cayennensis* and recovered the "new species" with 37 spines. The description is too incomplete to allow a decision as to the specific characters for it. Lutz seems to consider the host a distinguishing character. He says that it is most similar to Dietz's *Echin-*

ostoma distinctum which is a form with 35 rather than 37 spines. There is no question about its being a doubtful species and there is a very great probability that it is identical with *E. revolutum.*

Echinostoma microrchis Lutz, 1924 (? = *E. revolutum*).—This is another of Lutz's 37-spined species. The description of it is based upon several young specimens from *Gallinula galeata* and one mature one from *Creciscus viridis.* It is characterized as follows:

Der ausgewachsene Wurm enthaelt zahlreiche Eier. Seine Laenge betraegt nur 5 mm bei einer Breite von 1 mm. Der Stachelkranz enthaelt 37 Stacheln in der gewoehnlichen Anordnung; vom freien Ende aus gezeehlt, ist der fuenfte der laengate. Stachelschuppen sind zahlreich und auffallend bis auf die Hoehe des Ovariums, nachher werden sie seltener und undeutlicher.

This description fits exactly the average 5 mm *E. revolutum* from birds, particularly those that are contracted or slightly flattened. The statement regarding the longest spine is especially characteristic of *E. revolutum;* in fact in every one of several hundreds of specimens examined the most lateral one of the aboral corner spines is the longest (Text-figs. 1, 3, 5). He says further:

. . . . die neue Art unterscheidet sich leicht von allen beschriebenen, da sie selbst in erwachsenem Zustande sehr kleine Hoden besitzt. Dagegen ist das Ovarium bedeutend grosser und voluminoeser, als bei verschiedenen ashnlichen Arten. Es ist kugelig und liegt mit seinem Hintertande dem mittleren Querdurchmesser auf. Die Hoden liegen ziemlich weit zurueck, aber nahe hintereinander. Die Vitellarien sind lang und anastomosiren briet zwischen dem zweiten Hoden und den Enden der Blinddaerme.

Lutz's figure of this species shows the one mature worm which he collected, and it is clear that the specimen is somewhat flattened and comparisons of relative sizes of the parts are therefore uncertain. In spite of this, however, it is clear that the specimen is indeed different from the average *E. revolutum.* I have never seen a specimen whose ovary is one and one-half times larger than the testes, but a number of the barely mature specimens have all three of the gonads about equal in size. The testes of Lutz's worm are considerably larger than the pharynx, and the uterus contains about 150 eggs. I have specimens of *E. revolutum* which also have small testes and consider them as slightly more abnormal than Lutz's worm. The testes in one of these specimens are slightly smaller than the pharynx and the uterus contains fifteen eggs. The ovary in this specimen is only slightly smaller than the testes. Since Lutz found but one mature specimen of this type and such abnormalities in the gonads are known to occur, I believe that the similarities in his specimen and *E. revolutum* are striking enough to almost, if not actually, establish their identity. If it is shown that a form exists in which the testes are consistently smaller than the ovary, then it may be distinguished from *E. revolutum* on the basis of that character.

Echinostoma echinocephalum (Rud., 1819) (? = *E. revolutum*).—*Echinostoma echinocephalum* is also a 37-spined species. It was originally described by Rudolphi (1819) and redescribed from the type material by Dietz (1910, p. 303, Pl. X, fig. 4, text fig. C). The specimens are immature, only 2.7 to 3.3 mm in length. The width of the head crown is 0.23-0.27 mm and body width at the level of the acetabulum is 0.47-0.60 mm. The head crown contains 37 spines described by Dietz as follows:

Davon liegen wie bei *Echinost. revolutum* 27 Stacheln in einer doppelten Reihe am Rande des Kragens; dieselben sind gleichgross, 0.0544-0.0720 mm lang und 0.012-0.0144 mm briet. An den beiden Enden dieser Stachelreihen befinden sich beiderseits in den ventralen Ecklappen noch je eine Gruppe von 5 Stacheln, von denen je 2 paarweise übereinander gelagert und ein fünfter einzelner Stachel median neben dem innern Stachel der vordern Reihe dieser Gruppe gelegen ist. Diese Eckstacheln sind im Gegensatz zu *Echinost. revolutum* (bei denen sie die gleiche Grösse besitzen wie die Randstacheln) kleiner als die Randstacheln; ihre Länge beträgt 0.048-0.060 mm, ihre Dicke 0.01-0.012 mm. Eine Hautbestachelung konnte ich bei keinem Exemplar nachweisen.

The contrast which Dietz describes in the spines of *E. revolutum* and *E. echinocephalum* is very slight when actually compared. That the corner spines are slightly smaller (54-72 μ, by 12-14 μ as compared with 48-60 μ by 10-12 μ) than the others is really no contrast, for contrary to Dietz's statement it is the usual condition in *E. revolutum*. Dietz's figure "A" (see Pl. III, fig. 18) shows a very considerable addition to the difference given in his text. The measurements given in the text overlap both in length and width, and they are very near the mean length for the spines of *E. revolutum* at the same body size (Chart 6). If the distribution of the spines is as inaccurately figured as the size of the corner spines it can be assumed that no difference is shown in the cephalic spination in the two species. The figure shows a character that would distinguish *E. echinocephalum* but Dietz does not mention it. He has shown a distinct alternate arrangement in the lateral spines, a condition that does not occur in *E. revolutum*. If the figure could be accepted as reliable there would be no reason to discuss this species further for it could be distinguished from all of the other 37-spined forms of the genus by the character of these lateral spines. Since, however, as was shown above, the figure does not support the text and practically none of the written description actually disagrees with my own observations on specimens of *E. revolutum*, and furthermore, since only seven immature specimens were used for the description, and no one has since reported finding this species, it is very probable that *E. echinocephalum* is identical with *E. revolutum*. The measurements given by Dietz are added here since they do not agree in every detail with *E. revolutum:* oral sucker 90-120 μ in diameter; pharynx 100-120 μ by 80-110 μ; acetabulum

0.37-0.44 mm in diameter. These measurements agree with corresponding ones for *E. revolutum* with the exception of the acetabulum, which is slightly larger than the same organ in unflattened specimens of *E. revolutum*. The figure given by Dietz (1910, Pl. X, fig. 4) appears slightly flattened although it is not so designated. Zunker (1925) reexamined the type material and confirmed Dietz's observations. His only significant remarks are that he could see no cuticular spination and that the acetabulum is large. Although I am not convinced that *E. echinocephalum* is distinct from *E. revolutum,* it is possible that it differs in the following characters: smooth cuticula, acetabulum four times larger than oral sucker while in *E. revolutum* it is only two to three times larger than the oral sucker in immature specimens. The six lateral spines are alternating instead of being unalternating, and the corner spines may be somewhat smaller in relation to the other cephalic spines.

Echinostoma miyagawai Ishii, 1932 (= *E. revolutum*).—This is a 37-spined form which Ishii (1932, pp. 91-100, Pl. 2, figs. 10, 11; Pl. 1, fig. 1) described from the domestic fowl and wild and domestic ducks in Japan. It is described as being 21-26 mm long by 2-3.5 mm wide. The head crown is 0.89-1.10 mm in diameter, and the 37 spines are arranged exactly as in *E. revolutum* in the figure. In the text, however, it is stated that six spines lie on each ventral side of the crown and the other 25 are arranged around the crown in two rows. It is clearly shown in the figure that only five spines occur on the ventral corners, the six laterals do not alternate, the fifteen remaining spines alternate across the dorsal side of the head, and the median spine is aboral as it is in *E. revolutum* (Figs. 22, 23). The figure also shows that the relative size of the spines is identical with *E. revolutum*. They are 84-108 μ in length and 26-28 μ in width, which also checks with *E. revolutum*. The oral sucker is 0.221-0.476 mm in diameter, the acetabulum is 0.714-1.088 mm and is 2.0-2.3 mm posterior to the oral sucker. The cuticular spines are closely set in the anterior part of the body but more scattered posteriorly. The diameter of the acetabulum is slightly less than the average for *E. revolutum* but is just within the range. The relative size of the two suckers is the same for the two forms and the diameter of the collar is also the same. The length of the pharynx is equal to the diameter of the oral sucker in both forms, and the internal organs of the two are identical throughout. Ishii says that his species resembles *Echinostoma cinetorchis* in body size and number of crown spines but differs from it in the character of the testes. Ishii's figures show the testes distinctly lobate while his figure of *E. revolutum* shows more rounded testes. It has already been pointed out that under some conditions the testes of *E. revolutum* are also lobate, more so than is shown in the figure for *E.*

miyagawai. Since there are no other characters, with the possible exception of the size of the acetabulum, by which to separate the two forms it seems very probable that they are identical. Ishii's *E. miyagawai* is merely a description of the very large specimens of *E. revolutum.* It is important to note that Ishii does not describe the individuals of his species that are less than 21 mm in length, and I am sure that a 10 mm specimen of *"E. miyagawai"* could not be distinguished from a 10 mm specimen of *E. revolutum.**

Echinostoma cinetorchis Ando and Ozaki, 1923 *(= E. revolutum).*—*Echinostoma cinetorchis* is a 37-spined form which Ando and Ozaki found in the intestine of wild rats in Japan (Dollfus 1925, pp. 85-91, text figs. 1-3). It is a form that reaches a length of 21 mm and a corresponding width of 3.7 mm in flattened specimens. The oral sucker has a diameter of 0.26-0.30 mm in average specimens and 0.31-0.38 mm in the larger ones. The distance between the two suckers is 1.75-2.18 mm (about one-sixth of the body length) in average specimens and 2.29-2.95 mm (about one-ninth of the body length) in larger specimens. The acetabulum is 0.72-0.73 mm in diameter in average specimens and 1.01-1.18 mm in larger ones. Cuticular spination reaches the region of the posterior testis. The cephalic crown is 0.44-0.53 mm wide. The arrange-

*Two specimens and a communication from Ishii have been received since the above was written. The following notes are based on a study of them. Specimens 12.8 mm and 15 mm in length. Cuticula entirely lacking on both. On the smaller one there are thirty-seven cephalic spines arranged exactly as in *E. revolutum;* the relative size is typical and the actual size is near the average for specimens taken at delayed autopsy where they usually are shorter with points more blunt than the normal ones. The actual and relative size of the suckers, pharynx, and collar agree with *E. revolutum.* Vitellaria, uterus, ova, ovary, and cirrus also agree in general character, size and position. The testes, however, are more deeply lobed than any I have previously seen in this species, being constricted into four very definite lobes. The larger specimen has been fixed under considerable pressure and appears to have been dead in the host longer than does the smaller one. The cephalic spination does not differ from that in *E. revolutum* and as far as can be determined all other organs are identical with it. The testes are less deeply lobed than those in the smaller specimen but more so than is usually seen in *E. revolutum.* I have, however, several specimens in my collection with more deeply lobed testes, and I do not separate them from "sister" individuals on the basis of this character. In view of the fact that the cuticula is lacking on both specimens and the cephalic spines are rather blunt it seems evident that post-mortem changes must be taken into account here. The tissue's reaction to the stains support this view.

Through a personal communication Ishii has pointed out that he believes that *E. miyagawai* has a very long neck, longer esophagus, greater head crown, and longer crown spines than *E. revolutum.*

It is possible that a study of specimens that were fixed while still active would alter my conclusion, which is at present that the two are identical.

ment of the cephalic spines is the same as in *E. revolutum.* In average specimens the dorsal spines are 55-59 μ long by 11-13 μ wide. Those of the end groups are slightly smaller, 36-38 μ by 10-11 μ. The figure (see Fig. 15) of the crown shows that the relative size of the various spines is the same as for *E. revolutum.* The internal organs are apparently identical also. The egg size is 96-100 μ by 61-70 μ, which is about the same as for the largest specimens of *E. revolutum.* The testes are smooth or slightly lobed and are somewhat variable in position, the anterior testis being in some cases on the level with the ovary or even anterior to it. I have not observed any such extreme anomolies in my specimens of *E. revolutum.* Ando and Ozaki do not compare this form with *E. revolutum* or any of the other very numerous 37-spined species of the genus. Very extensive tabular comparisons of the descriptions and comparisons of every detail indicated in the figures fail to show a single difference in *E. cinetorchis* and *E. revolutum.* It is interesting and important to observe that both Ishii (1932) and Kurisu (1932) reported finding both species in the same region. The only assumption necessary to account for this, however, is that they have been misled by the very pronounced changes in body proportions that always occur in *E. revolutum* as its length increases over 12 mm.

Echinostoma armigerum Barker and Irvine, 1915 (= *E. revolutum*). —This is one of the four species of muskrat echinostomes described by Barker *et al* as having about 37 cephalic spines. Two of these species, *Echinoparyphium contiguum* and *Echinost. callawayensis,* have already been discussed. *Echinostoma armigerum* has been reported recently from the muskrat in Canada, and a brief description of it given by Law and Kennedy (1932, p. 24, fig. 15). Although both authors report it as common I have not been fortunate enough to obtain specimens for comparison with specimens in my own collection which seem to be identical with *E. armigerum* and are at the same time identical with *E. revolutum.* This form according to the original description (Barker, 1915, p. 189, text-fig. A) is 9.4 to 12.4 mm long by 1.2 to 1.8 mm wide. Cephalic spines 37 in number, 27 on the rim, and 5 on each ventral point of the collar. Collar spines 61 to 94 μ in length, and the ventral corner ones are the smaller. The anterior third of the body spiny. Uterine coils fairly compact in transverse coils. Eggs 84 to 105 μ by 57 to 66 μ. All other parts of the description would apply to almost any species of the genus. The figure (Barker's text-fig. A) so closely resembles a large number of the specimens in my own collection that I am convinced that they are identical and that Barker's *E. armigerum* is *E. revolutum* after it has reached a length of 9.4 to 12.4 mm. Comparison of the measurements for *E.*

revolutum and all points of description fail to show any difference in the two forms. *E. armigerum* is doubtless a synonym for *E. revolutum.*

Echinostoma coalitum Barker and C. A. Beaver, 1915 (= *E. revolutum*).—The fourth species of muskrat echinostome described by Barker is *E. coalitum.* It is also identical with *E. revolutum* and the same as *E. armigerum* but described from only the older and larger individuals. It is difficult to take muskrats alive and in most cases autopsies are made several hours post-mortem. The large specimens of *E. revolutum* die much sooner than the smaller ones and consequently most of the largest specimens in my collection were taken after the worms had died. I have, however, several specimens over 20 mm in length with firm bodies and good spination. The original description of *E. coalitum* was based upon "22 specimens of an unusually large trematode . . . . found among several hundred specimens of different species of trematodes in the intestine of 46 muskrats." The body is described as "very flabby," which suggests that the specimens were dead when collected. They are 22 to 30 mm long by 1.5 to 2.3 mm wide at the level of the ovary. The anterior part of the body is covered with minute spines. The collar bears "35 spines arranged in a single or slightly alternate rows, 25 large spines on the rim and 5 smaller spines on each lappet." Oral sucker 0.37 to 0.46 mm in diameter. Acetabulum at the level of the second anterior sixth of the body, 1.12 to 1.32 mm in diameter. Uterus with dense transverse coils. Eggs numerous, 104 to 108 μ by 67 to 70 μ. When this form is compared with *E. revolutum* there is but one significant difference in the two. That is in the number of spines in the collar, there being only 35 instead of 37. There are, however, five on each corner lappet, and since most of the specimens studied were in all probability dead before fixation, there is a very strong probability that the 22 specimens studied did not show typical spination. Through the kindness of E. W. Price of the B. A. I. of the U. S. D. A., I have been able to study a slide (#30850) containing three specimens which were accessioned in the Washington collection under the name of *E. coalitum.* These specimens are 16 to 20 mm in length and identical with specimens which I have collected from the muskrat and have identified as *E. revolutum.* Although none of the three specimens on slide #30850 have a complete set of cephalic spines and all have lost the cuticular spination, it is evident from the pattern of the spines still present that they are the 37-spined *E. revolutum.* Law and Kennedy (1932, p. 16, fig. 9) report *E. coalitum* from the muskrat in Ontario, Canada. The description and figure given by them are about the same as Barker's. Leidy (1888, p. 126) also reported on an *Echinostoma* sp. from the muskrat. He had two collections, and in the

first one he says "there are 25 worms, and in their present condition they are pale brown bordered by dark brown, and measure from 12 to 18 mm long by 1 to 1.5 mm broad. If not identical, they are closely allied to *Distomum echinatum,* which in the mature state lives in ducks and other water birds and in the larval state in fresh-water snails. The muskrat eats the latter so that it may likewise become infested in the same manner as the ducks and this would also seem to make it probable that the parasite is the same." Leidy characterizes them as follows: 36 straight spines on the collar. Acetabulum much larger than the head, "situated at the base of the neck." The oral sucker is small and the pharynx oval. Body nearly the same width throughout. Testes almost midway between acetabulum and posterior end. Head 0.5 to 0.6 mm broad, spines about 100 by 20 μ, oral sucker 0.25 mm in diameter, pharynx 0.30 by 0.225 mm, and acetabulum 0.875 to 1.25 mm. Ova 100 by 72 μ. Regarding the second collection Leidy states: "Since the above communication a collection of worms, from the small intestine of a muskrat, has been received. Eighteen of the worms pertain to the supposed *Distomum echinatum,* and range from 18 to 25 mm long. In all, the fore part of the body a short distance behind the acetabulum is finely echinate, while the rest is smooth." Leidy did not report any observations on the cephalic spination of the second collection, but it is very probable that 37 instead of 36 spines could have been counted on these since they were, judging from the fact that the cuticular spination was intact, in a much better state of preservation than the first lot. Assuming that the spines on the first lot were atypical, the worms reported by Leidy can be considered correctly diagnosed by him as *Distomum echinatum (= Echinostoma revolutum).* It is perhaps important to note too that *Distomum echinatum* was at the time Leidy made this report sometimes characterized as having 36 rather than 37 cephalic spines.*

Echinostoma mendax Dietz, 1909 *(= E. revolutum).—Echinostoma mendax* is a 37-spined form that is so similar to *E. revolutum* that it has already been considered a doubtful species (Lutz, 1924, p. 81). Sprehn (1932, p. 306) lists it as a synonym of *E. revolutum* but does not indicate whether he has himself re-examined the types or not. It is obvious from the original description that it cannot be distinguished from *E. revolutum.* The only differences noted by Dietz himself are expressed in the following statements:

*Krull (1935) has recently made a brief report on a form which he has called *E. coalitum.* Since the morphology of the worm is not discussed, no confirmation of the above conclusion is given, although certain host records have been confirmed and certain others added (Tables 1, 2, and 3).

In der vordern Körperpartie ist die Cuticula sowohl auf der Dorsal—wie auf der Ventralseite mit feinen Hautstacheln besetzt, die sich jedoch im Gegensatz zu *Echinost. revolutum* auch in den Seitenrändern nicht über den Bauchsaugnapf nach hinten verfolgen lassen. . . . Der Bauchsaugnapf ist in Verhältnis zu *Echinost. revolutum* nur schwach entwickelt, kugelig (0.60-0.64 mm) oder ein wenig nach hinten verlängert (0.65-0.72 : 0.60-0.62 mm).

It has been shown that the cuticular spination varies with both the age of the worm and the type of host, and differences in it are unreliable for diagnosis (Chart 8). That there is a difference in the development of the acetabulum in the two forms is doubtful, for the measurements given by Dietz fall very near the average for *E. revolutum.* The figures given by Dietz (1910, text fig. A, Pl. 10, fig. 2) are identical in all the important diagnostic features with *E. revolutum.* In fact text figure A (see Pl. III, Fig. 16) shows very distinctly both the typical arrangement of the spines and the typical order in the relative lengths of the various spines. *Echinostoma mendax* Dietz 1909 is without question identical with *Echinostoma revolutum.*

Echinostoma paraulum Dietz, 1909 (= *E. revolutum*), and *Echinostoma columbae* Zunker, 1925 *(= E. revolutum).—Echinostoma paraulum* was desecribed by Dietz (1909, p. 300, Pl. 10, fig. 3, text-fig. B) from *Colymbus cristatus* and several species of ducks from middle Europe. Several authors (Baylis, 1929, p. 40; Iwata and Tamura, 1933, p. 2) have expressed strong doubts as to its legitimacy, while others (Skrjabin, 1915, p. 399; Sprehn, 1927, p. 454, 1930, p. 767, 1932, p. 314; Wetzel, 1933, p. 772) report its occurrence and consider it a valid species. Sprehn (1927, 1930, 1932) considers *E. columbae* Zunker 1925 a synonym of *E. paraulum,* and in 1930 he put it into the genus Echinoparyphium, although I am doubtful if this can be justified. The situation is made still more complex by the fact that *E. columbae* Zunker is variously described by Zunker (1925), Bolle (1925), Krause (1925a, b), Otte (1926), and Sprehn (1927, 1930, 1932). According to Dietz's original description *E. paraulum* is a form that ranges in length from 4 to 7 mm. Its other measurements are: greatest width 0.94 to 1.66 mm, width of head crown 0.43 to 0.54 mm, diameter of oral sucker and acetabulum, respectively 0.21 to 0.26 mm and 0.54 to 0.83 mm, pharynx 0.21 to 0.22 mm by 0.14 to 0.20 mm. The body is spiny as far posterior as the acetabulum on the dorsal side and to the testes on the ventral side. The 37 spines of the cephalic collar are arranged exactly as in *E. revolutum* and the relative size of the various spines is also identical with *E. revolutum* (Fig. 17). His figure of the entire worm is in all respects identical with a slightly contracted 6-7 mm specimen of *E. revolutum.* The middle constriction in the testes occurs not uncommonly in this species, particularly if the

specimen is either under pressure or fixed in the contracted condition. There is positively not a single character by which *E. paraulum* can be separated from *E. revolutum.*

Echinostoma columbae has been restudied by Sprehn and shown to be a synonym of *E. paraulum,* which probably is sufficient evidence that both *E. paraulum* and *E. columbae* are identical with *E. revolutum.* Since, however, the several descriptions of *E. columbae* differ, and especially since Sprehn (1930) has put this form into the genus Echinoparyphium a more complete study of the descriptions is necessary.

Krause (1925a) made a preliminary report on a number of pigeons infected with echinostomes of two species, a 37-spined form and one with forty-two spines. Then Zunker (1925) published a description of a 37-spined echinostome from the pigeon, which he considered closely related to *E. echinocephalum* but distinct from it. He called it *Echinostoma columbae* spec. nov. Then Krause (1925b) followed Zunker with a second report on his material. He described his specimens in good detail and considered it similar to *E. revolutum,* which it is indeed. Krause's description leaves no doubt in my mind that both his and Zunker's worms are *E. revolutum.* Zunker could find no cuticular spines on his worms, but Sprehn (1927) re-examined Zunker's worms and found them identical with a collection which he himself described as having a distinct cuticular spination over nearly all of the ventral side and the dorsal side of the anterior end. Bolle (1925) also corrected Zunker's observation regarding the cuticular spination. It was Bolle who first compared the pigeon echinostomes with *E. paraulum,* but he decided against their identity on the basis of differences in size of the crown spines and the position of the cirrus sac. Bolle described the dorsal spines of the head crown as being larger in the aboral row than in the oral. This feature prompted him to compare it with members of the genus Echinoparyphium. He could distinguish it from all members of that genus and decided that it was indeed a distinct species which should be designated as *Echinostoma columbae.* There is very little disagreement among the various authors that cannot be readily explained on the basis of differences in degree of maturity in the specimens studied by them. Zunker and Bolle both used smaller specimens while Krause and Sprehn had about average specimens. Sprehn emphasized the size difference in the spines of the two dorsal rows and observed (1927, p. 452) that "eine deutliche pars prostatica lässt sich in Schnitten nachweisen." He compared his specimens with the types for *E. paraulum* and found that they were identical. On the basis of these studies he declared all of the pigeon echinostomes with 37 cephalic spines to be

E. paraulum (= E. columbae). Krause, however, argued that these worms were most similar to *E. revolutum.* In spite of the fact that he had in a former communication declared that a pars prostatica was distinctly present in the worms studied by him, Sprehn (1930) offered the following reason for transferring them to the genus Echinoparyphium:

. . . . bei ihm die Pars prostatica fehlt, die Hautbewaffnung die ganze Bauchseite überzieht und sich auch noch auf die Rückenseite ausdehnt und ausserdem ein doppelter Stachelkranz vorhander ist. Bei den Arten der Gattung Echinostoma s. str. ist die Bestachelung dagegen nur auf die Bauchfläche des Vorderkörpers beschränkt und ausserdem eine Pars prostatica vorhanden.

That the two genera Echinostoma and Echinoparyphium cannot be separated on the basis of the cuticular spines is evident since all of the most recent researches on *E. revolutum* have shown that at least some dorsal cuticular spination is present. It is furthermore evident that Sprehn is in error in considering the crown spines of the pigeon echinostomes as more typical of the genus Echinoparyphium, for all of the figures given by himself (1927, fig. 1), Zunker (1925, fig. 2), and Bolle (1925, fig. 1) picture the head crown as being typical of Echinostoma, not Echinoparyphium (Figs. 19, 20, 21). They show a very slight size difference in the two rows, a condition characteristic of *E. revolutum* (Text-figs. 1, 2, 4, p. 24). The figures of both Bolle and Sprehn are drawn in a manner that emphasizes this difference, since the oral row is more vertical than the aboral, and spines thus seen in end view appear to be shorter. Were it not for the question of the pars prostatica, all of the pigeon echinostomes described by the above authors could be readily put into the genus Echinostoma s. str. and considered as unquestionable synonyms of *E. revolutum.* In 1927 Sprehn declared that in *Echinostoma paraulum (= E. columbae)* the pars prostatica is distinctly seen in sections. Three years later he contradicted his earlier statement and stated that "die Pars prostatica fehlt." The only figure of an entire specimen of one of these disputed forms is given by Zunker (1925, fig. 1). The length of the uterus alone prohibits putting it into the genus Echinoparyphium (Fig. 24). If this form can be distinguished from *E. revolutum* at all it is on the basis of Sprehn's later statement that the pars prostatica is absent.

Van Heelsbergen (1927) and Picard (1930) have also reported finding echinostomes in pigeons, but neither has made a final identification of his material. The figures and data given by them suggest that each found both *E. revolutum* and *Echinop. recurvatum.*

Misdeterminations.—In addition to the large number of doubtful species and synonyms there are a few obvious misdeterminations that should be mentioned. Looss (1899, p. 681) has already called attention

to the misdetermination by Wedl (1857, pp. 245-246, Pl. 1, figs. 5-6). Wedl's form which was taken from *Phalocrocorax carbo* is not even closely related to *Echinostoma revolutum.* A form which Ercolani (1881, 1882) determined as *E. echinatum* obviously does not belong to the genus Echinostoma s. str. His figures (1881, Pl. 1, figs. 50-58, Pl. 2, figs. 1-5; 1882, Pl. 3, figs. 19-21) were copied by Railliet (1895, fig. 244) and designated as *D. hérissé (D. echinatum* Zeder*).* Neumann (Neumann-Macqueen, 1905, fig. 237) also copied these figures and put them under the name *Echinostomum echinatum.*

Johnson (1920, p. 368) has doubted Looss' determination of a form which he found in *Machetes pugnax* (1899, pp. 683-684, Pl. 24, figs. 6, 7). Looss described it as a variation of *Echinostoma revolutum.* Several of the crown spines had been lost but the exact point where some of them had been could be located, so that a total of 34 spines was accounted for. Since this form did not differ in other respects from the *E. revolutum* of other hosts Looss did not make a new species of it. Johnson was of the opinion that the host in this case was so different from the "normal" that it might be expected to be a new species of worm. He argued also that the spination in the two forms (those from ducks and those from *M. pugnax*) is very different, a point on which I cannot agree with him. The fact that only 34 spines are shown does not mean that the form is a 34-spined species. Looss made it clear in the text that the spines were not all present and that only 29 were actually observed. The difference in the size range in the eggs was also a point in Johnson's argument, but this is not, in my opinion, significant, since the measurements given by Looss (101 to 110 μ by 59 to 63 μ) are easily within the range for *E. revolutum.* Taking all of these facts into consideration there can be little doubt that Looss has correctly identified his material and that *Echinostoma limicoli,* the name which Johnson suggested for Looss' worm, is a synonym of *E. revolutum* (cf. Figs. 28, 29).

Looss' figure of the metacercaria of *E. echinatum* (1894, Pl. 9, fig. 192) is, on the other hand, a misdetermination. Sewell (1922, p. 122) has suggested that Looss' figure (1894, fig. 191) of the cercaria is also a misdetermination because of the difference in the excretory system of it and the young adult. Judging solely on the basis of the cephalic spination there can be no doubt that the figure of the metacercaria is not that of *E. revolutum.* It is instead very similar to *Echinoparyphium aconiatum* Dietz, a form which also has 37 spines. The corner spines in Looss' figures are four in number, all nearly equal and very much larger than the other crown spines. These characters are present in *Echinop. aconiatum* but not in *Echinost. revolutum.*

Summary of Forms Discussed.—The present status of the forms discussed and the conclusions the present study has led to may be summarized as follows: *Echinostoma revolutum* can be distinguished from all other described forms by the following characters: (1) 37 cephalic spines arranged in the manner indicated by $(3+2)+6+(7+8)+6+(3+2)$, which means that there are 5 on each ventral lappet, 3 of which are on the surface or oral in position and 2 deeper in the flesh or aboral, 6 unalternating laterals, 15 dorsals arranged in alternating fashion with 8 orals and 7 aborals. The median spine is in the aboral row which is continuous with the row containing the six lateral spines. (2) The ventral side of the body is spiny as far back as the anterior testis or farther, and the dorsal side bears spines in most specimens but may be without them in older worms from bird hosts. (3) The uterus is long, containing eight or more coils even in the smallest mature worms. (4) The proportions of the body change with age as indicated in Charts 1-7.

Echinoparyphium aconiatum and *Echinoparyphium contiguum* are distinguished from *Echinostoma revolutum* by the very short uterus and the different arrangement of the cephalic spines, *Echinop. aconiatum* having an arrangement that is different in all parts of the crown. *Echinoparyphium contiguum* must be redescribed before it can be definitely separated from *Echinop. aconiatum.* None of the other species discussed can be positively separated from *Echinostoma revolutum.* It may seem almost incredible that all sixteen of the species discussed here must be either reduced to synonymy or considered as doubtful species. It may also seem that too much emphasis has been placed on the similarity of the cephalic spination, and too little attention given to other structures. In most cases the character of the cephalic spination has borne more weight in my decisions than any other one factor. It must be conceded, however, that it is highly improbable that any two species could be identical in number, arrangement, and relative size of these 37 structures and at the same time be sufficiently dissimilar in other respects to be recognized as separate species. *Echinostoma miyagawai, cinetorchis, armigerum, mendax, paraulum,* and *columbae* are all unquestionably identical in this respect, and more than sufficient evidence is at hand to prove beyond reasonable doubt that the other two species which have been declared synonyms are indeed identical. Insufficient information regarding variations due to the age and the host as well as the question of host specificity account in part at least for the rather long list of synonyms. The procedure recommended by Kowalewski (1897) for designating synonyms is followed in the summary below:

Echinostoma revolutum (Froelich, 1802)

Syn. dem.

(Dietz, 1910) *Distomum echinatum* Zeder, 1803
(P. Beaver, present paper) *Echinostoma miyagawai* Ishii, 1932
(Idem) *Echinostoma cinetorchis* Ando and Ozaki, 1923
(Idem) *Echinostoma armigerum* Barker and Irvine, 1915
(Idem) *Echinostoma coalitum* Barker and C. A. Beaver, 1915
(Lutz, 1924; Sprehn, 1932; P. Beaver, present paper) *Echinostoma mendax* Dietz, 1909.
(Baylis, 1929; Iwata and Tamura, 1933; P. Beaver, present paper) *Echinostoma paraulum* Dietz, 1909.
(Krause, 1925a, b; P. Beaver, present paper) *Echinostoma columbae* Zunker, 1925.
(P. Beaver, present paper) *Echinostoma limicoli* Johnson, 1920.

Syn. inq.

Echinostoma sudanense Odhner, 1911
Echinostoma acuticauda Nicoll, 1914
Echinostoma callawayensis Barker and Noll, 1915
Echinostoma erraticum Lutz, 1924
Echinostoma neglectum Lutz, 1924
Echinostoma nephrocystis Lutz, 1924
Echinostoma microrchis Lutz, 1924
Echinostoma echinocephalum (Rud., 1819)
Echinostoma oxycephalum (Rud., 1819)
Echinostoma dilatatum (Miram, 1840)
Echinostoma armatum (Molin, 1850)

PROTOCOL OF EXPERIMENTS

On the Determination of Methods, Purity of Stocks, and Hosts

Preliminary experiments had shown that by feeding the cysts of the 37-spined echinostome cercariae to various birds and mammals, 37-spined adults could be recovered from them. This was not always true, however, as sometimes a negative was obtained even though it was definitely known that an abundance of cysts had been fed. This was particularly true with rats; and in one case a negative was obtained from a pigeon although it is probable that the snail fed did not contain cysts. With these experiences unexplained and Tubangui's (1932b) somewhat similar experience with pigeons and rats in mind it seemed possible that more than a single species or at least two varieties of the same species were being handled. Careful examination of the cercariae from each of the infected snails collected in various localities failed to disclose, however, what could be considered more than one species with 37 spines. Since several echinostome species have been described as having 37 cephalic spines, it was thought imperative to check the purity of the material employed.

The experiments to check on the above question are as follows: infected snails (*H. trivolvis*) were collected from three different localities (Table 8, p. 77), which represent three different streams and two entirely separate drainage systems, although only about thirty miles separate the headwaters of one from the other at the nearest point. The snails were wiped dry as they were collected in the field so that no cercariae could emerge and become mixed. Then as soon as they were brought into the laboratory each of them was cleaned, marked with india ink, and registered before they were isolated in various clear glass containers, usually eight-dram vials. Special care was employed to guarantee that no cercariae escaping from one snail might be allowed to mix with those of another. At no time during the experiments were any two infected snails in the same container. As soon as cercariae emerged they were examined to check on the various morphological characters used in diagnosis of species, especially the number, distribution, and relative size of the cephalic spines; body spination; character of the tail fin; number of cephalic ducts; and the main ducts of the excretory system. Size was also checked. In every case the characters exhibited were apparently identical. After the cercariae were checked the snail was given an additional number and again registered, so that complete records were kept on each infected snail. Various secondary intermediate hosts of laboratory origin were employed: *Physa gyrina, Helisoma trivolvis,* and tadpoles. Usually Physa were used. No careful check was kept on the number of cysts fed as this was thought to consume too much time to be worth while in this particular series of experiments. The following is a brief outline of the separate feedings and results. The experiments carry the same designation as the registered number of the snails which produced the cercariae, in addition to "1, 2, 3," etc. A single snail produced all of the cercariae used in each experiment. For example, in experiment 1, snail C3-A was used; in experiment 2, C4-28; etc.

EXPERIMENT 1(C3-A)

On 7/28/33 Pigeon 7 was fed two small Physa carrying many C3-A cysts, 1 day to 2 weeks old. Ten days later this pigeon developed a severe bloody diarrhea which continued into the next day. No ova were present yet in the stools when it was killed and examined on 8/8. The examination was made immediately, and 621 worms were recovered, none of which were really mature although about 30% of them had 5-20 ova in the lower uterus. These worms were scattered along the gut from anus to proventriculus. There were 44 worms in the gizzard but over half of these were dead. Most of the worms were in the lower duodenum and upper ileum. It is also of interest to note that two active worms were taken from above the gall bladder in the liver and nine were in the oviduct (upper). Death appeared to be imminent at the time the host was killed.

On 8/13/33 a half-grown albino rabbit (Rabbit 2) was fed three small Physa carrying many C3-A cysts, 1-14 days old. This animal died on 8/17. The autopsy was made 6-8 hours post-mortem and 284 specimens were recovered from about the middle of the ileum. Whether the worms were responsible for the death of this host is uncertain. It developed a rather severe diarrhea 36 hours before death, which may have been due to a change from a dry to a green diet. As all the other rabbits procured at the same time were fed infective cysts and as they too died, there was no control animal.

EXPERIMENT 2(C4-28)

Pigeon 10 was fed one small Physa with cysts on 8/13/33. Ova appeared in the feces on 8/22 and were more numerous each day until 8/26 when it was killed and 18 mature worms were taken from the middle ileum.

Rabbit 4, a half-grown albino, was fed 2 Physa with numerous cysts on 8/13/33. It died on 8/20 and was autopsied 3-4 hours post-mortem. One hundred twenty-two immature worms were taken from the ileum, for the most part from the second and third twelve inches above the cecum. It is uncertain as to whether or not the worms caused the host's death.

EXPERIMENT 3(C4-16)

Rabbit 5 was fed 2 Physa, each with about 100 cysts, on 8/31/33. Ova appeared in the feces on 9/12 and became more numerous daily until 9/17 when the host was killed and 28 mature worms taken from the lower and middle ileum.

Rabbit 6 was supposedly fed 1 Physa on 8/31. It fought violently, however, and it was thought doubtful at the time that any part of the snail was actually swallowed. It was not surprising then that its feces were negative, and no worms were found when it was killed on 9/18.

Pigeon 11 was fed 2 small Physa on 8/31/33. Ova appeared in the feces on 9/12 but were not numerous. They were easily found, however, by direct examination for five days, but none were found on the sixth. The pigeon was then killed and no worms were found.

EXPERIMENT 4(C5-A)

Pigeon 9, Rabbit 3, and Dog 1 were used in this experiment.

Pigeon 9 was fed 2 Physa with many C5-A cysts one to seven days old on 8/13/33. A few ova appeared in the feces on 8/22, were fairly numerous the next day, and very numerous on 8/24 when the pigeon was killed and 580 worms recovered. Some of those from the upper ileum were immature.

Rabbit 3 was fed C5-A cysts in one medium-sized Physa on 8/13/33. This rabbit died on 8/21/33 before the worms matured. The autopsy was made 12-14 hours post-mortem and 540 worms were recovered from the ileum, most of them about half-way between the duodenum and cecum. Many of those near the upper and lower ends were dead.

Dog 1 was fed C5-A cysts in 3 small Physa on 8/31/33. It was killed on 9/18 and 7 slightly immature worms were taken from the cecum and just above it in the lower ileum.

EXPERIMENT 5(C6-2)

Chicken 5, Dog 3, Cat 5, and Rats 30 and 31 were given cysts of C6-2.

Chicken 5 was fed a small Helisoma containing relatively few cysts on

9/19/33. On 10/1 the feces became positive for ova and on the next day the ova were abundant. The host was killed on 10/2 and 46 worms were taken from the middle ileum.

Dog 3, a setter bitch pup, was given 3 small Physa each harboring about 25 cysts and a tadpole with an unknown number of cysts on 9/15/33. This dog was anesthetized on 9/26 and devocalized, a procedure which caused an illness of about one week. Afterwards, however, it maintained good health and was found by fecal examination on 10/8 to be infected. It was then killed and 64 mature worms recovered from the second and third twelve inches of the ileum above the cecum.

Cat 5 was fed about 200 cysts in 6 very small Physa on 9/24/33. This cat was only about one-fourth grown and in rather poor health when given the cysts. It ate a normal meal of bread and milk at the time the cysts were given but refused to eat afterwards. It was passing a little blood in frequent stools the next day and soon developed a severe bloody diarrhea and died just 48 hours after it had eaten the cysts. Autopsy was delayed 12-14 hours, and was not thoroughly done. Only three specimens were discovered and these were badly disintegrated.

Rats 30 and 31 were fed 7 crushed Physa containing many metacercariae on 9/27/33. No ova were discovered in the feces, and when they were killed on 10/18 no worms were found.

EXPERIMENT 6(C6-3)

Chicken 6, Cat 2, and Pigeon 19 were fed the metacercariae of C6-3.

Chicken 6 was given 4 small Physa, each with a few cysts, on 9/19/33. Ova were found in the feces on 10/1 but were never numerous. Only two specimens were present when the host was examined on 10/5 and these were both in the cloaca.

Cat 2 was fed numerous metacercariae in a tadpole and one Helisoma. It became ill soon afterwards and on 10/2 looked as though it soon would die. The next day it was found in a state of coma. It was then killed and 39 immature worms taken from the lower ileum, 13 of which were dead. As the autopsy was made immediately it is certain that the worms were dead even before the host was killed.

Pigeon 19 was fed metacercariae on 9/27/33 in 4 small Physa. Ova appeared in the feces on 10/9 and nine days later the animal was killed. There were 93 worms in the middle ileum. This pigeon was also heavily parasitized with an internal mite.

EXPERIMENT 7(C6-5)

Rabbit 8, Pigeon 16, and Mice 14, 15, 16, and 17 were fed metacercariae of C6-5. They developed in the rabbit and pigeon but the mice were negative.

Rabbit 8 was fed metacercariae in a tadpole (*R. pipiens*) and 2 small Physa on 9/15/33. Ova were found in the feces on 9/27 and increased daily until the animal was killed (10/2). There were five mature worms in the middle ileum.

The four mice were fed five small Physa with metacercariae on 9/14/33. They were all negative when examined on 9/29.

Pigeon 16 was fed metacercariae in four small Physa on 9/28/33. The cysts were 1-7 days old. Ova did not appear until 10/12, several days later than was expected. It was killed on 10/13 and found heavily parasitized. There were 291 of the worms and about 1000 internal mites in addition. This may account for the delayed appearance of the ova. The 291 worms were scattered from lower ileum to duodenum, but most of them were in the middle section.

EXPERIMENT 8(C6-6)

Chicken 7, Guinea pig 1, and Rats 25 and 26 were fed metacercariae of C6-6. The chicken, guinea pig, and Rat 26 were positive but no worms were found in Rat 25.

Chicken 7 was fed metacercariae in 2 Physa on 9/19/33. It was killed on 9/30 in order to get some immature worms for another experiment. Only one worm was present, however. It was located in the lower ileum about ten inches above the ceca.

Guinea pig 1 was given a medium-sized Helisoma on 9/20/33 which had been in a finger bowl with the snail for about one week. It ate well and maintained good health until 9/30 when it became restless and refused food. The next morning it was dead. Autopsy was made about six hours post-mortem and the immediate cause of its death was found to be a large perforated duodenal ulcer. As there were 354 worms recovered from this host and nearly half of them were almost mature it seems very probable that they were in part at least responsible for the host's death. They were rather unevenly distributed between the middle colon and the stomach.

Rat 25 had been fed metacercariae from another snail (C6-10) on 9/15/33 but as it was never positive by fecal examination it was not killed. Then on 10/15 it was given a medium sized Physa carrying about 100 C6-6 metacercariae. The stools were never positive but as the feces were always solid it was thought that worms were possibly present even though no ova could be found. No worms were found, however, when it was killed on 11/4.

Rat 26 had been fed C6-11 metacercariae on 9/15/33 and although they must have been present no ova had been found in the feces up to 10/12. The pellets were always dry enough to collapse the ova and thus render the examination very difficult. A soft pellet was examined on 10/24, only twelve days after it had been fed C6-6 metacercariae and ova were easily found. This rat had been fed 26 or more metacercariae teased out of the kidneys of a tadpole (*R. pipiens*). These cysts were administered by mixing them into a bit of cornmeal and water. On 10/24 there were 33 worms in the middle ileum, three of which were much larger than the others and had the uterus well distended with ova. These were doubtless C6-11, which were fed on 9/15, and the other 30 were the C6-6. As was noted above, 26 metacercariae were counted in the kidney tissue, but several others could have been present but obscured from view.

EXPERIMENT 9(C6-7)

Rabbit 7, Chicken 3, and Pigeon 17 were used in this experiment. Worms developed in all three.

Rabbit 7 was fed 4 small Physa on 9/15 which carried metacercariae less than four days old. It was found positive by fecal examination on 9/27 and was killed six days later. There were 29 worms recovered, mostly from the middle ileum.

Chicken 3 was fed metacercariae in 3 small Physa on 9/19. A few ova were found in the feces on 10/1, and they were somewhat more numerous the next day when the animal was killed. Autopsy was made three hours post-mortem and 22 worms recovered. Most of them were in the middle ileum.

Pigeon 17 was given metacercariae in 7 small Physa on 9/28/33. A few ova were found on 10/9 and 10/11. It was killed on 10/11 and only two specimens were present, one of them not quite mature. They were in the middle ileum.

EXPERIMENT 10(C6-8)

Duck 3, Cat 4, and Pigeon 18 were fed metacercariae from C6-8. The cat and pigeon were positive; the duck was negative.

Duck 3 was fed 3 Physa carrying very many metacercariae on 9/15. No fecal examinations were made and when it was killed on 9/23 no worms were present. Although two or more explanations may be made for this the most probable is that the host was allowed to go without food for about a week. This occurred by accident, not as a part of the experiment.

Cat 4 was fed a Helisoma and 7 small Physa, each carrying a few metacercariae, on 9/23/33. Ova appeared in the feces on 10/13/33 and were found in abundance the following day. On 10/14 the host was killed and 352 worms taken from the caudal three feet of the ileum; 297 of these were concentrated into the second foot above the cecum.

Pigeon 18 was fed metacercariae on 9/28/33 in 2 small Physa. The feces became positive for ova on 10/9 and three days later the host was killed. The autopsy was delayed three hours. There were 80 worms entangled in the feathers around the anus, 74 in the cloaca and rectum, and 41 in the caudal four inches of the ileum.

EXPERIMENT 11(C6-9)

Pigeon 15 and Rat 27 were fed metacercariae of C6-9 and worms developed in both of them.

Pigeon 15 was fed the metacercariae in 2 small Physa on 9/15/33. Positive stools were passed on 9/27. Two days later it was killed and 63 worms taken from the middle ileum.

Rat 27 was fed a single tadpole (*R. pipiens*) that had been in four ounces of water with snail C6-9 during the 36 hours which immediately preceded the feeding on 9/15/33. Thus the cysts were 1-36 hours old. This host was killed by another rat on 9/29. The autopsy was made about ten hours post-mortem. There were 431 worms in the ileum, most of them near the middle. It is important to note that these 431 worms all developed from the metacercariae in one frog larva. It is also of importance to note that none of the cysts were over 36 hours old when administered.

EXPERIMENT 12(C6-11)

Hog 2, Pigeon 13, and Rat 26 were fed metacercariae of C6-11. The worms developed in all three.

Hog 2 was fed the cysts in 4 small Physa on 9/14/33. On 9/26 it was slaughtered and the autopsy made fourteen hours post-mortem. There were 32 worms recovered from the caudal eight feet of the ileum. Ten of these worms were dead and all were immature.

Pigeon 13 was given metacercariae in 2 small Physa on 9/15/33. Ova were found in the feces on 9/27, but may have been present earlier as no examination was made previous to that date. It was killed and 31 worms recovered on 9/30. Most of them were in the middle ileum.

Rat 26 was fed metacercariae in one tadpole on 9/15/33. It was killed on 10/24 and 3 large worms recovered from the middle ileum.

EXPERIMENT 13(C6-12)

Hog 3 and Pigeon 14 were fed metacercariae of C6-12. Both were positive.

Hog 3 was given the metacercariae in 5 small Physa on 9/14/33. No fecal

examinations were made. On 10/16 the animal was slaughtered and eight mature worms were recovered from the caudal one-fifth of the ileum, the most posterior one being about eighteen inches above the cecum. The autopsy was made about three hours post-mortem.

Pigeon 14 was given the metacercariae in 2 small Physa on 10/5/33. Ova appeared in the feces on 10/15 and were numerous for four days, then suddenly decreased and no more were found after the fifth day. The host was not killed.

EXPERIMENT 14(C6-17)

Rat 28 and Chicken 4 were fed metacercariae of C6-17 and worms developed in both of them.

Rat 28 was fed the metacercariae on 9/18/33 in one small tadpole (*R. pipiens*) that had been dead about eighteen hours. This rat seemed normal in every way until 10/2 when it was noticed that it had not eaten on that day. The next day it was obviously very ill. It was killed and an autopsy performed immediately. Ninety-five worms were taken from the middle and upper ileum. Some of them had ova in the uterus.

Chicken 4 was fed metacercariae in 2 small Physa on 9/19/33. Ova appeared in the feces on 10/1, and increased in abundance daily. On 10/4 the host was killed and 30 mature worms taken from the cloaca, rectum, ceca, and ileum. The autopsy was made about one hour post-mortem.

A summary of the above experimental feedings is given in Table 5. In all fourteen of the experimental feedings in which both birds and mammals were fed encysted cercariae from the same snail host, there is no instance of failure to infect at least one of each. Some of the hosts were entirely negative and others lost their infection before they were killed. Results with the rats were particularly inconsistent and somewhat confusing. Several races of rats were used, however, and there is a very great difference in the way each serves as a host for this species of worm (Table 6). Mature worms of each of the stocks were not always procured from more than one host, but there are, it seems, more than enough cases, nine in all, in which mature worms were recovered in considerable numbers from both birds and mammals to show that only one species was used in infecting both types of hosts. As was indicated above, there are not enough morphological differences in any of the cercariae used even to suggest a mixture of species, and the morphological variation in the adults is interpreted as being induced by the different host species (see p. 27).

On the Transfer of Adult Worms from Host to Host

In order to test further the possibility of physiologic differences in the worms from the different hosts a number of experiments were undertaken in which identical worms rather than worms of common parentage were used. Worms taken from mammals and introduced into the cloaca of pigeons were able to adjust themselves to the higher temperature, more

active gut, etc., and generate large numbers of ova for from several days to several weeks (Tables 7 and 9, p. 77). The individual transfers were made as described below. Some of them were bird-to-bird transfers which gave data on questions other than the one mentioned above.

TRANSFER I

Six mature worms (13 days old) were transferred from the lower ileum of Chicken 5 into the cloaca of Pigeon T1 at 9 A.M., 10/2/33. This pigeon like most of the others was put in a cage over a shallow pan of water for a day or two in order to check whether the worms were immediately expelled. None had been expelled after 24 hours. Ova appeared immediately in the feces and were very numerous the next day. There was no apparent reduction in the numbers until after twelve days (10/16) when there was a rather sharp decline. During the next ten days there was no further decline. On 10/27 the eggs were scarce and none were found after 10/28. At least one of the original six worms lived 26 days in the new host.

TRANSFER II

Six mature worms, 18 days old, were taken from the middle ileum of Rabbit 7 and transferred to the cloaca of Pigeon T2 on 10/3. Two hours later one of the worms was discovered in the pan under the cage. None others were found 24 hours later, however, so it was considered that five worms were successfully introduced. Ova appeared immediately and were numerous for five days. None were found after the sixth day (10/9).

TRANSFER III

Ten mature worms, 23 days old, were taken from the lower ileum of Dog 3 and transferred to the cloaca of Pigeon T3 on 10/8. Seven were expelled as soon as the pigeon was released but when they were reintroduced they established themselves successfully. Ova were found in very great abundance until 10/27 when there was a noticeable decline. A gradual decrease followed and none were found after 11/2.

TRANSFER IV

The pigeon used in Transfer II was also used in this one. It was given ten mature worms from the upper ileum of Pigeon 18 on 10/12. The worms were 14 days old. Ova were found daily until 10/25. They were never very numerous, however. It is important to note that this pigeon had only three days previous to this transfer lost the last of the worms from Transfer II.

TRANSFER V

This transfer was made from the upper ileum of Pigeon 19 to the cloaca of Pigeon T5 on 10/18. Only one robust worm was transferred. It was then 21 days old. Ova were found in very great abundance, 3,000 per 24 hours on 10/22. On 11/26 this worm was producing only 700 per 24 hours, and on 11/28 none could be found.

TRANSFER VI

A transfer was made from the middle ileum of Rat 26 to Pigeon T6. A single mature worm 39 days old was used. The pigeon had been used as Pigeon 14,

which had been infected from stock C6-12 and has lost its infection after fifteen days (10/20). The transfer was made on 10/24. Ova appeared in the feces immediately and continued for six days only.

TRANSFER VII

A single specimen, age 12 days, was taken from the middle ileum of Rat 26 and introduced into Pigeon T7. This pigeon had never been infected before. Ova did not appear until the fifth day after the transfer and continued for about two weeks. The last positive stool was found on 11/14.

TRANSFER VIII

Same as Transfer VII. Pigeon T8 had never been used in previous experiments. Ova appeared in the feces on the fifth day and continued three days longer than in T7. The last positive stool was found on 11/17.

TRANSFER IX

Like the above. Pigeon T9 lost its worm, however, before it matured.

TRANSFER X

Similar to VII, VIII, and IX. Pigeon T10 had never been infected before. The ova appeared on the fifth day and continued five days longer than in T8 and eight days longer than in T7. The last positive stool was found on 11/22, on the 28th day after the transfer.

A summary of these experiments is given in Table 9.

From the above experiments in which metacercariae of common parentage were fed to several hosts including in most cases both birds and mammals, and in the experiments in which worms were transferred from one host to another, the conclusions that may be reached are: (1) that the experimental worms constitute but a single species which (2) demonstrates a very high degree of adaptability, being able even after maturity is reached, to adjust itself to a new host that is very different from the original, thus (3) inter-class transfers are easily made, and (4) it is also probable that the life of the worms is prolonged somewhat by the transfer, which suggests that (5) there is probably a slight immunity developed which is of an accumulative nature. These points are discussed at length in the text (pp. 18-33).

CONCLUSIONS

When the previous studies on the life history and individual stages of the cycle were restudied it was found that relatively few of the older researches could be definitely shown to pertain to *Echinostoma revolutum*.

The eggs of this species hatch in about three weeks into miracidia which penetrate any one of a great variety of pulmonate snails and prob-

ably metamorphose into sporocysts which produce mother rediae which in turn produce a second generation of rediae. These daughter rediae produce cercariae which begin to emerge in about three months.

The cercaria resembles the adult so closely in cephalic spination that positive identifications can be made from this character alone. Other features by which it can be recognized are the tail-fin membrane and the twelve cephalic ducts and gland cells arranged in two groups. The arrangement of the units of the excretory system may also aid in identification.

In addition to the descriptions of cercariae that have been identified as *Cercaria Echinostomi-revoluti* by feeding experiments there are eleven other descriptions of 37-spined cercariae in the more recent literature. These cercariae are easily separated into two types and perhaps only two species. At least only two species of the 37-spined cercariae can be positively recognized.

Cercaria helvetica XXIV Dubois and *Cercaria trivolvis* Cort are synonyms of *Cercaria Echinostomi-revoluti.*

It is shown by this study that the cephalic spination is the most reliable character for diagnosis of the adult worm. The arrangement and relative size of the cephalic spines distinguish it from the other 37-spined echinostomes.

The morphology of the adult worm is best described by the use of graphs which describe any given size from the entire range of from 2 to over 20 mm in length.

The only character that is not variable with age is the number, arrangement, and relative size of the cephalic spines.

By transferring single individuals from one host to another it was found that individuals of this species produce as many as 4,600 ova per day. The same experiments have shown that self-fertilization is possible and that it does occur when the possibility of cross-fertilization is precluded.

A study of the original and other earlier descriptions of *E. revolutum* have shown that none of these records and descriptions can be accepted without so much reservation as to render them almost valueless.

Eighteen species of adult echinostomes have been described more recently as having about 37 cephalic spines. Two of them belong to the genus Echinoparyphium and are easily distinguished from *Echinostoma revolutum.* None of the other forms can be positively distinguished from one another or from *E. revolutum.* In addition to *Echinostoma echinatum,* which has already been reduced to synonymy, eight others are shown to be synonyms and eight are shown to be probable synonyms or species of dubious standing.

Echinostoma revolutum has been found in the temperate zone of all parts of the world excepting South Africa and the interior of Asia. The cercaria is recorded from eleven species of pulmonate snails in diverse geographical regions. The metacercaria may be found in molluscs, amphibia, fish, and (?) planaria. It is recorded from sixteen molluscs, primarily pulmonate gastropods; probably eight species of amphibia; and one scaleless fish, *Ameirus melas*. The adult worm is recorded from 32 species of birds and nine species of mammals, including man. The present study has contributed ten new records for the metacercaria and seventeen for the adult.

It is shown that experimental and "incidental" hosts are "normal" if not natural hosts for this species of parasite and that natural infection would probably occur in almost any bird or mammal whose feeding habits are similar to the so-called "natural" hosts.

By repeatedly infecting both tadpoles and snails from the same stock of cercariae and feeding these to both birds and mammals it was shown that only one species of parasite was being studied rather than two or more indistinguishable forms, and that there is relatively no specificity shown in the metacercaria and adult stages.

Experiments in which worms were transferred from mammals to birds support the above conclusions.

The same transfer experiments indicate that the life of the parasite is prolonged by introduction into a new host.

Transfer and reinfection experiments have demonstrated that although prolonged captivity of the host and unsuitable diet produce more obvious reactions which tend to obscure immunity phenomena, the pigeon does develop a slight immunity to *E. revolutum*.

A total of 42 different measurements on each of about 300 worms were made and the worms from birds compared by biometric methods with those from mammals. While the differences are in most respects slight they are nevertheless in many instances significant, and may account for a good deal of the confusion regarding the morphology of the adult worm. The most obvious differences were found in the character of the cuticular spination. Differences in proportions are probably due to the very pronounced difference in growth rate in the two types of hosts.

SUMMARY

The life history of the trematode parasite *Echinostoma revolutum* has been experimentally determined and the morphology of the various stages described.

The adult worm has been described by biometric methods, which was found to be the only adequate means of description for a worm whose size range in adult life is so great as to allow much confusion in diagnosis.

Extensive studies have been made on normal variation, especially the variation induced by the different types of hosts.

Related species have been reviewed and eight species reduced to synonymy.

All of the more reliable records of hosts and distribution have been reviewed and summarized.

For the first time adult worms have been successfully transferred from mammal- to bird-hosts.

Certain immunity phenomena have been observed and discussed.

TABLE 1.—*Cercaria Echinostomi revoluti*
(e = experimental infection only)

Host	Locality	Reference
Helisoma trivolvis	Illinois	Cort, 1914, 1915
	Illinois	Faust, 1918
	Illinois	Beaver (this paper)
	Ontario	Fallis, 1934
Lymnaea peregra	Philippine Islands	Tubangui, 1932b
Lymnaea pervia	Taiwan	Tsuchimochi, 1924
	Taiwan	Morishita, 1929
Lymnaea radix	Taiwan	Tsuchimochi, 1924, 1926
Lymnaea stagnalis	Italy	Vevers, 1923
	Switzerland	Dubois, 1928
Lymnaea swinhoe	Taiwan	Tsuchimochi, 1924
Physa gyrina	Illinois	E. L. Miller, 1936
	Ontario	Fallis, 1934
Physa occidentalis (?)	California	Johnson, 1920
Physa rivalis	Brazil	Lutz, 1924
Planorbis sp.	Taiwan	Tsuchimochi, 1924, 1926
Pseudosuccinea columella	Maryland	Krull, 1935 (e)
Stagnicola palustris	Ontario	Fallis, 1934

TABLE 2.—*Metacercaria Echinostomi revoluti*
(e = experimental infection only)

Host	Locality	Reference
Corbicula producta	Taiwan	Anazawa, 1929
Fossaria abrussa	Ontario	Fallis, 1934
Fossaria modicella	Illinois	Beaver (this paper) (e)
	Ontario	Fallis, 1934
Helisoma trivolvis	Illinois	Beaver (this paper)
	Ontario	Fallis, 1934
	Maryland	Krull, 1935 (e)
Lymnaea peregra	Philippine Islands	Tubangui, 1932b
Lymnaea pervia	Taiwan	Tsuchimochi, 1926
Lymnaea radix	Taiwan	Tsuchimochi, 1926
Lymnaea stagnalis	Italy	Vevers, 1923
Lymnaea traski (?)	California	Johnson, 1920
Musculium partumeium	Maryland	Krull, 1935 (e)
Physa gyrina	Illinois	Beaver (this paper)
	Ontario	Fallis, 1934
Physa halei	Maryland	Krull, 1935 (e)
Physa occidentalis (?)	California	Johnson, 1920
Physa rivalis	Brazil	Lutz, 1924
Planorbis sp.	Brazil	Lutz, 1924
Pseudosuccinea columella	Illinois	Beaver (this paper) (e)
	Maryland	Krull, 1935 (e)
Viviparus sp.	Taiwan	Tsuchimochi, 1926
Pisidium sp.	Illinois	Beaver (this paper) (e)
Sphaerium sp.	Illinois	Beaver (this paper) (e)
	Ontario	Fallis, 1934
Stagnicola palustris	Ontario	Fallis, 1934
Planaria sp. (?)	California	Johnson, 1920
Rana esculenta	Japan	Miki, 1923
Rana japonica	Japan	Dollfus, 1925
Rana pipiens (larva)	Illinois	Beaver (this paper)
Rana pipiens (adult)	Illinois	Beaver (this paper)
Rana rugosa	Japan	Dollfus, 1925
Rana catesbiana (larva)	Japan	Dollfus, 1925 (e)
Rana nigromaculata	Japan	Dollfus, 1925
"Tadpoles"	Taiwan	Tsuchimochi, 1926
"Tadpoles" (*Bufo americana*)	Ontario	Fallis, 1934
Ameirus melas	Illinois	Beaver (this paper) (e)

TABLE 3.—*Echinostoma revolutum*, ADULT
(e = experimental infection only)

Host	Locality	Reference
Anas brasiliense	Brazil	Dietz, 1910
Anas platyrhynchos (domestic)	Poland	Kowalewski, 1895, 1897
	Taiwan	Anazawa, 1929 (e)
	Taiwan	Iwata and Tamura, 1933
	Taiwan	Tsuchimochi, 1924
	Japan	Yamaguti, 1933
	Japan	Ishii, 1932
	Philippine Islands	Tubangui, 1932a
	England	Vevers, 1923 (e)
	England	Brown, 1926 (e)
	Turkestan	Skrjabin, 1913
	Middle-Europe	Dietz, 1910
	Illinois	Beaver (this paper) (e)
	Illinois	Miller, 1936 (e)
Anas P. platyrhynchos (wild)	Japan	Ishii, 1932
	Middle-Europe	Dietz, 1910
	Illinois	Beaver (this paper)
Anas superciliosa	Queensland, Australia	Johnston, 1913, 1916
	Queensland	Nicoll, 1914
Anser anser (domestic)	Taiwan	Iwata and Tamura, 1933
	Middle-Europe	Dietz, 1910
	Ontario	Fallis, 1934 (e)
Anser cinerius	Ontario	Swales, 1933a
Anseranas semipalmata	Queensland	Nicoll, 1914
Bueteo lagopus-s. johannis	Illinois	Beaver (this paper)
Cairina moschata	Brazil	Lutz, 1924 (e)
	Brazil	Dietz, 1910
Cheniscus pulchellus	Queensland	Nicoll, 1914
Chenopsis atrata	Queensland	Nicoll, 1914
Coloeus monedula	Poland	Markowski, 1933
Columba domestica	Philippine Islands	Tubangui, 1932b (e)
	Germany	Krause, 1925a (*Echinostoma* sp.)
	Germany	Zunker, 1925 (*E. columbae*)
	Germany	Bolle, 1925 (*E. columbae*)
	Germany	Sprehn, 1927 (*E. paraulum*)
	Illinois	Beaver (this paper) (e)
Colymbus cristatus	Middle-Europe	Dietz, 1910 (*E. paraulum*)
Coracias garrula	Egypt	Looss, 1899
Corvus cornix	Poland	Markowski, 1933
Dendrocygna viduata	Brazil	Dietz, 1910 (*E. mendax*)
"Duck"	Ontario	Swales, 1933b (corrected)
Gallus gallus (domestic fowl)	Turkestan	Skrjabin, 1913
	Taiwan	Tsuchimochi, 1924
	Taiwan	Anazawa, 1929 (e)
	Japan	Ishii, 1932
	Japan	Kurisu, 1932

TABLE 3.—*Concluded*

Host	Locality	Reference
Gallus gallus (domestic fowl) cont'd..	Japan	Iwata and Tamura, 1933
	Poland	Kowalewski, 1895, 1897
	Middle-Europe	Dietz, 1910
	Illinois	Beaver (this paper) (e)
Mareca americana	Alberta, Canada	Beaver (this paper)*
Mareca penelope	Middle-Europe	Dietz, 1910
Marila marila	California	Johnson, 1920
Meleagris gallopavo	Ekaterinburg, U.S.S.R.	Skrjabin, 1915
Neochen jubata	Brazil	Dietz, 1910 (*E. mendax*)
Nyroca fuligula	Japan	Yamaguti, 1933
Nyroca marila	California	Johnson, 1920
Oidemia nigra	Middle-Europe	Dietz, 1910
Pernis apivorus	Egypt	Looss, 1899
Philomachus pugnax	Egypt	Looss, 1899
Scolopax rusticola	Ekaterinburg, U.S.S.R.	Skrjabin, 1915
"Swan"	Taiwan	Iwata and Tamura, 1933
Tadorna tadorna	Middle-Europe	Dietz, 1910
Canis familiaris	Taiwan	Tsuchimochi, 1924 (e)
	Taiwan	Anazawa, 1929 (e)
	Illinois	Beaver (this paper) (e)
Cavia porcellus	Illinois	Beaver (this paper) (e)
	Maryland	Krull, 1935 (e)
Epimys norvegicus	Japan	Dollfus, 1925 (*E. cinetorchis*)
	Taiwan	Tsuchimochi, 1924 (e)
	Illinois	Beaver (this paper) (e)
Felis domestica	Illinois	Beaver (this paper) (e)
Homo sapiens	Taiwan	Anazawa, 1929
Mus musculus	Taiwan	Anazawa, 1929 (e)
	Illinois	Beaver (this paper) (e)
Ondatra zibethica	Pennsylvania	Leidy, 1888
	Nebraska	Barker, 1915 (*E. armigerum* and *E. coalitum*)
	Ontario	Law and Kennedy, 1932 (*E. armigerum* and *E. coalitum*)
	Quebec	Beaver (this paper)*
	Alberta	Beaver (this paper)*
	Pennsylvania	Beaver (this paper)†
	Illinois	Beaver (this paper)
	Colorado	Beaver (this paper)†
	Maryland	Krull, 1935 (*E. coalitum*)
Rabbit (domestic)	Illinois	Beaver (this paper) (e)
	Maryland	Krull, 1935 (e)
Sus scrofa	Illinois	Beaver (this paper) (e)

*Collected by W. E. Swales.
†Records obtained from a study of specimens in the collection of Henry B. Ward.

TABLE 4.—SUMMARY OF REINFECTION EXPERIMENTS

Host	Days infected	Dates of infections	Remarks
Im 1	6	10/3 —10/9	5 worms administered *per anus*
	12	10/12—10/24	12 worms administered *per anus*
	?	11/2 —	Negative by fecal examinations
	?	11/22—	Negative by fecal examinations and autospy 12/15
Im 2	15	10/5 —10/20	Positive by fecal examination from 10/15 to 10/20
	6	10/24—10/30	1 worm administered *per anus*
	?	11/2 —	Negative by fecal examination
	8	12/6 —12/14	1 immature worm recovered
Im 3	24	10/8 —11/1	10 worms administered *per anus*
	16	11/2 —11/18	Positive by fecal examination from 11/2 to 11/18
	10	11/6 —12/16	3 immature worms recovered
Im 4	26	10/2 —10/28	6 worms administered *per anus*
	32	11/2 —12/4	Positive by fecal examination from 11/18 to 12/5
	?	12/6 —	Negative by autospy on 12/15
Im 5	28	10/24—11/22	1 worm administered *per anus*
	18	12/10—12/28	2 mature worms recovered
Im 6	40	10/18—11/28	1 worm administered *per anus*
	21	12/10—12/31	11 mature worms recovered
Im 7	20	10/24—11/14	1 worm administered *per anus*
	?	12/10—	Negative by fecal examination and autospy on 12/27
Im 8	?	10/24—	1 worm administered *per anus;* probably expelled immediately
	19	12/10—12/29	1 worm recovered, mature

TABLE 5.—EXPERIMENTS IN WHICH CERCARIAE OF COMMON PARENTAGE WERE FED TO BIRDS AND MAMMALS

Exp. no.	Cercaria used	Host	Number of worms	Age in days	Position in host
1	C3-A	Pigeon 7	621	11	Anus to proventriculus
		Rabbit 2	284	4	Cecum to duodenum
2	C4-28	Pigeon 10	18	13	Middle ileum
		Rabbit 4	122	7	Lower and middle ileum
3	C4-16	Rabbit 5	28	17	Lower and middle ileum
		Rabbit 6	0	0	
		Pigeon 11	0	?	?
4	C5-A	Rabbit 3	540	8	Cecum to duodenum
		Pigeon 9	580	11	Anus to duodenum
		Dog 1	7	18	Cecum and lower ileum
5	C6-2	Chicken 5	46	13	Middle and lower ileum
		Dog 3	64	23	Lower part of middle ileum
		Cat 5	3	2	Lower ileum
		Rat 30	0	0	
		Rat 31	0	0	
6	C6-3	Chicken 6	2	16	Cloaca
		Cat 2	39	13	Lower ileum
		Pigeon 19	93	21	Middle and upper ileum
7	C6-5	Rabbit 8	5	17	Middle ileum
		Pigeon 16	291	15	Middle and upper ileum
		Mice 14-17	0	0	
8	C6-6	Chicken 7	1	11	Lower ileum
		Guinea pig 1	354	11	Upper colon to stomach
		Rat 25	0	0	
		Rat 26	30	12	Middle ileum
9	C6-7	Rabbit 7	29	18	Middle ileum
		Chicken 3	22	13	Cloaca, lower and middle ileum
		Pigeon 17	2	13	Middle ileum
10	C6-8	Duck 3	0	0	
		Cat 4	352	21	Lower and middle ileum
		Pigeon 18	195	14	Cloaca, rectum, and lower ileum
11	C6-9	Pigeon 15	63	14	Lower, middle, and upper ileum
		Rat 27	431	14	Lower, middle, and upper ileum
12	C6-11	Hog 2	32	12	Lower ileum
		Pigeon 13	31	15	Lower, middle, and upper ileum
		Rat 26	3?	39?	Middle ileum
13	C6-12	Hog 3	8	32	Lower ileum
		Pigeon 14	0	14-15	Host not killed
14	C6-17	Rat 28	95	15	Middle and upper ileum
		Chicken 4	30	15	Cloaca, lower and middle ileum

TABLE 6.—SUMMARY OF DATA ON HOSTS USED IN ALL EXPERIMENTS

Host no.	Cercaria used	Number of worms	Mature in days	Age of worms in days	Host type
Pigeons					
1	C1-A	38	..	4 and 1	
2	C1-A	300	11	11	
3	C1-A	0	11	Worms expelled on 12th day	
4	C1-A	6	10	16	
5	C3-A	0	..	Cysts not swallowed	
6	C3-B	14	..	6	
7	C3-A	621	11	11	
8	C4-mix	82	9	11	
9	C5-A	580	9	11	
10	C4-28	18	9	13	
11	C4-16	0	12	Worms expelled on 17th day	
12	C6-10	96	10	13	
13	C6-11	31	11	15	
14	C6-12	0	10	Worms expelled on 15th day	
15	C6-9	63	10	14	
16	C6-5	291	12	15 (Many immature)	
17	C6-7	2	11	13	
18	C6-8	195	11	14	
19	C6-3	93	11	21	
20	C7-4	1	..	8 (Used as T6 and Im 2)	
21	C7-4	0	..	0 (Used as T1 and Im 4)	
22	C7-4	0	..	0 (Used as T2, T4, Im 1)	
23	C7-4	3	..	10 (Used as T3 and Im 3)	
24	C6-6	0	..	0 (Used as T7 and Im 7)	
25	C6-6	2	17	18 (Used as T10 and Im 5)	
26	C6-2	1	19	19 (Used as T9 and Im 8)	
27	C6-6	11	12	21 (Used as T5 and Im 6)	
Domestic Fowl					
1	C4-mix	2	..	9 (Immature)	
2	C4-mix	11	14	17	
3	C6-7	22	12	13	

TABLE 6.—*Continued*

Host no.	Cercaria used	Number of worms	Mature in days	Age of worms in days	Host type
Domestic Fowl (Concluded)					
4	C6-17	30	12	15	
5	C6-2	46	12	13	
6	C6-3	2	12	16	
7	C6-6	1	..	11 (Immature)	
8	C6-13	9	12	13	
Domestic Duck					
1	C4-10	1	..	14	
2	C4-mix	2	..	10 (Mature)	
3	C0-0	27	..	14	
4	C6-8	0	..	.. (Worms expelled)	
5	C7-3	6	9	17	
Rats					
1	C0-0	3–400	4	4	Wild
2	C0-0	3–400	..	1	Wild
3	C0-0	20	17	20	Wild
5	C0-0	0	18	Worms expelled on 24th day	Brown
6	C0-0	0	18	Out on 22nd day	Wild
7	C1-A	0	Neg.	..	Hooded
8	C1-A	0	Neg.	..	Albino
9	C1-A	0	Neg.	..	Albino
10	C1-A	0	Neg.	..	Wild
11	C3-B	0	Neg.	..	Hooded
12	C3-A	0	Neg.	..	Albino
13	C3-B	0	Neg.	..	Albino
14	C3-B	0	Neg.	..	Hooded
15	C3-B	0	Neg.	..	Albino
16	C4-mixed	0	Neg.	..	Albino
17	C4-mixed	0	Neg.	(Not well fed)	Hairless
18	C4-mixed	75	19	22	Hairless

Note: Rat 4 escaped; no data.

TABLE 6.—*Continued*

Host no.	Cercaria used	Number of worms	Mature in days	Age of worms in days	Host type
Rats (Concluded)					
19	C4-mixed	124	..	16	Hairless
20	C4-6	0	Neg.	..	Hooded
21	C4-mixed	0	Neg.	..	Albino
22	C5-A	0	Neg.	..	Albino
23	C4-28	0	Neg.	..	Albino
24	C4-40	0	Neg.	..	Wild
25	C6-10	0	Neg.	..	Wild
26	C6-6	33	..	12	Wild
27	C6-9	431	..	14	Hairless
28	C6-17	95	..	15	Hairless
29	C6-13	0	Neg.	..	Wild
30	C6-2	0	Neg.	..	Wild
31	C6-2	0	Neg.	..	Wild
32	C7-4	0	Neg.	..	Yellow
33	C7-4	0	Neg.	..	Blue
34	C7-4	0	Neg.	..	Hooded
Mice					
1	C0-0	350	..	2	Black
2	C0-0	2	..	3	Albino
3	C0-0	0	Neg.	..	Yellow
4	C0-0	500	..	9 (None mature)	Yellow
5	C1-A	0	Neg.	..	Black
6	C0-0	0	Neg.	..	Yellow
7	C0-0	0	Neg.	..	Yellow
8	C1-A	0	Neg.	..	Yellow
9	C4-mixed	10	..	7	Yellow
10	C4-40	1	..	12 (Immature)	Wild
11	C4-40	0	Neg.	..	Wild
12	C6-5	0	Neg.	..	Wild
13	C6-5	0	Neg.	..	Wild
14	C6-5	0	Neg.	..	Wild
15	C6-5	0	Neg.	..	Wild

TABLE 6.—*Concluded*

Host no.	Cercaria used	Number of worms	Mature in days	Age of worms in days	Host type
Rabbits					
1	C5-A	75	..	1	Albino
2	C3-A	284	..	4	Albino
3	C5-A	540	..	8	Albino
4	C4-28	122	..	7	Albino
5	C4-16	28	12	17	Wild
6	C4-16	0	..	(Not well fed)	Chocolate
7	C6-7	29	12	18	Wild
8	C6-5	5	12	17	Wild
Dogs					
1	C5-A	7	..	18 (Immature)	Collie
2	C4-mixed	700	..	8	Setter
3	C6-2	64	22	23	Setter
Cats					
1	C4-40	0	16?	(Expelled on 17th day)	Mongrel
2	C6-3	39	..	13	Mongrel
3	C6-13	1	..	2	Black
4	C6-8	352	18	21	Mongrel
5	C6-2	3	..	2	Mongrel
6	C6-6	0	?	(None present on 26th day)	Mongrel
Hogs					
1	C6-10	1	..	12	Hampshire
2	C6-11	32	..	12	Hampshire
3	C6-12	8	..	32	White
4	C6-8	..	..	(Gut mixed by butcher)	White
Guinea pigs					
1	C6-6	354	(Few with ova in the uterus when autopsied on the 11th day)		

TABLE 7.—RATE OF DEVELOPMENT IN THE EXPERIMENTAL HOSTS

Host	Body temperature (Approximate)	Days required to reach maturity	
		Range	Average
Rat	100	17–19	18
Mouse[1]	.....	.....	..
Rabbit	102	12	12
Dog	101.5	22	22
Cat	102	16?–18	18
Hog	102	32	32
Guinea Pig	101.7	11	11[2]
Duck	110	10–11	10.2
Fowl	107–109	12–14	12.5
Pigeon	106–108	9–12[3]	11
All mammals	.....	11–32	19
All birds	.....	9–14	11

[1]No mature worms taken.
[2]Worms almost mature, eggs in uterus but none in host's feces.
[3]Pigeons with reinfection required longer than 12 days.

TABLE 8.—COLLECTIONS OF *Helisoma trivolvis* IN THE VICINITY OF URBANA, ILLINOIS

Collection number	Date	Source	Number collected	Number infected
C1	6/ 9/33	Camp Creek, Seymour	14	1
C2	6/20/33	Drainage ditch at Crystal Lake Park	35	0
C3	7/ 8/33	First north tributary of Salt Fork, West of Homer Park	14	2
C4	8/ 1/33	Camp Creek, Seymour	100	43
C5	8/ 5/33	Leverette, Illinois, Drainage Ditch	41	1
C6	9/11/33	Camp Creek	72	38
C7	10/28/33	Camp Creek	9	1
C8	3/10/34	Camp Creek	12	1

TABLE 9.—TRANSFER EXPERIMENTS

Transferred from	Transferred to	Number of worms	Days in original	Days in second	Total age in days
Chicken 5	Pigeon T1	6	13	26	39
Rabbit 7	Pigeon T2	5	18	6	24
Dog 3	Pigeon T3	10	23	24	47
Pigeon 18	Pigeon T4 (T2)	10	14	12	26
Pigeon 19	Pigeon T5	1	21	40	61
Rat 26	Pigeon T6 (14)	1	39	6	45
Rat 26	Pigeon T7	1	12	20	32
Rat 26	Pigeon T8	1	12	23	35
Rat 26	Pigeon T9	1	12	?	?
Rat 26	Pigeon T10	1	12	28	40

EXPLANATION OF CHARTS

The charts serve three separate purposes. Charts 1, 2, 3, 4, 5, 6, 7, 19, and 20 give the average measurements for specimens of any given size under 20 mm. The range of variability that is due to factors other than host is best shown on Charts 8, 9, 10, 11, 12, 13, 14, 19, and 20, where measurements of individual worms are plotted separately. Charts 7, 8, 9 and 10, 11 and 12, 13 and 14, 15, 16, 17, 18, 19 and 20, and especially 8, serve to show the differences and similarities to be found in specimens taken from the two types of hosts in which they commonly occur in the adult stage.

The curves are made from lines drawn through points obtained by averaging all of the individual points within arbitrary limits. The lines radiating from zero are drawn through points of equal proportion and this proportion is indicated in each case on the line as 1:2, 1:3, etc.

Chart 1 shows the diameter of the acetabulum and the width of the body in relation to the total length of the body. The former shifts from one-fifth in very small worms to about one-sixteenth the body length in specimens over 13 mm in length. Similarly the body width shifts from one-third to nearly one-tenth.

Chart 2 shows the size relationship between the acetabulum and the collar. The acetabulum is the larger in worms over 4 mm in length.

Chart 3 shows the size relationship between the acetabulum, oral sucker, and collar. The body length may be determined from Chart 1, and the relative size of the pharynx may be seen in Charts 19 and 20.

Chart 4 shows the relative lengths of the three regions of the body limited by the acetabulum and the ovary.

Chart 5 shows the relative size of the gonads in relation to the body length.

Chart 6 shows the relative length of the lateral posterior ventral collar spine (see Text-figs. 3 and 5) in relation to the body length.

Chart 7 shows the relationship between the body length and the length of the cuticular spines in specimens from the two types of hosts.

Chart 8 shows the relationship between the body length and the number of complete rows of dorsal cuticular spines (see Text-fig. 2) in specimens from the two types of hosts. In this chart the lengths of the individuals having the same number of complete rows have been averaged and the total number within the class indicated by a number near each point.

Charts 9 and 10 show the relationship between the body length and the greatest width of the body in specimens from the two types of hosts. The larger worms are slightly stouter in bird hosts.

Charts 11 and 12 show the size relationship between the body length and the acetabulum in specimens from the two types of hosts. The acetabulum is slightly smaller in the smaller specimens from mammals.

Charts 13 and 14 show the length relationship between the whole body and the pre-acetabular region in specimens from the two types of hosts. Differences here appear to be insignificant.

Chart 15 shows the length relationship between the body and the region of the uterus. It appears here that there is no significant difference in the length of this region in the specimens from the two types of hosts.

Chart 16 shows the length relationship between the body and the post-ovarian region in specimens from the two types of hosts. The greater length of this region in larger specimens from bird hosts is probably significant.

Chart 17 shows the difference in the size relationship between the acetabulum and the collar in specimens from the two types of hosts.

Chart 18 shows the size relationship between the acetabulum and the oral sucker in specimens from the two types of hosts. The oral sucker is relatively larger in the larger specimens from birds.

Charts 19 and 20 show the size relationship between the pharynx and the oral sucker. The variation in this character in the specimens from the two types of hosts is probably not significant.

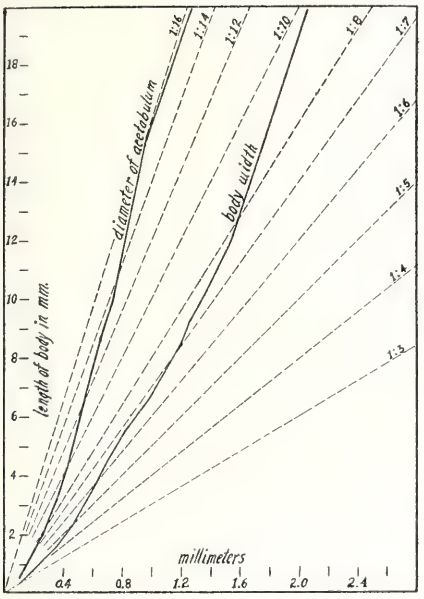


CHART 1

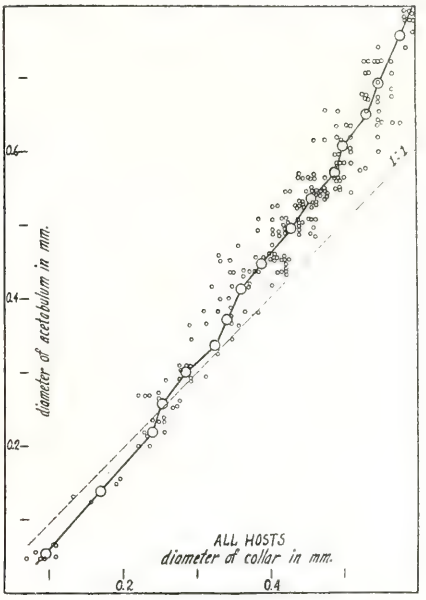


CHART 2

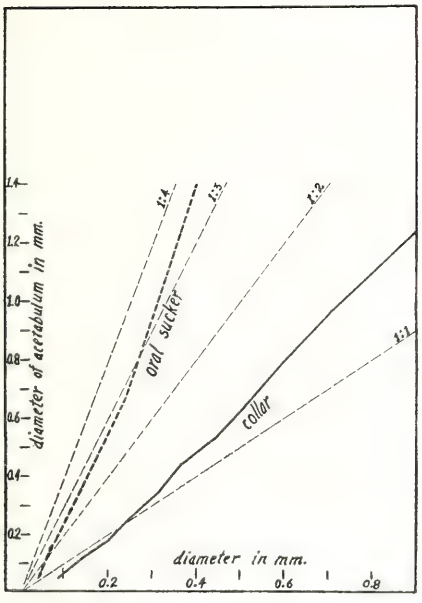


CHART 3

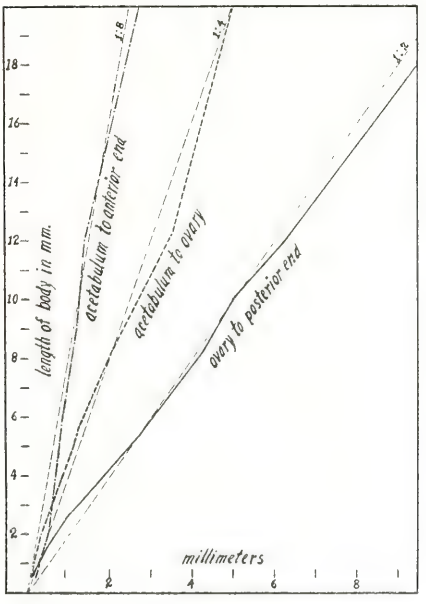


CHART 4

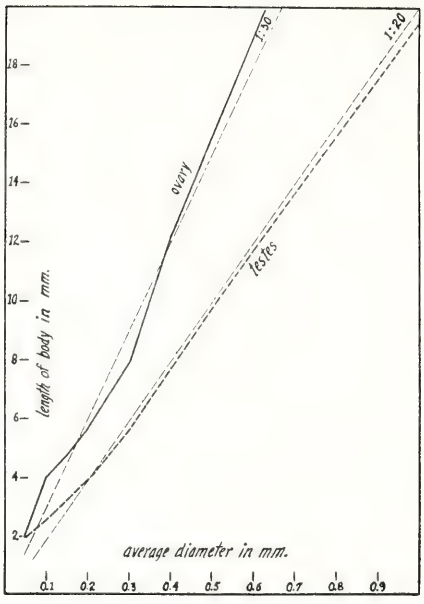


CHART 5

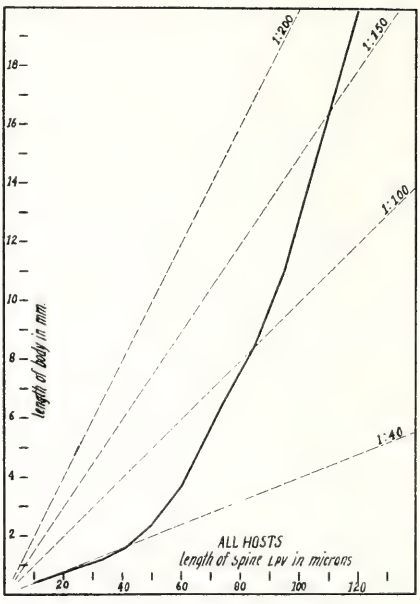


CHART 6

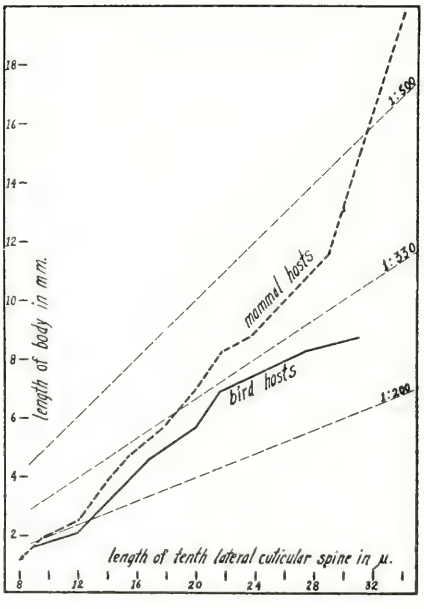


CHART 7

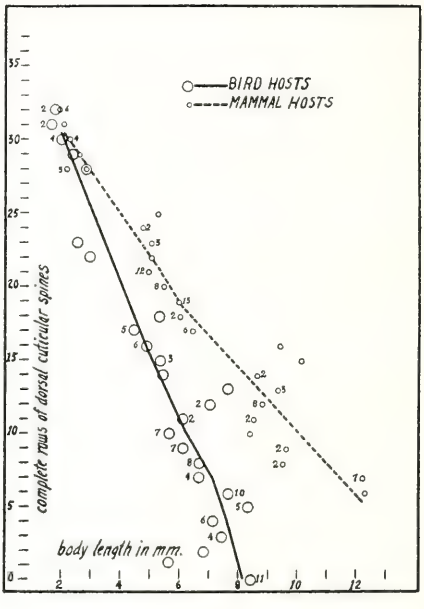


CHART 8

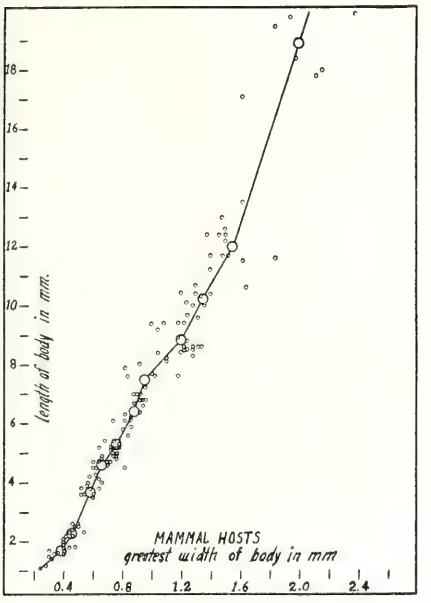


CHART 9

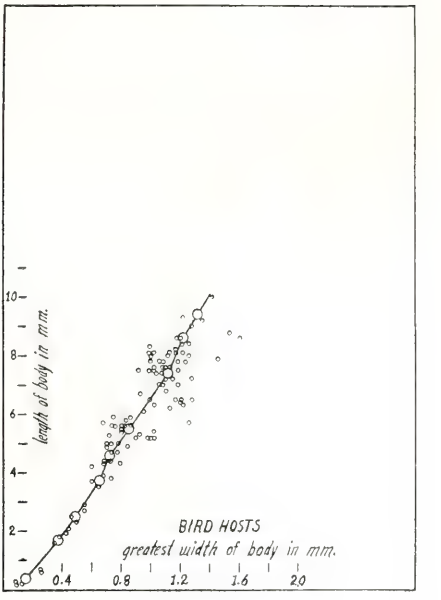


CHART 10

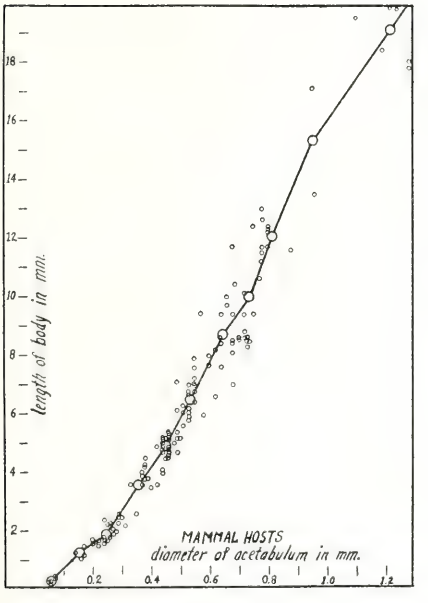


CHART 11

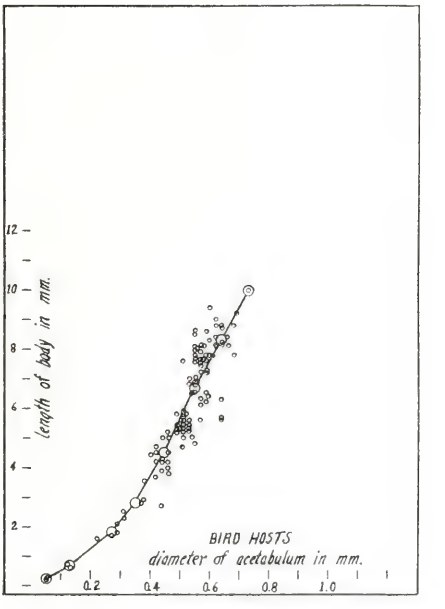


CHART 12

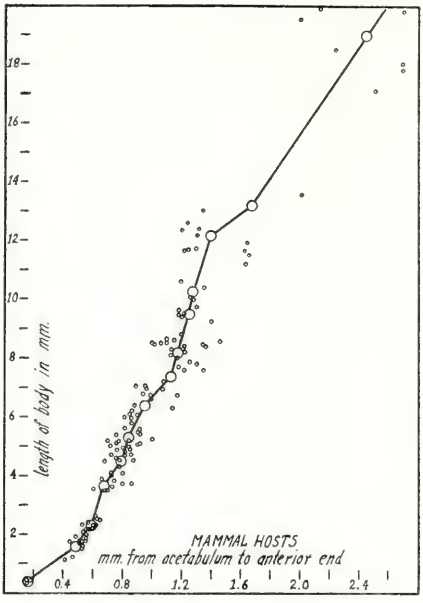


CHART 13

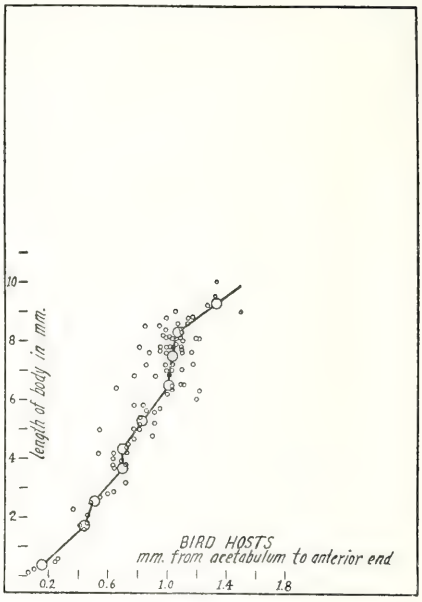


CHART 14

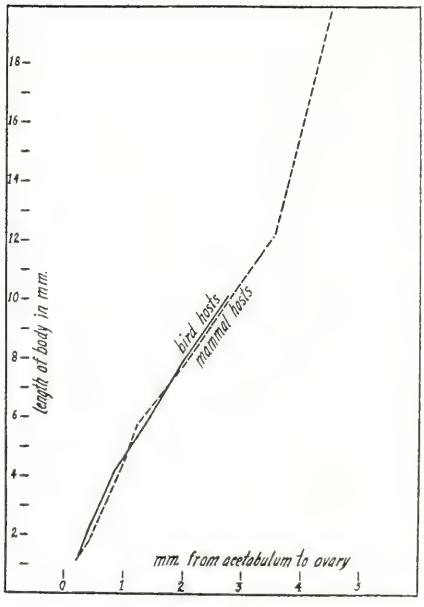


CHART 15

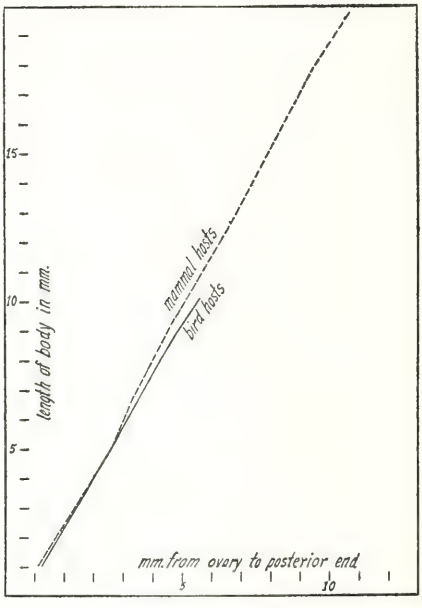


CHART 16

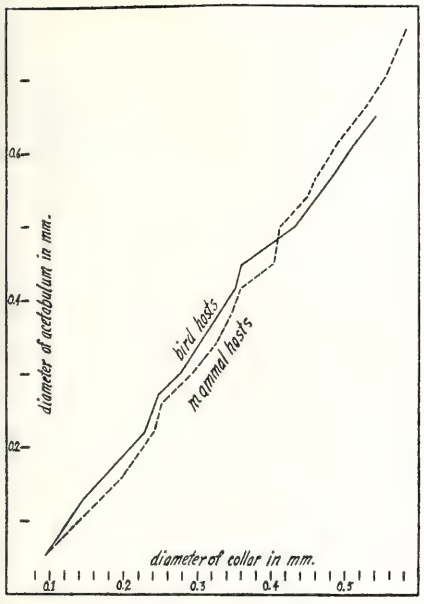


CHART 17

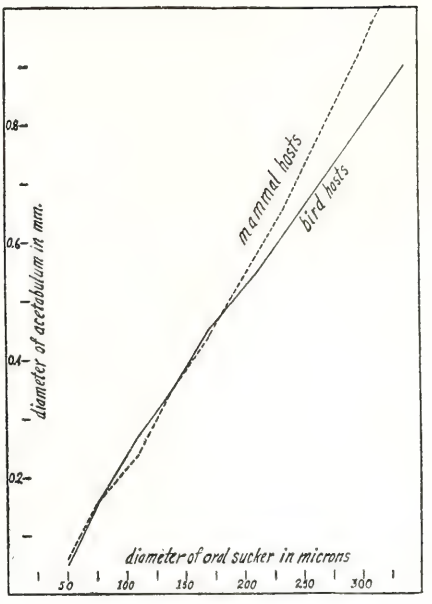


CHART 18

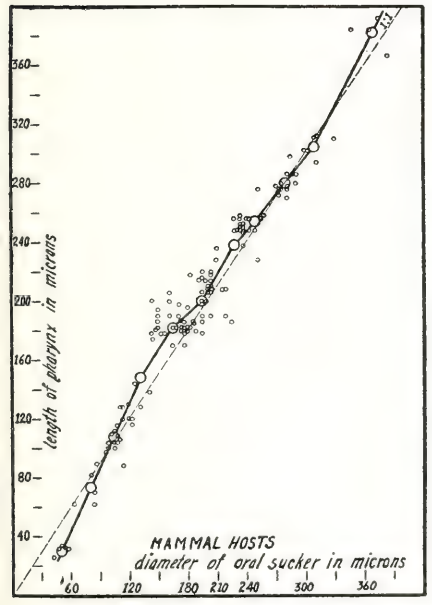


CHART 19

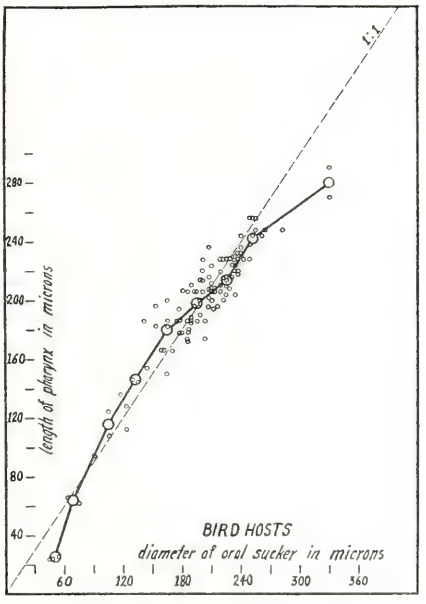


CHART 20

PLATE 1

FIGS. 1, 2, 3.—Head of cercaria, showing cephalic spination in lateral, ventral, and dorsal views, respectively. (Camera lucida drawings).

FIG. 4.—Camera lucida drawing of a fixed specimen of the cercaria.

FIG. 5.—Diagram of tadpole showing the usual course followed by creeping cercariae.

FIG. 6.—Diagram showing the cephalic ducts as seen in the unflattened specimen (cercaria).

FIG. 7.—Diagram of the cuticular plates of the miracidium in dorsal view.

FIG. 8.—Camera lucida drawing of the partially contracted tail of the cercaria. Lateral view.

FIG. 9.—Camera lucida drawing of the head of an 8.55 mm adult from a chicken.

FIG. 10.—Camera lucida drawing of the tip of the tail of a fixed cercaria, showing the extent of the circular muscles and the fin fold membrane.

FIG. 11.—The miracidium, almost mature within the egg. Outlines made with the camera lucida, details free-hand.

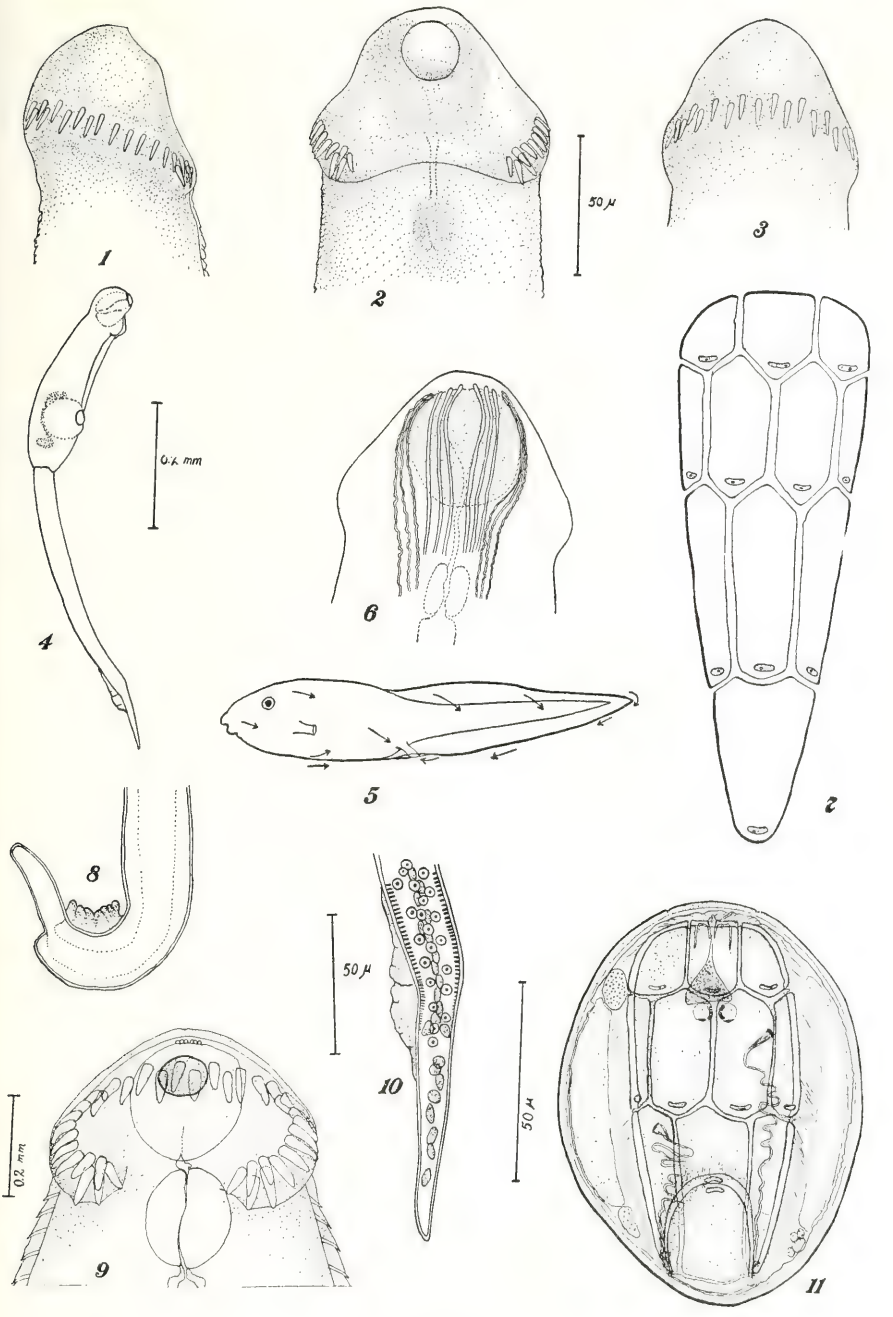


PLATE I

PLATE II

FIG. 12.—Camera lucida drawing of a fixed specimen of the cercaria, ventral view. Excretory system added free-hand from drawings made from living specimens.

FIG. 13.—Camera lucida drawing of a 20 mm specimen from a muskrat. Ventral view.

FIG. 14.—Camera lucida drawing of an 8.8 mm specimen from a chicken, experimentally infected. Ventral view.

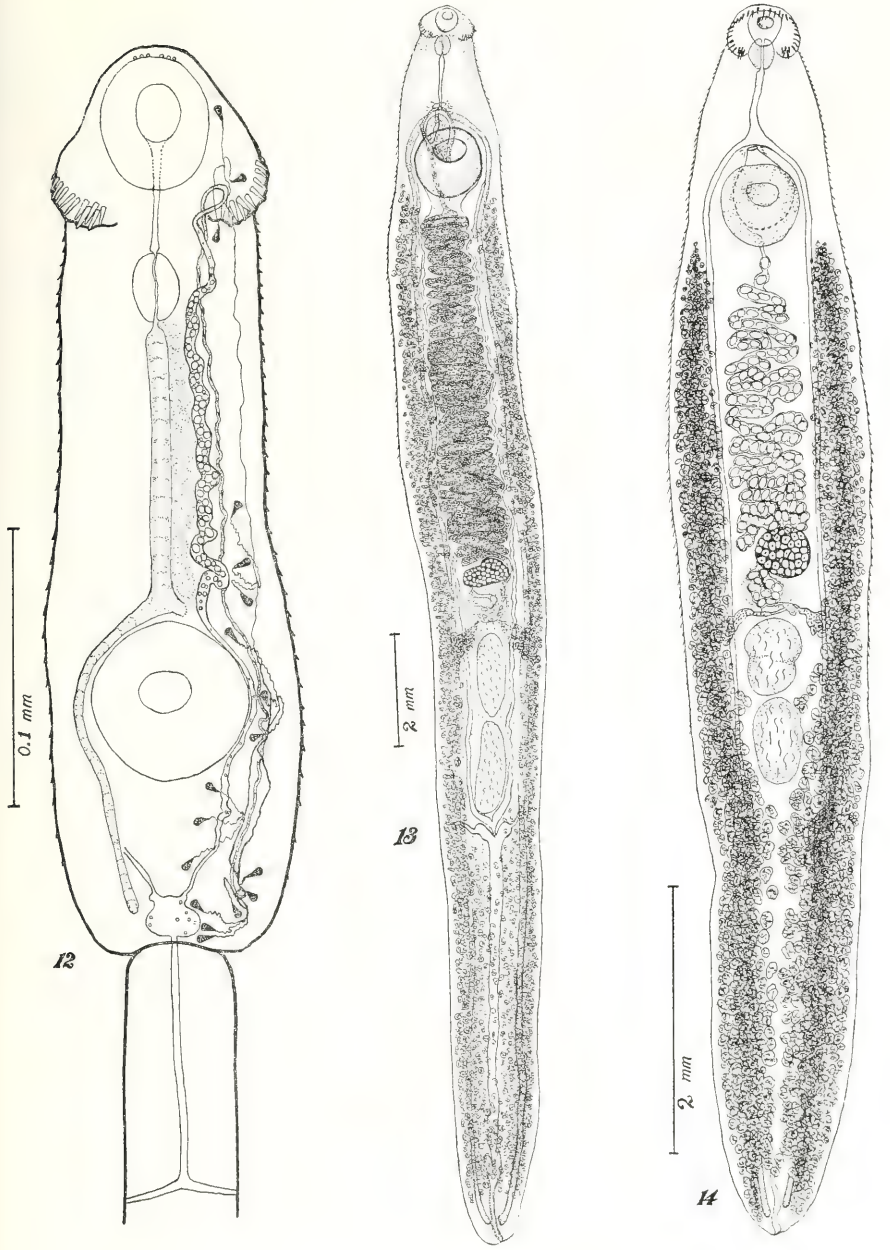


PLATE II

PLATE III

FIG. 15.—Cephalic spines of *Echinostoma cinetorchis.* Text-fig. 1, Ando and Ozaki, 1923. (From Dollfus, 1925, text-fig. 2.)

FIG. 16.—Cephalic spines of *E. mendax.* (From Dietz, 1910, text-fig. A.)

FIG. 17.—Cephalic spines of *E. paraulum.* (From Dietz, 1910, text-fig. B.)

FIG. 18.—Cephalic spines of *E. echinocephalum.* (From Dietz, 1910, text-fig. C.)

FIG. 19.—Cephalic spines of *E. columbae.* (From Bolle, 1925, text-fig. 1.)

FIG. 20.—Cephalic spines of *E. columbae.* (From Zunker, 1925, text-fig. 2. Scale is 0.1 mm.)

FIG. 21.—Cephalic spines of *Echinoparyphium (?Echinostoma) paraulum.* (From Sprehn, 1932, text-fig. 8.)

FIG. 22.—*Echinostoma miyagawai.* (From Ishii, 1932, Plate II, fig. 10.)

FIG. 23.—Cephalic spines of *E. miyagawai.* (From Ishii, 1932, Pl. II, fig. 11.)

FIG. 24.—*Echinostoma columbae.* (From Zunker, 1925, text-fig. 1. Scale is about 1 mm.)

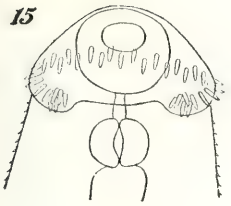
FIG. 25.—Cephalic spines of *E. echinatum (= revolutum).* (From Kowalewski, 1895, Plate 8, fig. 1.)

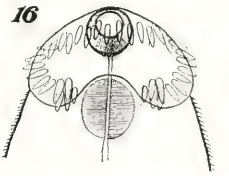
FIG. 26.—*"Fasciola revoluta" (= Echinostoma revolutum)* as figured in the original description. (From Froelich, 1802, Plate II, fig. 7.)

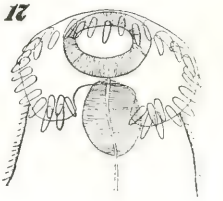
FIG. 27.—Cephalic spines of *E. sudanense.* (From Odhner, 1911, Plate III, fig. 13.)

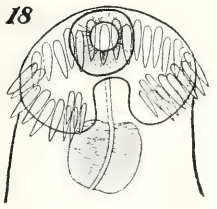
FIG. 28.—Cephalic spines of *E. echinatum (= revolutum)* from a goose. (From Looss, 1899, Plate XXIV, fig. 7.)

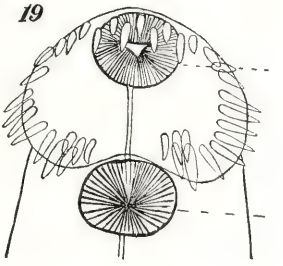
FIG. 29.—Cephalic spines of *E. echinatum* (?syn. *limicoli*) from *Machetes pugnax.* (From Looss, 1899, Plate XXIV, fig. 6.)

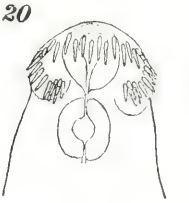


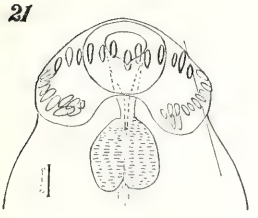


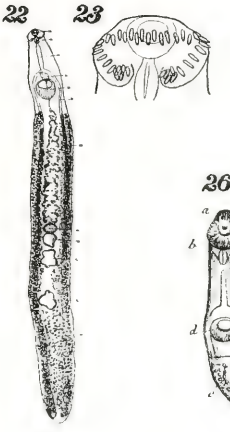


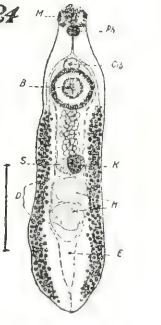


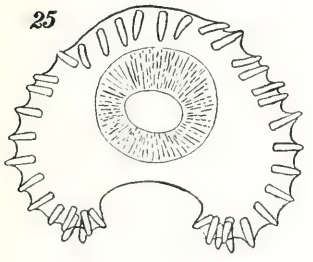


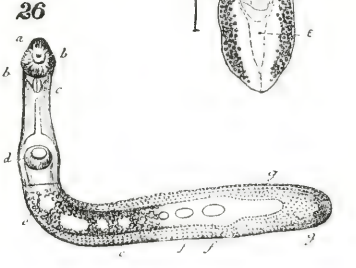


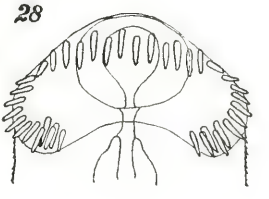












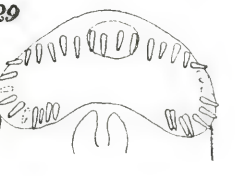


PLATE III

BIBLIOGRAPHY

ACKERT, JAMES E., *et al.*

1931a. Resistance of Chickens to Parasitism Affected by Vitamin A. Amer. Jour. Hygiene, 13:320-336.

1931b. Resistance of Chickens to Parasitism Affected by Vitamin B. Amer. Jour. Hygiene, 13:337-344.

ANAZAWA, K.

1929. The First Instance of *Echinostoma revolutum* in Man and Its Infection Route. Taiwan Ig. Kw. Z., 288:221-241. (Abs. 25, Japan. Jour. Zool., 3, 1930).

ANDO, A., and OZAKI, Y.

1923. Sur quatre nouvelles espèces de trématodes de genre Echinostoma. Dobutsu G. Z., 35:108-119.

BARKER, F.

1915. Parasites of the American Muskrat (*Fiber zibethicus*). Jour. Parasitol., 1:184-197.

1916. A New Monostome Trematode Parasitic in the Muskrat with a Key to the Parasites of the American Muskrat. Trans. Amer. Micros. Soc., 35:175-184.

BAYLIS, H.

1929. A Manual of Helminthology. 303 pp. London.

BOLLE, E.

1925. Über einen Taubentrematoden aus der Gattung Echinostomum. Deutsche tierärztl. Wochenschr., 33:529-531.

BROWN, F.

1926. Some British Freshwater Larval Trematodes with Contributions to Their Life-histories. Parasitol., 18:21-34.

1931. Some Freshwater Larval Trematodes from Cheshire. Parasitol., 23:88-98.

CORT, W. W.

1914. Larval Trematodes from North American Freshwater Snails (Preliminary Report). Jour. Parasitol., 1:65-84.

1915. Some North American Larval Trematodes. Illinois Biol. Monogr., 1:447-532.

DIETZ, E.

1909. Die Echinostomiden der Vögel. Zool. Anz., 34:180-192.

1910. Die Echinostomiden der Vögel. Zool. Jahrb., Supple., 12:265-512.

DOLLFUS, R.

1925. Distomiens Parasites de Muridae du Genre Mus. Ann. Parasitol., 3:85-102, 185-205.

DUBOIS, G.

1928. Les Cercaires de la Région de Neuchâtel. Bull. Scc. Sc. nat. Neuchâtel, 53:1-160.

ERCOLANI, G.

1881. Dell'adattamento della specie all'ambiente. Nouve richerche sulla storia genetica dei Trematodi. Mem. Acad. d. Sci. d. Istit. di Bologna, 2:239-334.

1882. Dell'adattamento della specie all'ambiente. Nouve richerche sulla storia genetica dei Trematodi. Mem. Acad. d. Sc. d. Istit. di Bologna, 3: 43-111.

FALLIS, A.
1934. A Note on Some Intermediate Hosts of *Echinostoma revolutum* (Froelich). Proc. Helminthol. Soc., 1:4-5.

FAUST, E.
1918. Studies on Illinois Cercariae. Jour. Parasitol., 4:93-110.

FROELICH, J.
1802. Beiträge zur Naturgeschichte der Eingeweidewürmer. Naturforscher (Halle), 29:5-96.

GENERALI, G.
1881. Sul *Distoma echinatum* nel cane. Spallanzani, Modena, ser. 2, 10:614-615.

HASSALL, A.
1896. Check List of Animal Parasites of Chickens, U. S. Dept. Agric., Bureau Animal Industry, Circular 9.

HEELSBERGEN, T. VAN.
1927. Echinostomiasis bij de duif door Echinostoma. Tijdschr. Diergeneeskde, 54:414-416. (Abs. Deutsche tierärztl. Wochenschr., 35:852).

ISHII, N.
1932. Studies on Bird Trematodes. I. Bird Trematodes in Japan. II. Four New Bird Trematodes. Jikken Ig. Z., 16 (English text: Japan. Jour. Exp. Med., 11:91-100 (1933)).

IWATA, S., and TAMURA, O.
1933. Some Intestinal Parasites in the Duck from Japan. Annot. Zool. Japon., 14:1-6.

JOHNSON, J.
1920. The Life Cycle of *Echinostoma revolutum* (Froelich). Univ. California Public. Zool., 19:335-388.

JOHNSTON, S.
1913. On some Queensland Trematodes. Quart. Jour. Micr. Sci., 59:361-400.
1916. On the Trematodes of Australian Birds. Jour. Proc. Roy. Soc. N.S. Wales, 50:187-261.

KOWALEWSKI, M.
1895. Studya helminthologiczne I. Rozpr. Wydz. mat.-przyrodn. Akad. Umiejet. Kraków, ser. 2, 9:349-367.
1897. O przedstawicielach rodzaju Echinostomum Rud. u Kaczki i kury. (Résumé: Sur les représentants du genre Echinostomum Rud. chez la poule et le canard domestique). Kosmos, Lemberg, 21:554-565.

KRAUSE, C.
1925a. Gehäuftes Sterben bei Tauben durch Echinostomiden (Vorläuf Mitteilung). Berlin. tierärztl. Wochenschr., 41:262-263.
1925b. Bemerkungen zu der Vorstehenden Arbeit von Dr. M. Zunker: "*Echinostoma columbae* n. sp., ein neuer Parasit der Haystaube." Berlin. tierärztl. Wochenschr., 41:484-485.

KRULL, W. H.
1935. A Note on the Life History of *Echinostoma coalitum.* Barker and Beaver, 1915. Proc. Helminthol. Soc. Washington, 2:76.

KURISU, T.
1932. Studies on Trematodes of Domestic Fowl in Japan (Japanese). Kumamoto Ig. Kw. Z., 8:283-298. (Abs. 150, Japan. Jour. Zool., 5, 2, 1933).

LAW, R., and KENNEDY, A.
1932. Parasites of Furbearing Animals. Bull. Dept. Game and Fisheries, Ontario, 4:1-30.

LEIDY, JOS.
1888. On the Trematodes of the Muskrat. Proc. Acad. Nat. Sc. Phila., 40-126.
LOOSS, A.
1893. Ist der Laurer'sche Kanal der Trematoden eine Vagina? Centralbl. f. Bakteriol. u. Parasitenk., Jena, 13:808-819.
1894. Die Distomen unserer Fische und Frösche. Biblioth. Zool., 16:1-296. (Stuttgart).
1899. Weitere Beitrage zur Kenntnis der Trematoden-Fauna Aegyptens, zugleich Versuch einer naturlichen Gliederung des Genus Distomum Retzius. Zool. Jahrb., Jena, Abt. f. Syst., 12:521-784.
LÜHE, M.
1909. Parasitische Plattwürmer. I. Trematodes. Die Süsswasserfauna Deutschlands, 17:1-217.
LUTZ, A.
1924. Untersuchung über die Entwicklungsgeschichte brazilianischer Trematoden. Spezieller Teil. I. Echinostomidae. Mem. Inst. Oswaldo Cruz, Rio de Janeiro, 17:75-93.
MARKOWSKI, ST.
1933. Untersuchungen über die Helminthenfauna der Raben (Corvidae) von Polen. Mem. acad. Polon. d. Sci. et d. Lettres. Ser. B., Sci. nat., 5:1-65.
MATHIAS, P.
1925. Recherches expérimentales sur le cycle evolutif de quelques Trématodes. Bull. biol. France Belgique, 59:1-123.
MIKI, T.
1923. On the secondary intermediate host of *Echinostoma cinetorchis* Ando and Ozaki. Aichi Ig. Kw. Z., 30:169-174.
MILLER, E. L.
1936. Studies on North American Cercariae. Illinois Biol. Monogr., 14: no. 2, 125 pp., 8 pls.
MORISHITA, K.
1929. Some Avian Trematodes from Japan, especially from Formosa: with a Reference List of All Known Japanese Species. Annot. Zool. Jap., 12:143-171.
MÜLLER, A.
1897. Helminthologische Mittheilungen. Arch. f. Naturg., 63:1-26.
NEUMANN, L.
1905. A Treatise on the Parasites and Parasitic Diseases of the Domesticated Animals. (Translation by G. Fleming; revised and edited by J. Macqueen). 697 pp. London.
NICOLL, W.
1914. The Trematode Parasites of North Queensland. II. The Parasites of Birds. Parasitol., 7:105-126.
ODHNER, T.
1911. Nordostafrikanische Trematoden, grösstenteils vom Weissen Nil. Results Swed. Zool. Exped. White Nile, Part IV, 166 pp.
OTTE, W.
1926. Betrachtungen über Geflügeltrematoden in Liv- und Kurland. Berlin. tierärztl. Wochenschr., 42:444-446.

PAGENSTECHER, H.
1857. Ueber Erziehung des *Distoma echinatum* durch Fütterung. Arch. f. Naturg., Berlin, 1 (J. 23):244-251.

PICARD, W.
1930. Pluimveeziekten in Nederlandsch-Indie. Echinostomiasis bij. duiven. Ned.-Ind. Blad. Diegeneesk., 42:424-428.

RAILLIET, A.
1895. Traité de Zoologie médicale et agricole. 2 ed. 1303 pp. Paris.

RASÍN, K.
1933. *Echinoparyphium recurvatum* (Linstow, 1873) und seine Entwicklung. Biol. Spisy Vysoké Skoly Zverol. Brno, C.S.R., 12:1-104.

REES, G.
1931. Some Observations and Experiments on the Biology of Larval Trematodes. Parasitol., 23:428-440.

RIECH, F.
1927. Beiträge zur Kenntnis der Echinostomiden. I. Der Lebenszyklus von *Echinoparyphium aconiatum* Dtz. II. *Cercaria laticaudata* n. sp. Centralbl. f. Bakt. u. Parasit. Abt. 1. Orig., 103:279-290.

RIVAS, D. DE
1932. Internal Parasitism; Diagnosis and Treatment. Amer. Jour. Trop. Med., 12:477-492.

RUDOLPHI, C. A.
1819. Entozoorum Synopsis cui accedunt mantissa duplex et indices locupletissimi. x + 811 pp., 3 pl. Berolini.

SEIFERT, R.
1926. *Cercaria limbifera,* eine neue echinostome Cercarie. Zool. Aug., 67: 112-119.

SEWELL, R.
1922. Cercariae Indicae. Indian Journ. Med. Res., Suppl. 10:1-370.

SIEBOLD, C. TH. VON
1837. Zur Entwickelungsgeschichte der Helminthen. K. F. Burdach's: Die Physiologie als Erfabrungswissenschaft. Bd. 2, 2 Aufl. Leipzig.

SKRJABIN, K.
1913. Vogeltrematoden aus Russisch-Turkestan. Zool. Jahrb. Abt. Syst. Ökol. Geogr. Tiere, 35:351-388.
1915. Trématodes des oiseaux de l'Oural. Ann. Mus. Zool. Acad. Sci. Petersburg, 20:395-417.

SPREHN, C.
1927. Echinostomiden bei Tauben. Deutsche tierärztl. Wochenschr., 25:451-455.
1930. Wichtige Endoparasiten des deutschen Hausgeflügels. Berlin. tierärztl. Wochenschr., 46:765-774.
1932. Lehrbuch der Helminthologie. 998 pp. Berlin.

SWALES, W.
1933a. A Review of Canadian Helminthology. I. The present status of knowledge of the helminth parasites of domesticated and semidomesticated mammals and economically important birds in Canada, as determined from work published prior to 1933. Canadian Jour. Res., 8:468-477.

Swales, W. (*continued*)

1933b. A Review of Canadian Helminthology. II. Additions to Part I, as determined from a study of parasitic helminths collected in Canada. Canadian Jour. Res., 478-482.

Taliaferro, W.

1929. The Immunology of Parasitic Infections. 414 pp. New York.

Tsuchimochi, K.

1924. On the Life Histories of Two Echinostome Trematodes. Dobutsu G. Z., 428:245-252.

1926. On Larval Flukes Infesting Limnaea in Formosa (Jap. Text with English Summary). Taiwan Ig. Z., 257:733-754.

Tubangui, M.

1932a. Trematode Parasites of Philippine Vertebrates. V. Flukes from Birds. Philippine Jour. Sci., 47:369-404.

1932b. Observations on the Life-histories of *Euparyphium murinum* Tubangui 1931, and *Echinostoma revolutum* (Froelich 1802). Philippine Jour. Sci., 47:497-513.

Vevers, G.

1923. Observations on the Life-Histories of *Hypodaerium conoideum* (Bloch) and *Echinostomum revolutum* (Froel.): Trematode Parasites of the Domestic Duck. Ann. Appl. Biol., 10:134-136.

Wedl, C.

1857. Anatomische Beobachtungen über Trematoden. Sitzungsb. d. k. Akad. d. Wissensch., Wien, math.-naturw. Cl., 26:241-276.

Wesenberg-Lund, C.

1934. Contributions to the Development of the Trematoda Digenea. Part II. The Biology of the Freshwater Cercariae in Danish Freshwaters. Mem. Acad. Roy. Sci. et Lettres Danemark, Copenhague, ser. 9, 5:1-223.

Wetzel, R.

1933. Zum Wirt-Parasitverhältnis des Saugwurms *Echinoparyphium paraulum* in der Taube. Deutsche tierärztl. Wochenschr., 41:772-775.

Yamaguti, S.

1933. Studies on the Helminth Fauna of Japan. I. Trematodes of Birds, Reptiles, and Mammals. Jap. Jour. Zool., 5:1-134.

Zeder, J.

1803. Anleitung zur Naturgeschichte der Eingeweidewürmer. 432 pp. Bamberg.

Zunker, M.

1925. *Echinostoma columbae* n. sp., ein neuer Parasit der Haustaube. Berlin. tierärztl. Wochenschr., 41:483-484.

INDEX

Ameiurus melas, 32, 66, 68
Anas brasiliense, 69
  platyrhynchos, 69
  superciliosa, 69
Anser anser, 69
  cinerius, 69
Anseranas semipalmata, 69
Aramides cayennensis, 43
Ascaridia lineata, 37
Bueteo lagopus-s. johannis, 69
Bufo americana, 32, 68
Cairina moschata, 10, 69
Canis familiaris (See Dog)
Carphibis spinicollis, 42
Cat, 32, 33, 70, 76, 77
Catfish (See *Ameiurus melas*)
Cavia porcellus (See Guinea pig)
Cercaria, Agilis group, 14
  Coronata group, 16
  Echinata group, 16
  Echinatoides group, 16
Cercaria echinata, 9, 15, 17, 18
  Echinoparyphii-aconiati, 17, 18
  Echinostomi-revoluti, 13, 17, 65, 67
  helvetica XXII, 18
    XXIII, 18
    XXIV, 18, 65
    XXV, 18
  limbifera, 17, 18
  trivolvis, 18, 65
Chamaepelia passerina, 43
Chemiscus pulchellus, 69
Chenopsis atrata, 69
Chicken (See Domestic fowl)
Coloeus monedula, 69
Columba domestica (See Pigeon)
Colymbus cristatus, 51, 69
Coracias garrula, 69
Corbicula producta, 68
Corvus cornix, 69
Crecisсus viridis, 43, 44
Crotophaga ani, 43
Dendrocygna viduata, 69
Distomum armatum, 40
  dilatatum, 40
  echinatum, 8, 9, 50, 56
  hérissé, 54
  oxycephalum, 40
Dog, 9, 32, 33, 76, 77
Domestic fowl, 33, 46, 69, 70, 73, 74, 77
Duck, 8, 9, 32, 33, 46, 50, 51, 69, 74, 77
Echinoparyphium, 28, 51, 52, 53, 65
  aconiatum, 15, 17, 40, 54, 55
    cercaria, 17
  contiguum, 40, 48, 55
  recurvatum, 11, 12, 53
  sp., 40, 42
Echinostoma, 28
  acuticauda, 41, 42, 56
  armatum, 56
  armigerum, 49, 55, 56
  callawayensis, 41, 42, 48, 56
  cinetorchis, 10, 32, 46, 47, 48, 55, 56, 88
  coalitum, 19, 49, 50, 56
  columbae, 51, 52, 53, 55, 56, 88
  dilatatum, 56
  distinctum, 44
  echinatum, 40, 54, 65, 88
  echinocephalum, 41, 45, 46, 52, 56, 88
  erraticum, 41, 43, 56
  limicoli, 54, 56
  mendax, 50, 51, 55, 56, 88
  microrchis, 41, 44, 56
  miyagawai, 46, 47, 55, 56, 88
  neglectum, 41, 43, 56
  oxycephalum, 56
  paraulum, 51, 52, 53, 55, 56, 88
  revolutum, acetabulum, 19, 20
    body spines, 21
    cercaria, 13, 67
      description, 14, 15, 16, 24, 25
      geographical distribution, 29, 67
      hosts, 30, 67
      related species, 17, 18
      synonyms, 17, 18
    collar, 20
      spines, 22
    diagnosis, 55
    genital system, 25
    geographical distribution, 29, 69, 70
    hosts, 29, 32, 56, 62, 69, 70, 72
    immunity against, 33, 71
    life history, 8
    metacercaria, 30, 31, 32, 68
    methods used, 10, 12, 14, 18, 19, 26, 56, 57
    miracidium, 10
    misdeterminations, 53
    morphology, 18, 78
    mother-redia, 13
    oral sucker, 20
    ova, 25, 26
    ovary, 19, 20
    pathology, 31, 57, 58, 59, 60
    pharynx, 20
    rate of maturity, 28, 77
    redia, 12
    self-fertility, 26

Echinostoma revolutum (continued)
size and form, 19, 20
spination of body, 21
crown, 22, 23, 24
synonyms, 39, 55, 56
testes, 20
transfer to pigeons, 26, 37, 62, 64, 77
uterus, 20
variation, induced, 27, 56, 62, 78
natural, 18, 56, 78
sp. (Leidy), 49
(Heelsbergen), 53
(Picard), 53
sudanense, 41, 42, 56, 88
Ephippiorhynchus senegalensis, 41
Epimys norvegicus (See Rat)
Euparyphium (Echinostoma?) murinum, 13
Fasciola revoluta, 88
Felis domestica (See Cat)
Fossaria abrussa, 68
modicella, 31, 68
Gallinula galeata, 43, 44
Gallus gallus (See Domestic fowl)
Gosling, 10
Guinea pig, 32, 33, 70, 76, 77
Helisoma trivolvis, 30, 57, 67, 68, 77
Hog (See Pig)
Homo sapiens, 70
Hyla regilla, 9
Hypoderaeum conoideum, 12
Lymnaea peregra, 67, 68
pervia, 67, 68
radix, 67, 68
stagnalis, 67, 68
stagnicolis, 8
swinhoe, 67
traski, 9, 68
Machetes pugnax, 54
Mareca americana, 70
penelope, 70
Marila marila, 9, 70
Meleagris gallopavo, 70
Mouse, 9, 32, 33, 70, 75, 77
Mus musculus (See Mouse)
Musculium partumeium, 68
Muskrat, 25, 40, 42, 48, 49, 50, 70
Neochen jubata, 70
Notophthalamus torosus, 9
Nycticorax violaceus, 43
Nyroca marila, 70
fuligula, 70
Oidemia nigra, 70
Ondatra zibethica (See Muskrat)
Paludina sp., 8
Pernis apivorus, 70
Phalocrocorax carbo, 54
Philomachus pugnax, 70
Physa halei, 68
gyrina, 30, 57, 67
occidentalis, 9, 67, 68
rivalis, 43, 67, 68
Pig, 32, 33, 70, 76, 77
Pigeon, 10, 32, 33, 34, 53, 69, 77
Pisidium sp., 31, 68
Planaria, 9, 68
Planorbis corneus, 8
sp., 43, 67, 68
Pseudosuccinea columella, 31, 67, 68
Rabbit, 32, 33, 70, 76, 77
Rana esculenta, 9, 32, 68
catesbiana, 31, 32, 68
japonica, 32, 68
nigromaculata, 32, 68
pipiens, 30, 31, 32, 57, 68
rugosa, 32, 68
Rat, 9, 32, 33, 70, 74, 77
Scolopax rusticola, 70
Scopus umbretta, 41
Sphaerium sp., 31, 68
Spirulina sp., 43
Stagnicola palustris, 67, 68
Sus scrofa (See Pig)
Swan, 70
Tadorna tadorna, 70
Tadpole, 10, 15, 31, 32, 68 (See also, Bufo, Hyla, Notophthalamus, and Rana)
Viviparus sp., 68

UNIVERSITY OF ILLINOIS-URBANA
3 0112 065097195