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PROCEEDINGS OF THE HELMINTHOLOGICAL SOCIETY OF WASHINGTON

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Observations on the Morphology and Biology of *Longibucca eptesica N. Sp.* (Nematoda: Cylindrocorporidae) Parasitic in the Bat[1]

JOHN R. ELSEA

The genus *Longibucca* was founded by Chitwood (1933), who described the species *Longibucca vivipara* from the stomach of a South American snake, *Pseudoboa cloelia.* The only other species of the genus is *Longibucca lasiura,* described by McIntosh and Chitwood (1934) from the red bat, *Lasiurus borealis,* taken at Washington, D. C. Two other species of bats were recorded by Tromba and Smith (1952) as new hosts for *L. lasiura.*

Members of the genera *Longibucca* and *Goodeyus* are the only known parasitic cylindrocorporid nematodes; the known species of other genera are saprophagous (Chitwood and Chitwood, 1950). Because of this close relationship between free-living and parasitic nematodes, it was thought worthwhile to study certain of the biological features of a member of the genus *Longibucca.*

Longibucca Chitwood, 1933

GENERIC DIAGNOSIS EMENDED: Cylindrocorporidae; Oral opening surrounded by 6 indistinct lips each bearing a papilla of the inner circle; external circle of 6 papillae. Amphids situated at base of lateral lips; amphidial opening round, minute. Esophagus consisting of corpus, isthmus, and pseudobulb, with corpus well set off from isthmus or not. Lateral alae present. MALE: testis single, reflexed, caudal alae absent; tail attentuated; 5 or 6 pairs of genital papillae; 2 curved spicules, short, equal; gubernaculum present. FEMALE: vulva posterior, near anus; one reflexed ovary; viviparous. Found in stomach of snakes and bats.

TYPE SPECIES: *Longibucca vivipara* Chitwood, 1933.
OTHER SPECIES:
Longibucca lasiura McIntosh and Chitwood, 1934
Longibucca eptesica, n. sp.

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[1]From the Department of Biology, The Catholic University of America, Washington, D. C. This paper is based on the author's dissertation submitted in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

The author is deeply grateful to Dr. B. G. Chitwood who suggested the problem and to Dr. Edward G. Reinhard who directed the course of the investigation. He also wishes to extend his appreciation to Drs. Merritt P. Sarles and Henry E. Wachowski who read the manuscript and offered helpful suggestions. Mr. Allen McIntosh and Mrs. M. B. Chitwood of the Zoological Division, Bureau of Animal Industry, Beltsville, Maryland were most helpful in assisting in certain technical aspects of the problem. Mss Nancy Rogers of the Washington Grotto, National Speleological Society, and Brother G. Nicholas, La Salle High School, Cumberland, Maryland gave generously of their time and knowledge in aiding the author in the collection and identification of bats used in this study.

Longibucca eptesica, n. sp.

SPECIFIC DESCRIPTION: *Longibucca;* Stoma 45-53 μ long. Esophagus 46-51 μ long with cylindroid corpus indistinctly set off from isthmus and pseudobulb. Ratio of length of stoma to length of esophagus approximately 1:1. Excretory pore 78-92 μ from head. MALE: 485-647 μ long by 20-32 μ wide. Tail with slight terminal enlargement. Genital papillae: one pair preanal subventral, 3 pairs postanal subventral, and 2 pairs postanal subdorsal. Spicules 26-29 μ long. Gubernaculum 22-25 μ long (measured along internal rim) and 6-8 μ in thickness. FEMALE: 710-825 μ long by 28-44 μ wide. Tail conical. 2-8 developing larvae in uterus.

HOST: *Eptesicus fuscus fuscus* (Beauvois 1796)[2]

LOCATION: Stomach

LOCALITY: Frederick County, Virginia

TYPE SPECIMEN: USNM Helminthological Collection No. 47893

PARATYPES: USNM Helminthological Collection No. 47894

KEY TO SPECIES OF *LONGIBUCCA*

1. Male with 5 pairs of genital papillae, male tail conical ..... *L. lasiura*
   Male with 6 pairs of genital papillae, male tail not conical 2
2. Male tail curved dorsally ..... *L. vivipara*
   Male tail with slight terminal enlargement ..... *L. eptesica, n. sp.*

ADULT MORPHOLOGY[3]

A. EXTERNAL CUTICLE. The external cuticle is thin and very finely striated. The most striking feature of the cuticle is the lateral alae which are present in all stages, including first stage larvae *in utero*. These prominent projections (Fig. 2, A) arise from a thickened cuticular plate and extend practically the entire length along either side of the body. In molting specimens the alae resemble cordons (Fig. 3, E).

In stained sections the cuticle has a dark bluish color, noticeable particularly in thickened portions such as the lateral alae and the vaginal lining.

In the male there are 5 pairs of genital papillae. These are located either subventrally or subdorsally and appear to be somewhat sucker-like. Probably they serve as copulatory aids.

B. HYPODERMIS. The hypodermis is extremely thin and presumably a syncytial mass, since no cell boundaries were observed. It is somewhat thickened in the post-anal portion of the body, but elsewhere it is merely a thin membrane separating the cuticle and the somatic musculature. In favorable sections a slight elevation of the hypodermis is present in the lateral, dorsal, and ventral fields, suggesting the presence of longitudinal chords. None of the structures customarily present in chordal tissue, such as excretory ducts, nerves, etc. were observed, undoubtedly because of their small size.

C. SOMATIC MUSCULATURE. The somatic musculature forms the innermost layer of the body wall. It consists of a single layer of longitudinal platymyarian cells, usually 5 to 7, in each of the 4 sublateral fields (Fig. 2, C).

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[2]Organisms apparently the same were also found in the stomachs of 2 other species of bats: *Myotis lucifugus* (LeConte 1831) and *Pipistrellus subflavus* (Cuvier 1832).

[3]The morphological observations are based chiefly upon examination of *L. eptesica* contained in serial sections (6_{μ}) of *Pipistrellus subflavus* stomachs stained with Harris' hematoxylin and eosin. Adult worms were also sectioned individually. In both cases the technique of preparation was that previously reported by the author (1951). The specimens used for the whole mount studies were placed in 0.85% NaCl solution and inactivated with gentle heat. Other worms were preserved in a 10% formalin solution. All drawings were made with the aid of the camera lucida. The magnifications of drawings and photographs are indicated in the figures.

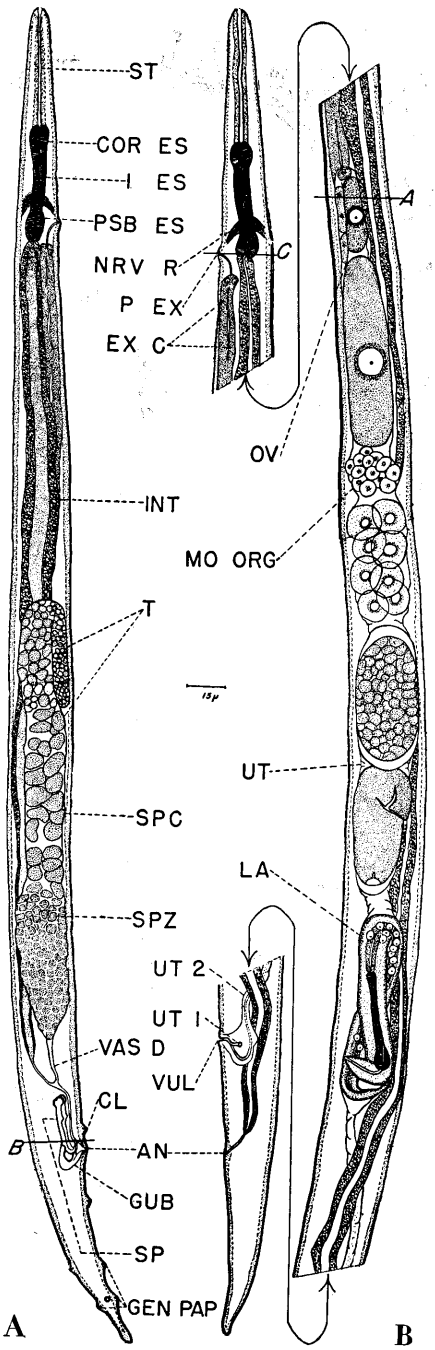

FIG. 1. *Longibucca eptesica.* A—Lateral view of adult male. B—Lateral view of adult female. Note: The levels at which the cross-section drawings in Figure 2 were made are indicated.

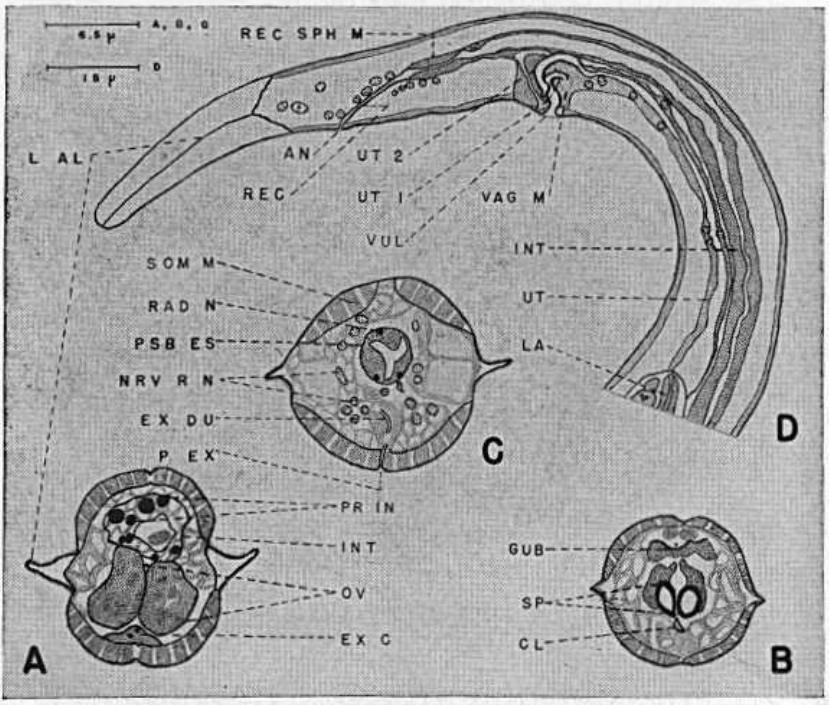

FIG. 2. *Longibucca eptesica.* A—Cross-section through adult female in region of ovarian flexure. B—Cross-section through adult male at level of spicules and gubernaculum. C—Cross-section through adult female at level of excretory pore. D—Longitudinal section through posterior end of adult female.

Abbreviations for Figures:

al	ala	ov	ovary
an	anus	p ex	excretory pore
c	cell	pap	papilla
cl	cloaca	pr. in	protein inclusion body
cor es	corpus region of esophagus	psb es	pseudobulb region of esophagus
cut	cuticle	rad	radial
du	duct	rec	rectum
er muc	eroded area of mucosa	som	somatic
ex	excretory	sp	spicule
gen	genital	spc	spermatocyte
gub	gubernaculum	sph	sphincter
i es	isthmus region of esophagus	spz	spermatozoa
		st	stoma
int	intestine	t	testis
l	lateral	ut	uterus
la	larva	vag	vagina
m	musculature	vas d	vas deferens
mo org	morular organ	vul	vulva
n	nucleus	w	worm
nrv r	nerve ring		

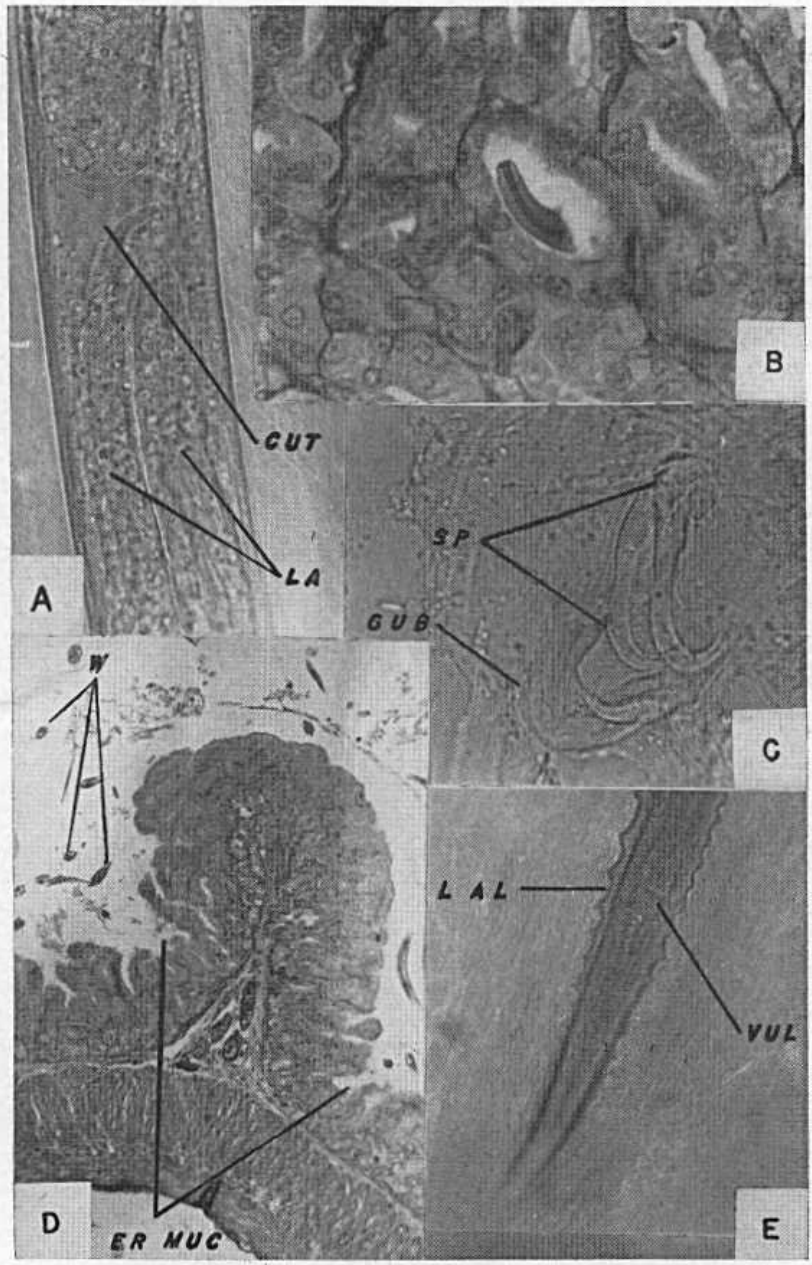

FIG. 3. *Longibucca eptesica.* A—Posterior region of mature female showing molt of larva *in utero.* B—Anterior end of mature male in gastric gland of *Pipistrellus subflavus.* C—Spicules and gubernaculum dissected from adult male. D—Surface of gastric mucosa of *P. subflavus* showing slight erosion produced by worms. E—Posterior end of pre-adult female in process of molting. Note: Magnifications for photomicrographs are: A, C, oil immersion; B, E, high dry; D, low dry.

D. EXCRETORY SYSTEM. The excretory pore, opening at the summit of a small elevation, is located in the region of the base of the esophagus (Fig. 2, C). Leading from the pore is a minute cuticularly lined duct, which appears to become embedded in the juncture of 2 excretory cells. These cells are extremely elongate, extending posteriorly in the female a distance approximately equal to 1/9 the body length. The cells are minutely granular and have a nucleus near their anterior end (Fig. 1: A, B).

E. DIGESTIVE SYSTEM. *Stoma.* The comparatively enormous length of the stoma is a prominent feature emphasized by the generic name of the organism. The cheilorhabdions flare out very slightly at the oral end to form a funnel-shaped opening. The protorhabdions forming the body of the stoma are of the same thickness throughout its length, and the internal diameter of the stoma is also uniform. The telorhabdions which form the base of the stoma are somewhat thickened and form a glottoid apparatus (Fig. 1: A, B).

Surrounding the stoma is a very thin sheath which contains several elongate, darkly-staining nuclei, probably a part of the arcade tissue.

Esophagus. The esophagus is divisible into three portions: corpus, isthmus and pseudobulb. The corpus and pseudobulb are slightly ovoid while the isthmus is of uniform diameter. (Fig. 1: A, B).

Radial nuclei were observed in the corpus and pseudobulb but owing to their small size the number could not be ascertained with certainty. They appear as concentrically arranged, ovoid, darkly-staining bodies (Fig. 2, C). The isthmus is anucleate. The dorsal esophageal gland orifice is located in the posterior part of the cropus (Fig. 1, A). The esophageal lumen is triradiate and is lined by a thin cuticular layer (Fig. 2, C).

Intestine. The intestine is a prominent tube which occupies a considerable portion of the body cavity, except where it is pushed aside and compressed by the reproductive organs (Fig. 1: A, B).

In section, from 2 to 4 cells are seen to compose the intestinal wall. The intestinal cells contain many small globular inclusions which appear greenish in living specimens. These globules probably represent reserve food material in the form of oil and protein (Chitwood, M. D., 1951; Elsea, 1951). In sectioned material the intestinal cells appear somewhat vacuolate, probably due to the loss of fatty materials in routine histological preparation. The protein granules stain either an intense blue or a faint yellowish-red, suggesting that there is more than one type of protein inclusion body.

The intestinal contents appear to be a finely granular material suggestive of bacteria. This material presents the same appearance at all levels of the digestive tract in living, preserved, and sectioned specimens. At no time was any cellular material observed inside the digestive tract.

The posterior end of the intestine is surrounded by a circular band of muscle tissue, which serves as a rectal sphincter (Fig. 2, D).

Rectum and Anus. In the female the rectum is a short tube which is heavily lined with cuticle (Fig. 2, D). In the male the cloaca which is formed from the junction of the intestine and vas deferens is finer and more elongate; it passes ventral to the spicules and opens to the outside via the anus (Fig. 2, B). In both sexes the anal orifice appears circular in a surface view and is surrounded by a slightly raised cuticular rim.

F. REPRODUCTIVE SYSTEM. In both sexes the gonad is a single tubular structure which extends through a considerable part of the body. There is a single anterior flexure (Fig. 1: A, B).

FEMALE: In the germinal zone of the ovary the oocytes are comparatively few in number and are arranged in single file, one oocyte filling the entire lumen. Near the ovarian flexure there is an accumulation of 15-20 round cells (Fig. 1, B) which probably provide the egg shell, since it is not until after the oocytes pass this structure that an outer membrane is observed. This accumulation of cells is probably homologous to the morular organ in *Meloidogyne hapla* (Elsea, 1951). The outer membrane is not a rigid structure but stretches with growth of the developing embryo.

In the uterus one can trace the gradual development of the embryo through cleavage stages to the ovoid and finally the fully formed vermiform larva, which is located in the terminal portion of this organ (Fig. 1, B). Occasionally two apparently fully developed larvae are present in the uterus simultaneously. Other Cylindrocorporidae (*Myctolaimus, Goodeyus,* and *Cylindrocorpus*) are oviparous and contain only one or at the most 2 eggs in the uterus. The occurrence of viviparity and the greater number of young produced in *L. eptesica* may be construed as a slight departure toward a parasitic existence.

The terminal portion of the uterus is cuticularly lined and is surrounded by extensive muscles (Fig. 2, D). Joining the functional uterus to form the vagina vera is the remnant of a second anterior uterus which is in the form of a medially-extending cuticularly lined blind tube (Fig. 2, D; 1, B). The vagina vera is short and possesses a thick cuticular lining. The lips of the vulva are somewhat elevated above the body surface and on surface view the transverse vulva is an elongated narrow opening. (Fig. 3, E).

MALE: The testis is divided into an anterior germinal zone and a posterior growth zone. In the latter area the spermatocytes are large and granular in appearance. The lower part of the gonad is somewhat expanded and appears to function as a storage space for spermatozoa. The vas deferens is a short tube which joins the lower end of the intestine to form the cloaca (Fig. 1, A).

The mature spermatozoa are round and contain coarse granules (Fig. 1, A). Spermatozoa were examined in the male gonad, in the uterus of the female, and expressed from the male gonad into warm saline solution, but at no time was motility observed.

The spicules[4] are best examined when dissected from the body (Fig. 3, C). Each spicule possesses a rather large capitulum and a ventral tubercle approximately 1/3 spicule length behind the capitulum. The terminal portion of the blade is curved anteriad. The gubernaculum is located postero-dorsal to the spicules and is curved along both margins; the inner margin is heavily sclerotized. The gubernaculum possesses a prominent postero-dorsal thickening and a distal bifurcation (Fig. 3, C).

DEVELOPMENTAL STAGES

Within the uterus of the female *L. eptesica* there is a complete series of developmental stages from the egg through cleavage stages to the fully formed larva (Fig. 1, B). In the terminal portion of the uterus, the larvae frequently are surrounded by a cast cuticle (Fig. 3, A). That this is a molt rather than the stretched egg membrane is probable, since it is not closely applied to the body.

[4]Reexamination of *L. lasiura* from the type host showed that there are 2 equal spicules which are completely separated from the gubernaculum, rather than fused as originally reported by McIntosh and Chitwood (1934).

In the lumen of the bat stomach, organisms ranging from a size only slightly larger than that of the larvae *in utero* to the adult size are encountered. Only rarely are molting specimens found in the stomach. These are inactive and the cast cuticle with its peculiar cordon-like lateral alae (Fig. 3, E) may be recognized without difficulty.

The developing larvae are similar to the adult in body proportions and contour except that the youngest larvae have a terminal enlargment of the tail (Fig. 1, B). This condition is retained in the adult male but the female tail is conical.

The digestive system is remarkably constant throughout the life of the individual. For example, in a series of 15 larval *L. eptesica* ranging in size from 194 to 470 μ, the stoma and esophague averaged 34 μ and 46 μ in length respectively. Thus it is seen that the esophagus remains fairly constant in size while the stoma elongates during the pre-adult stages. The intestine, rectum and anus present essentially the same appearance in all stages.

The reproductive system is recognizable in first stage larvae as 4 cells located approximately 60% of the body length from the head. With growth of the individual the gonad elongates to become a very thin tubular organ. In the pre-adult female the flexure of the ovary is formed and developing oocytes are present. Just anterior to the anus the vaginal primordium is recognizable by the beginning sclerotization. In the pre-adult male the testis is well formed and contains from 40-50 germinal cells, some of which appear to be fully formed spermatozoa. There is a slight sclerotization adjacent to the dorsal wall of the gonad just anterior to the anus; this probably represents the spicular primordium.

DISTRIBUTION AND PREVALENCE OF INFECTION

The distribution of *Longibucca eptesica* in the stomachs of bats from the various colonies sampled appears quite irregular.

The greatest prevalence of infection was observed in a group of 25 *Pipistrellus subflavus* collected from Horse Cave, Cumberland, Maryland. All of these animals contained from 25 to 50 worms.

Four colonies of the big brown bat, *Eptesicus fuscus fuscus*, were examined. Only one of these showed a fairly high prevalence of infection; 69% of the bats examined (13 individuals) contained from 30-50 worms. In another colony, located 12 miles away, 15 bats were negative while the remainder of the 33 examined contained from 1 to 70 worms. One colony, located near Washington, D. C., had a very slight infection; 6 of 10 bats were infected with an average of less than 5 worms while the remainder were negative. From the fourth colony were collected 16 young bats of less than 1 month of age and 4 adults; none of these animals contained worms.

Infection is considerably less in the little brown bat, *Myotis lucifugus*. One colony (83 individuals) examined in the present study showed 23% infection with *L. eptesica;* infected bats contained from 1 to 80 worms each. A group of 25 *M. lucifugus* collected from Horse Cave showed a 20% infection with usually less than 10 worms per stomach. From another colony, a total of 111 bats in three samples collected at different times of the year failed to show any worms. It was concluded therefore that animals in this colony were infection-free; they were the bats used in the experimental infection studies described later.

Other helminths encountered in bat stomachs examined included a small lecithodendriid tremotode, 5 to 30 specimens of which were present in practi-

cally all stomachs examined. Nematodes encountered were *Rictularia sp.,* encysted third stage larvae of *Physocephalus sexalatus,* and a species of *Capillaria,* similar in most respects to *C. speciosa* (Van Beneden, 1873).

HOST-PARASITE RELATIONS

The original description of *Longibucca lasiura* was based on material supposedly collected from the small intestine of *Lasiurus borealis.* The other literature reports, however, and the results of the examinations made in the present study show all species of *Longibucca* to be confined to the stomach. Repeated examinations and charcoal cultures of the intestinal contents and feces of infected bats were negative. In addition, in a complete necropsy of approximately 50 bats *Longibucca eptesica* was not found outside the stomach. In an examination of the type host conducted in 1939 by Mr. Allen McIntosh of the Zoological Division, B. A. I., *Longibucca lasiura* was reported from the stomach; furthermore *L. vivipara* is also found in this location. In view of this information, the occurrence of *L. lasiura* in a location other than the stomach is highly doubtful.

Chitwood (1933) reported a deep penetration of *L. vivipara* into the stomach wall of its host, *Pseudoboa cloelia;* he found larval and adult worms in the mucosa, submucosa, and even in the muscular coat. In addition to freely migrating worms, some partially degenerate forms were found surrounded by tissue which showed some slight cellular reaction.

In *Eptesicus fuscus fuscus* McIntosh and Chitwood (1934) reported adult and larval *L. lasiura* embedded in the depths of the gastric glands and interglandular tissue. They found several adults in a layer of mucus and desquamated epithelium which also contained lymphocytes and polymorphonuclear leucocytes. They concluded that the tissue of the stomach showed no marked pathological reaction.

Our findings in sectioned material from *P. subflavus* containing *L. eptesica* agree with those of McIntosh and Chitwood (1934). Within the stomach the nematodes are largely found lying on the surface of mucosa, either free or surrounded by a small amount of mucus-like material, which appears to contain cellular fragments. Occasionally the head of a worm will be found inserted slightly in a gastric gland (Fig. 3,B); penetration, however, is never great in such cases and it appears that the presence of the head in the gland is merely fortuitous.

When several nematodes are localized in one portion of the stomach, it appears at times as though there is a slight sloughing and erosion of the surface layer of the gastric epithelium in that area (Fig. 3, D). In no case observed, however, was such damage extensive.

Apparently *L. eptesica* is not a cause of mortality in bats. For example, in one series of 83 *Myotis lucifugus* examined, 19 containing worms were encountered. Of these, 4 were found dead in their cages and the remaining 15 cases were discovered from examination of freshly killed bats. Deaths among the remaining 64 bats (uninfected) occurred at about the same rate and, in all cases, is attributed to diminution of natural food reserves and maintenance in unnatural surroundings.

There was no significant difference observed between the size (measured by wing spread) or sex of infected and non-infected animals.

CULTURE ATTEMPTS

Because of the close relationship of *L. eptesica* to certain freeliving nema-

todes, the possibility that the organism could be maintained *in vitro* appeared promising. Consequently male, female, and larval worms were placed on the following media at 37°, 23°, and 7° C.:

Solid: nutrient agar
nutrient tryptose agar
Nigon's agar (vide Nigon, 1949):
plain
with cholesterol
overlaid with a solution of baker's yeast
without lecithine
liver infusion agar at pH of 5.5 and 7.0:
plain
overlaid with a solution of baker's yeast

Liquid: liver infusion broth at pH of 3.0, 5.5, and 7.0
sterile rabbit serum
Locke's solution
Locke-serum medium
Ringer's solution
Ringer-serum medium
0.85% NaCl solution:
plain
with 1% human erythrocytes
with 1% bat erythrocytes

In another series of experiments minced bat stomachs and livers were placed with each of these media.

Immediately after removal from the bat stomach the worms were rinsed thoroughly in 3 changes of sterile saline before placing on media. In some cases sterile plates inoculated with the worms and incubated at 37° C. developed no bacterial growth during a 48 hour period. In other cases, however, bacterial and mold colonies did develop.

No significant difference was observed in survival time between worms which were maintained in assocation with bacteria and those which were not. Likewise, the addition of minced bat tissues had no effect upon the survival time. The results in all cases were uniformly poor, the survival time being less than 48 hours at 37° C. and never more than 4 or 5 days at 23° C. or 7° C. The worms survived longer in 0.85% NaCl solution at 7° C. than in any of the above media. They were inactive but survived for periods up to 11 days, when this solution was renewed daily. In no case was reproduction of the organisms observed.

LIFE CYCLE STUDIES

In an attempt to discover a possible intermediate anthropod host for *L. eptesica* a number of external parasites, including bedbugs (*Cimex* sp.), fleas, mites, and ticks, were removed from infected bats and examined for nematodes. In addition, several coprophagous beetles collected from bat droppings in nature were dissected. The results of these examinations were negative.

Finely minced stomachs, intestines, and feces from infected bats were placed on pieces of moist toweling in Petri dishes. Maggots of *Musca domestica* were placed in these dishes and allowed to remain at room tempera-

ture for several days. Upon dissection of the maggots no helminths were encountered.

Bats are notoriously difficult animals to maintain in captivity at normal temperatures. At a temperature of 5-10° C., however, they will survive for several months; at this low temperature the animals are quite lethargic and do not feed. Water, however, must be supplied.

It was observed that caged *Myotis lucifugus* and *Eptesicus fuscus fuscus* maintained at room temperatures would occasionally feed on an injured bat. Furthermore, it was found that dissected bat carcasses were completely eviscerated in a short time when offered to caged bats at room temperature. This cannibalism was not observed by the writer in the natural habitats from which the bats used in this study were obtained. It may be significant, however, that in no colony of bats examined were any dead animals or remains thereof discovered.

While carnivory is the normal method of feeding employed by certain tropical bats, it is not usual among Vespertilionidae (which includes the genera *Myotis* and *Eptesicus*). Several writers (Allen, 1939; Ryberg, 1947), however, record instances of cannibalism in species of *Eptesicus* and *Myotis* maintained in cages or in otherwise unnatural surroundings. The former author suggests the possibility that large species of cave-dwelling bats may devour smaller species which attempt to inhabit the same cave, thus accounting for the occurrence of only one species of bats in certain small caves.

In view of this information it was decided to attempt to transfer *Longibucca eptesica* directly from infected to non-infected animals (vide supra). This was done either by (a) transferring the worms directly or (b) feeding the entire stomach containing worms.

(a) Worms obtained from the stomachs of freshly killed *M. lucifugus* were concentrated in a small drop of 0.85% NaCl solution and fed to uninfected animals by means of a capillary pipette introduced into the mouth. The results are as follows:

Animal #1. Fed 5 worms; dead 21 days later; 4 worms recovered from stomach.
#2. Fed 15 worms; killed 95 days later; no worms recovered.
#3. Fed 25 worms; dead 11 days later; 12 worms recovered from stomach.
#4. Fed 88 worms over a period of 3 days; killed 71 days later; 32 worms recovered from stomach.
#5. Fed 88 worms over a period of 5 days; killed 73 days later; 26 worms recovered from stomach.

(b) Three other bats were fed orally either 1 or 2 stomachs removed from infected *M. lucifugus*. The number of worms present in these stomachs was not known, but judging from the extent of infection in other bats from the group used, there were probably from 30 to 50 worms in each stomach. The results are as follows:

Animal #1. Fed 1 stomach; killed 42 days later; 1 worm recovered from stomach.
#2. Fed 1 stomach; killed 45 days later; 10 worms recovered from stomach.
#3 Fed 2 stomachs; killed 45 days later; 14 worms recovered from stomach.

The worms recovered were examined for motility and general appearance; all were quite active and none appeared moribund. A good number were females containing fully formed larvae; in most cases a few extremely young larvae were also found free in the stomach. Although in no instance was the number of worms recovered greater than the number administered, it appears that except in one case the worms did become established in their new hosts. Because of the unavailability of infection-free animals it is not possible at the present time to continue these preliminary studies.

Although the results of these experiments do not prove the transfer of *Longibucca eptesica* in nature through the carnivorous habits of its hosts, on the basis of the evidence at hand, this method is the one most strongly suggested.

SUMMARY

1. The morphology of the adult and developmental stages of *Longibucca eptesica n. sp.* found in the stomach of the bat is described and illustrated. The original diagnosis of the genus *Longibucca* is emended and certain features of the original description of *L. lasiura* are corrected. A key to the species of *Longibucca* is given.
2. The distribution and prevalence of infection with *L. eptesica* in three species of bats is discussed.
3. The only pathological manifestation in *Pipistrellus subflavus* infected with *L. eptesica* is a slight erosion of the surface layer of the gastric mucosa. Apparently *L. eptesica* is not a cause of mortality in bats.
4. Attempts to culture *L. eptesica* apart from its normal host were unsuccessful.
5. *L. eptesica* transferred directly from infected to uninfected bats survived for periods up to 73 days. The results of these preliminary experiments together with observations of cannibalism among captive bats suggest the possibility that *L. eptesica* may be transferred in nature through carnivorous habits of its hosts.

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The Feeding of Artificially Altered Oocysts of *Eimeria Tenella* as a means of Establishing Immunity to Cecal Coccidiosis in Chickens*

WILLIAM A. URICCHIO

The study of immunity to cecal coccidiosis caused by *Eimeria tenella* has received considerable attention within the last few years by a number of investigators. Johnson (1923) stated that fowls developed an age immunity to coccidiosis, but later (1924) decided that age is not the determining factor. The work of Beach and Corl (1925) suggested that resistance was the result of coccidial infection, but definite proof of this phenomenon remained for future investigators. Johnson (1927) reported that a high degree of protection could be established experimentally by the feeding of sporulated oocysts but that one feeding to the chick is not likely to confer a marked resistance. Tyzzer (1929) found that feeding a single dose of from 2 to 5 oocysts followed by a larger dose 15 days later conferred immunity. Farr (1943) found that relatively small numbers of oocysts administered daily produced a marked resistance to subsequent inoculations. This investigator demonstrated that 1,000 oocysts of *E. tenella* administered daily for 15 days, or 15,000 oocysts administered in three doses of approximately 1,000, 5,000, and 9,000 at 5 day intervals, were sufficient to produce a high degree of resistance.

Some previous attempts have been made to establish a method of immunization against coccidiosis through the use of altered oocysts. Jankiewicz and Scofield (1934), using heat-attenuated oocysts, presented data to show that immunity could be developed if 3 separate doses of oocysts were fed at 5 day intervals. Waxler (1941) reported that chicks which had been infected with X-ray-attenuated oocysts acquired a certain amount of resistance to a later infection.

Since many of the above methods appear impractical for immunizing fowls against coccidiosis on commercial farms, the following experiments were carried out to determine the possible value of the use of a single dose of artificially altered oocysts in establishing immunity against cecal coccidiosis.

MATERIALS AND METHODS

The Rhode Island Red birds used in these experiments were obtained as day-old chicks from the Poultry Section of the Bureau of Animal Industry, Beltsville, Maryland. The chicks, upon being removed from the incubator, were placed in clean electric brooders. When 15 days old, except where otherwise noted, they were removed from the brooders, weighed, banded, and sorted into groups of comparable average weight. Each group was placed in all-metal cages which were equipped with wire floors, and with waterers and feeders that were attached to the outside to avoid contamination of the feed and water by the droppings.

Infective oocysts used to inoculate the birds were obtained from the scrapings of the cecal walls of young birds which had been inoculated with small

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doses of sporulated oocysts 8 days previously. The scrapings of the ceca containing unsporulated oocysts were placed in a 2.5 per cent solution of potassium dichromate and allowed to remain at room temperature until the oocysts sporulated. The oocysts were then washed a number of times in tap water and stored in a refrigerator until needed. The number of sporulated oocysts in the inoculum was counted with the aid of a Fuchs-Rosenthal counting chamber. Three counts were made and averaged, and the number of oocysts per ml. was calculated. The suspension was standardized by dilution to give the desired number of oocysts per ml. The birds were inoculated by introducing the sporulated oocysts into the crop by means of a graduated pipette.

All the experiments were set up and carried out under essentially similar conditions. In each experiment there were two groups of birds, the experimental and a corresponding group of controls. When more than one experiment was started on the same day, the same control group was used. The birds of each experimental group received an initial dose of 100,000 treated oocysts, while the corresponding control group received an initial dose of 100,000 untreated oocysts. After 12 days all the surviving birds of both groups received a challenging lethal dose of 100,000 oocysts. The virulence of the oocysts of the challenging dose was checked by simultaneous feedings to susceptible birds. Observations on the weights, time of appearance of blood in the droppings, and the general behavior of the birds were made daily. Dead birds were removed daily and the ceca were examined for coccidia and coccidial lesions.

EXPERIMENTAL RESULTS

Five groups of experiments were conducted, treating the oocysts by means of ultrasonics, radium, freezing, heat at 45° C, and heat at 60° C. The amount of hemorrhage, weight gains, and mortality were used as criteria in judging the effectiveness of each of the treatments.

ULTRASONICS. Oocysts were treated with ultrasonic vibrations (high frequency sound waves) and fed to chicks in an attempt to build up immunity to coccidiosis. Wood and Loomis (1927) observed that blood cells were broken and that bacteria and protozoa were torn apart as a result of treatment with ultrasound. It was, therefore, desired to learn whether or not ultrasonics would have any effect in altering oocysts of *E. tenella*. The high frequency sound source was a piezoelectric quartz crystal driven at its fundamental natural vibration frequency of 5 megacycles by a radio oscillator. The crystal was mounted in a brass holder and placed in water which served as the medium for the sound waves. To focus the waves a concave watch glass with a radius of curvature of approximately 4 inches was mounted on top of a petri dish and sealed with wax. By manipulation of the watch glass under the quartz crystal, a fine spray of water at the surface indicated that the sound waves were focused.

The suspension of oocysts was placed in a glass tube one-fourth inch in diameter which was sealed at one end with a collodion membrane. It was found that one-half ml. suspension of oocysts was the most effective volume to use since a greater volume was not completely affected by the vibrations. The tube containing the oocysts was held by means of a clamp at the apex of the fountain produced by the ultrasound for the desired time of exposure.

Oocysts were subjected to vibrations for periods ranging from 2 to 20 minutes to ascertain the most effective time exposure. The highest mortality

from the initial dose was found in birds fed oocysts treated for 7 minutes or less. These birds all passed large amounts of blood and showed typical symptoms of coccidiosis 5 days after being inoculated. Birds receiving oocysts treated for 10 minutes had a lower mortality and also passed smaller amounts of blood. There was no blood passed and very few deaths in birds fed with oocysts treated for 15 and 20 minutes. From the initial infection there was evidence indicating that as the exposure time was increased, the severity of the disease was decreased. On the 12th day of the experiment all surviving birds of the groups that were fed oocysts treated for 10, 15, and 20 minutes were given a lethal challenging dose to learn whether or not they had developed immunity. On the 16th day all the birds that were fed oocysts treated for 15 and 20 minutes started to take on the droopy appearance of typical acute coccidiosis and also began passing large amounts of blood. By the 17th and 18th days a great percentage of these birds had died and upon autopsy proved to be heavily infected. The surviving birds in the groups that were fed oocysts treated for 10 minutes showed no clinical symptoms, and upon necropsy few coccidia were found and the ceca appeared normal. The loss from the initial dose in birds receiving oocysts treated for 10 minutes was too great for this method to be of practical value. The control groups infected with untreated oocysts suffered an average of 66 per cent loss from the initial dose and no losses from the challenging dose.

It appears that as yet no great claims can be made for ultrasonic-treated oocysts in establishing immunity to cecal coccidiosis. While ultrasonic vibrations do have some effect on the oocysts, the exact effect is not known, and a satisfactory exposure time can not be obtained where there will be only a very few deaths from the initial dose and no deaths from the challenging dose.

RADIUM EMANATIONS. In the radiation experiments a suspension of sporulated oocysts was subjected to the gamma rays of radium sulfate. Two platinum cells, each containing 1.0 mg. of radium, were used conjointly. These two cells emit gamma radiation equivalent to 2.1 mg. of radium in equilibrium with its disintegration products when corrected to allow for the absorption of 1.5 percent of gamma rays that takes place in the 0.2 mm. walls of the cells. The cells were inserted in a fine glass tube which was then placed in a 25 X 10 mm. vial filled with oocysts in solution. Exposure periods were for 24, 48, and 72 hours.

Three experiments were performed to determine the amount of resistance chicks developed from radium-treated oocysts. A high percentage of deaths occurred in all the experimental and control birds following the initial dose; however, there was less blood in the droppings, and upon postmortem examination the ceca of the controls were not as thick as those of the experimental groups. The results of the challenging dose to the surviving birds was quite the opposite of that following the initial dose. The birds that received a treated initial dose were able to withstand the challenging dose much better and passed only small amounts of blood, while the controls looked rather sick after 5 days and were passing larger amounts of blood. However, at necropsy most of the treated birds showed large cores in the ceca, indicative of severe coccidiosis.

The data obtained from the above experiments gives a good indication that oocysts treated by radium are not effective in the development of an adequate resistance in chickens.

HEAT AT 45° C. The oocysts in these experiments were treated by being subjected to a constant temperature of 45° C. A 15 ml. wide mouth vial was filled with a suspension of oocysts, corked, and placed in a heated chamber for the desired length of time. With this method it was necessary to perform a number of preliminary experiments before a favorable exposure time was obtained.

Oocysts treated for 0.5 to 10 hours produced a very high percentage of deaths when fed to chicks. Birds fed oocysts treated for 14 and 15 hours showed no symptoms and continued to gain weight until 3 days after the challenging dose. After this period the birds did not appear well. The first clinical sign occurred on the 4th day after the administration of the challenging dose when there was profuse hemorrhage in the majority of chicks. On the 5th day the birds began taking on a droopy look, and on the 6th and 7th days they died. The controls had a high death rate from the initial dose, but the surviving birds gained more weight after the challenging dose and no losses occurred. When the data of these experiments were compared it became clear that heat at 45° C. has little effect on oocysts for the first 10 hours and too much effect on oocysts for periods of more than 13 hours.

A group of experiments were conducted using oocysts treated for 11, 12, and 13 hours in an attempt to find an exposure period where no deaths would occur following the initial and challenging doses. The most favorable results were obtained by feeding oocysts treated for 12 hours to 12 day old birds. There was a 10 per cent mortality from the initial dose and no deaths from the challenging dose. Further trials were undertaken to test the above results and to learn what effect oocysts treated for 12 hours had on younger and older chicks. Two experiments were carried out using birds 10 and 15 days old. In the 10-day-old chicks, blood first appeared in the droppings on the 4th day and was very heavy during the 5th and 6th days. On the 7th day 40 per cent of the birds died and upon examination were found to contain blood-filled ceca. No deaths or hemorrhage occurred in the 15-day-old chicks. The controls of both experiments which were fed untreated oocysts showed an average mortality of 63 per cent. No deaths occurred amnog the surviving birds of the experimental and control groups after the challenging dose. Ten days following the challenging dose 15 birds of the 15-day-old group were autopsied and were found to have normal sized ceca with thickened walls and small cores. The 5 remaining birds were carried for 1 more week and then killed to see if the ceca had changed in appearance from the rest of the group. The walls of the ceca were still thickened, but the cores had passed out and the contents appeared normal.

These findings indicate that oocysts exposed to heat at 45° C. for 12 hours and fed to 15-day-old chicks may confer a reasonably high degree of immunity to a subsequent lethal dose.

HEAT AT 60° C. These experiments were performed in the same way as the preceding series except that the temperature was maintained at 60° C.

The feeding of the initial dose was followed by a high mortality in birds receiving oocysts treated for less than 16 hours and no mortality in birds getting oocysts treated for 18 hours or more. However, the results of the challenging dose showed a very high mortality in birds which were given an initial dose of oocysts treated for 18 and 20 hours and only 1 death in birds receiving oocysts treated for 16 hours or less.

As a result of the above preliminary experiments it was decided to try

oocysts treated for 15 hours. The results from the initial dose were very promising, and it seemed that the correct exposure time had been obtained. However, the results of the challenging dose were not favorable. Four days after the challenging dose the experimental birds began passing small amounts of blood, and this was increased on the 5th and 6th days. The general behavior of the birds was that of a heavily infected group, with weight gains in only 2 birds. The first death occurred on the 7th day, followed by 2 more deaths on the 8th day. The surviving experimental birds improved considerably and became very active on the 9th day after the challenging dose. On the 12th day all the birds were autopsied and were found to have either normal ceca or ceca with small cores or large amounts of blood present. The controls in this group of experiments suffered a great loss from the initial dose and no loss from the challenging dose.

In an effort to obtain a lower percentage of deaths, an experiment was performed using oocysts treated for 14 hours, since 13 hours proved to be too short a time and 15 hours too long. The experimental birds were not clinically affected from the initial dose, whereas the control birds became very weak and passed considerable blood. Five control birds died on the 7th day and 3 on the 8th day. Twelve days following the initial dose all the surviving birds of both groups were given a lethal challenging dose. The experimental birds became weak and passed large amounts of blood. Beginning on the 5th day the chicks became pale and inactive and on the 6th day 4 died followed by 3 more deaths on the 7th day. Each chick that died had blood-filled ceca. The surviving members of the control group were not visibly affected by the challenging dose.

These results indicate that 0.5 to 20 hours exposure of oocysts to 60° C. was not effective in establishing immunity to subsequent lethal doses.

FREEZING. The oocysts in this group of experiments were treated by being placed in the freezing compartment of a refrigerator which was maintained at —5° C. A 15 ml. wide mouth vial was filled with a suspension of oocysts, corked, and placed in the compartment for the desired length of time.

Chicks receiving oocysts treated for 4 days or less showed a high mortality rate from the initial dose, while the chicks receiving oocysts treated for periods of 7 days or more had no deaths from the initial dose, but a high death rate following the challenging dose. The more favorable results from the preliminary experiments were obtained by feeding 12-day-old chicks oocysts that were treated for 5 days.

To learn whether oocysts exposed for a shorter period of time would be more effective on older chicks, oocysts that had been treated for 4.5 days were fed to 15-day-old chicks. There was a 50 per cent mortality in the experimental group and 80 per cent mortality in the control birds following the feeding of the initial dose. None of the surviving birds showed any outward signs of coccidiosis following the challenging dose.

Since 4.5 days exposure was not effective, another test was conducted using oocysts treated for 5 days and chicks 15 days old in an effort to obtain a lower percentage of deaths. Very small amounts of blood appeared in the droppings of the experimental group following the initial dose and the birds remained alert and active, showing no symptoms of the disease. The control group passed large amounts of blood and showed a mortality of 53 per cent. The challenging lethal dose was given to all the surviving birds of both groups

on the 12th day to learn whether or not any immunity had been developed. Another set of control birds of the same age were also inoculated to test the lethal potency of the challenging dose. The chicks of the experimental group and the surviving control birds proved to be well protected against the lethal dose, as no deaths resulted and there was a complete absence of any symptoms of the disease. Upon autopsy of the birds of the treated group the ceca and its contents appeared normal except for the thickening of the cecal wall in two birds. The ceca of the controls also appeared normal upon autopsy. The immunity which developed from the initial dose was the same in both the experimental and control groups. However, the control group suffered a 53 per cent mortality from the initial infection, whereas, no birds of the experimental group died. Six of the 8 control birds carried to test the lethal potency of the challenging dose died on the 7th day and were found to have a severe case of coccidiosis.

These results indicate that oocysts exposed to —5° C for 5 days may be effective in producing an immunity to a subsequent lethal dose.

CONCLUSIONS

The data obtained from the foregoing experiments indicate that chicks fed oocysts altered by means of ultrasonics, radium, and heat at 60° C developed very little resistance to subsequent lethal doses, while chicks fed oocysts treated by means of freezing and heat at 45° C appeared to have acquired an immunity to a second lethal infection.

No deaths and very little hemorrhage occurred in15-day-old birds fed an initial dose of oocysts treated by freezing for 5 days. Experimental birds lacked the droopiness, inactivity, and weakness of the control group. Following the challenging dose there were no deaths, hemorrhage, or change in appearance of the birds in the experimental group. The surviving control birds also suffered no deaths from the challenging dose. There was no noticeable difference in the weight gains of the experimental birds and the uninoculated controls. It may be concluded that oocysts treated by freezing for 5 days can be administered safely, since there were no deaths from either the initial or challenging doses.

A seemingly high degree of immunity was also established in 15-day-old birds fed oocysts altered by means of heat at 45° C for 12 hours. There were no losses from the initial and challenging doses and the weight gains were equal to those of the uninoculated controls. The birds in this experiment passed more blood following the initial infection than did those in the freezing experiment, and were also found to contain more irritated ceca upon examination after the challenging dose than did those of the freezing experiment.

SUMMARY

1. Fifty-two sets of experiments were undertaken to determine the efficacy of single doses of artificially altered oocysts of *Eimeria tenella* in establishing immunity against cecal coccidiosis in chickens.

2. No appreciable resistance was developed in birds fed oocysts altered by means of ultrasonics, radium, or heat at 60° C.

3. Fifteen-day-old chicks fed 100,000 oocysts treated by heat at 45° C for 12 hours appeared to have a marked immunity to a challenging lethal dose without fatalities. A more effective degree of immunity was produced in the same age birds fed 100,000 oocysts exposed to —5° C for 5 days.

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Meloidogyne brevicauda, n.sp. a cause of root-knot of mature tea in Ceylon.

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The root-knot eelworm which was formerly regarded as a single species, *Heterodera marioni* (Cornu) Goodey, has been transferred by Chitwood (1949) to the genus *Meloidogyne* Goeldi, 1887, and embraces a number of species. It has long been known as a destructive pest of young seedlings under six months old in Ceylon tea plantations. The same nematode causes severe galling of roots of *Tephrosia vogelii* and *Erythrina lithosperma,* two green manures commonly interplanted with tea.

The interplanting of tea with susceptible green manure plants has resulted, in many areas, in a marked increase in the root-knot population of the soil. Nevertheless mature tea bushes growing in such heavily infested soils usually show no ill-effects. The root-knot larvae find no difficulty in entering young rootlets of the mature tea bushes but are apparently unable to complete the moult between the second (infective) and the third (sedentary) stages.

As Barrons (1939) and Christie (1949) have suggested, resistance to root-knot damage lies in the failure of the larvae to survive after entering and not in their inability to penetrate the roots. This fact was shown by Gadd & Loos (1941) with *Crotalaria anagyroides* which is apparently resistant at all stages of growth to root-knot though its roots are highly attractive to the larvae. Tea appears to be unusual in that it is very susceptible in the seedling stage but develops a form of resistance with increasing age until an apparent complete resistance is acquired.

The presence on a few Ceylon tea estates of relatively small areas in which mature bushes have severely galled roots needed explanation. Gadd & Loos (1941) presumed that a specialized race of the root-knot nematode, capable of overcoming the adult bush's resistance had been evolved. Specimens of galled roots were sent to the Imperial Bureau of Agricultural Parasitology, St. Albans, England in 1938 and a tentative identification as *Heterodera marioni* was given. This identification was based entirely on females as males, eggs and second-stage infective larvae could not be found. The failure to find eggs, males and larvae, although hundreds of females have been observed, has been my experience over a period of several years at periodic examinations of infested material.

During a visit to Ceylon in January 1952, Mr. Gerald Thorne of the United States Department of Agriculture made a short study of infested material in which he found males and larvae. He then expressed the view that the eelworm concerned was probably an unrecorded species of *Meloidogyne*. The present study was undertaken to confirm or refute that view and, as I shall show later, his opinion has proved correct. Mr. Thorne's help, advice and criticism both in this instance and on many past occasions are gratefully acknowledged.

Tea plants of all ages, from seedlings to old bushes, are equally susceptible to infestation and gall formation by *Meloidogyne brevicauda*. The presence of empty cavities, where females have lodged, in galls on old mature roots indicates that infestation began when the plants were quite young. The effects of infestation on growth of the plant are most noticeable during the recovery period after pruning which usually takes place at 2-6 year intervals. Usually about 15 weeks elapses after pruning before a normal healthy bush is again pluckable whereas severely infested bushes may take 12-15 months to reach the plucking stage. The production of new branches becomes restricted and in consequence the cropping capacity of the bush is seriously reduced. Very severely infested plants fail almost completely to make new growth and become "passengers"—i.e. unproductive of crop though rarely are they killed outright.

DESCRIPTION

Order: Tylenchida Thorne, 1949. Sub family Heteroderinae Filipjev, 1934. *Meloidogyne brevicauda,* n. sp.

MALE: Length = 0.97-1.44 mm.; width = 28-52μ.; a = 26-44; b (length of body/length to base of median bulb) = 12.6-17.

The body is worm-like in shape, tapering anteriorly to a conoid neck (Fig. 1-A and G) and posteriorly to a bluntly sub-digitate tail terminus (Fig. 1-H). When killed by heat the sub-digitate portion of the tail usually turns upwards. The head, which is often difficult to obtain in a lateral position, is scarcely set off from the neck. The cuticle carries coarse transverse striations throughout the body except in the region of the head and the sub-digitate tail terminus. The striations, which occasionally do not completely circle the body, are about 1.7μ apart at the region of the spear and up to 3.2μ at the middle of the body. The hemizonid (J. B. Goodey 1951) is located about 3-4 annules in front of the excretory pore which is nearly two-thirds the distance down the oesophageal gland region. The lateral field starts as an inverted V about half-way down the spear. The two incisures, or arms of the

V, gradually diverge and between them in the region immediately above the median bulb a third central incisure commences. This third incisure, in the region opposite the hemizonid, itself divides into two, thus forming four incisures running almost parallel. The four incisures divide the lateral field into 3 strips which continue throughout the body length until they meet the opposite lateral field at the sub-digitate tail terminus (Fig. 1-H). The transverse striae of the body continue across the lateral field though occasionally a stria may not continue between incisures (Fig. 1-D). A phasmid is located on the lateral field immediately below the level of the gubernaculum (Fig. 1-H). Deirids were not observed.

The two lateral lips are considerably larger than the subventral and subdorsal lips; their basal attachment to the head takes up nearly three-fourths of the head area (Fig. 1-E). They are wide at the base but taper towards the

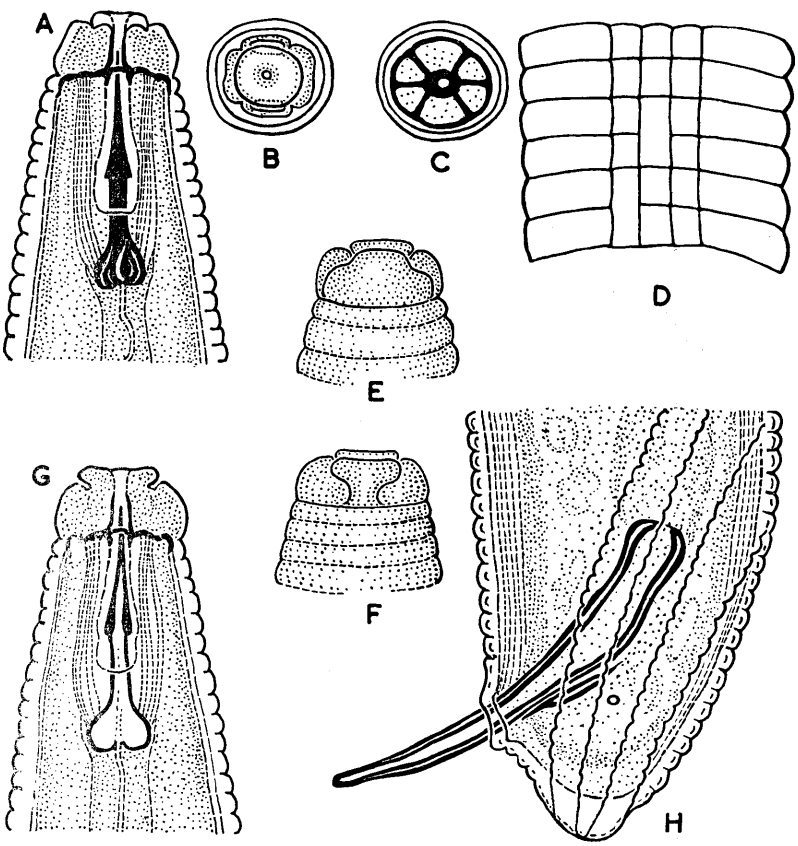

Fig. 1.—Male *Meloidogyne brevicauda*. A—Head, X 1400; B—Face view showing labial cap, amphids and lip formation, X 1450; C—Transverse section through cephalic basal plate, X 1450; D—Lateral field, X 1400; E—Head, lateral view showing wide lateral lip, X 1400; F—Head, dorsal view showing fused pair of lips; G—Head, dorsal view, X 1400; H—Tail, X 1400.

apex (Fig. 1-E). In this species the two subventral and also the two subdorsal lips are fused, narrow at the base and broad at the apex (Fig. 1-F). Crescent-shaped amphid apertures are located at the junction of the labial cap and lateral lips (Fig. 1-B). In a dorso-ventral view the labial cap is seen as a saucer-shaped structure with a deep indentation where the amphid openings meet the lateral lips (Fig. 1-G). The shape and position of the labial cap in relation to the lips and amphids are best observed in a face view using a combination of vertical illumination (Sartory 1948) and transmitted light. The cap is almost circular in shape, nearly four-fifths as wide as the lip region and, due to its saucer-like shape, slightly overlaps the amphidial apertures (Fig. 1-B). There are two ridges or plications which appear as crescent-shaped structures on the dorsal and ventral sides of the labial cap. (Fig. 1-B). By transmitted light alone this structure as well as the position of the labial cap relative to the amphids cannot be accurately observed.

The position and structure of the basal sclerotised framework of the head is shown in in Fig. 1-A, C and G. This structure appears in a face view as hexaradially thickened arms emanating from the heavily sclerotised lining of the vestibule (Fig. 1-C). In lateral or dorso-median views the heavily sclerotised structure of the vestibule is a striking feature (Fig. 1-A and G). A less heavily sclerotised continuation of the basal framework extends two to three annules into the anterior part of the pharynx to form a guiding ring for the spear (Fig. 1-A and G).

The massive finely pointed spear is 19.5-20.7μ long with basal swellings 5.2μ wide. The anterior tapering portion is about as long as the posterior knobbed region and is more heavily cuticularised (Fig. 1-A and G). The median bulb with its valvular apparatus is 14-17.5μ long by 8-10μ wide. The oesophageal gland region is crossed, at its narrow portion, by the nerve ring slightly posterior to the median bulb. The intestine with its highly granular contents overlaps the oesophageal gland for almost its entire length and makes observation of the oesophageal gland nuclei difficult.

The spicules are slightly arcuate, 34-42.5μ long, each with a bluntly rounded terminus (Fig. 1-H). The gubernaculum, which is 10-10.5μ long, tapers anteriorly (Fig. 1-H). The testis is 333-511μ long.

FEMALE: Length = 0.68-1.86 mm.; width = 0.31-1.06 mm.

The eelworms show fairly wide variation in shape and size, the majority being well over 1 mm. in length. Variation in shape is determined by the type of root in which the worm develops. In soft-tissued feeding roots they are pearly white, globular to pear-shaped with short necks. The convoluted ovaries are well developed with many eggs. In roots which have developed rapidly to form corky cortical tissues and a hard central woody cylinder the females have broadly cylindrical bodies with long narrow necks encased in tubular cavities within the woody tissues. The linings of these tubular cavities are smooth and slimy. The cylindrical body is surrounded by cortical tissues only. It appears probable that pressure exerted by the harder plant tissues prevents the warm from developing its normal shape and restricts it to the more elongate cylindrical form. The ovaries also do not develop normally; they remain as narrow tubes without convolutions and extend up to and sometimes into the neck. Eggs have not been observed. In all other respects these females appear healthy and are apparently able to obtain the necessary nourishment for existence. Both forms of females are soft-bodied and easily ruptured on teasing out from the plant tissues.

Egg masses are few and difficult to find. When galled roots are kept for 3 or 4 days in a damp chamber large numbers of gelatinous matrices may be seen protruding outside the root and connected with worms but are usually empty of eggs. The largest number of eggs obtained from a single female was 45, a number far less than is usual with other *Meloidogyne* species. Soil washings, too, seldom give large numbers of larvae.

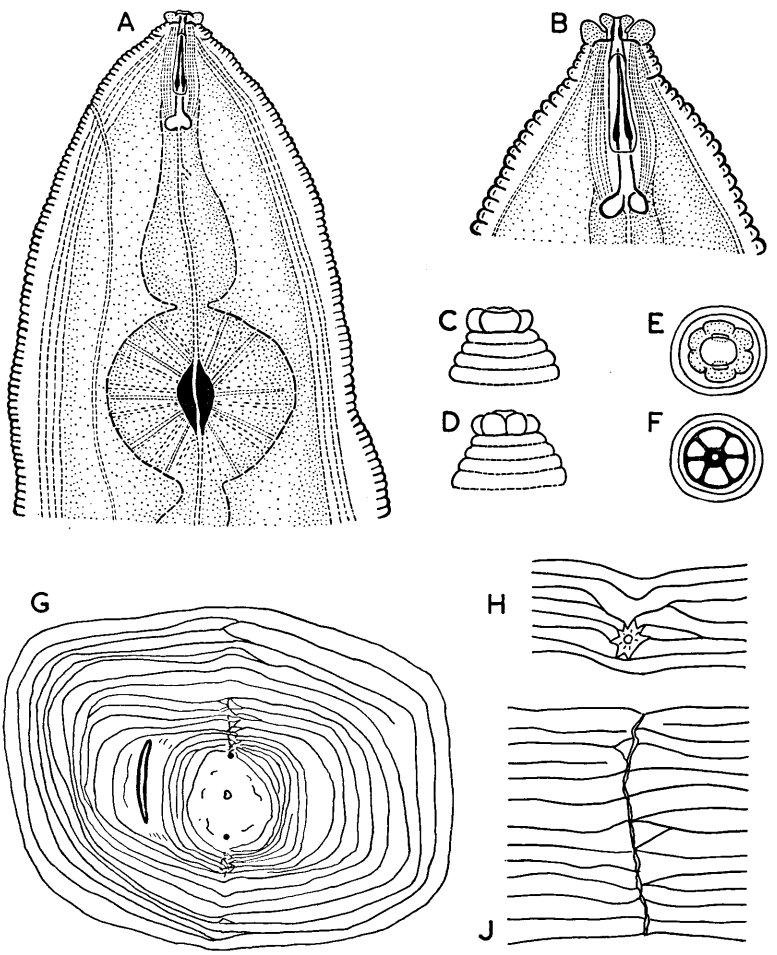

Fig. 2.—Female *Meloidogyne brevicauda*. A—Anterior portion of body X 510; B—Head, dorsal view, X 1120; C—Cip region, lateral view, X 1120; D—Lip region, dorsal view, X 1120; E—Face view showing labial cap and amphids, X 1160; F—Transverse section though cephalic basal plate, X 1160; G—Perineal region, X 1160; H—Ventral view of excretory pore region X 1160; J—Lateral line and transverse striae, neck region, X 1160.

In the position assumed on killing by heat the head usually is turned at right angles to the neck. The cuticle and the neck is coarsely transversely striated but the striations become less evident on the swollen body except in the perineal region. The excretory pore is well defined and its duct may be

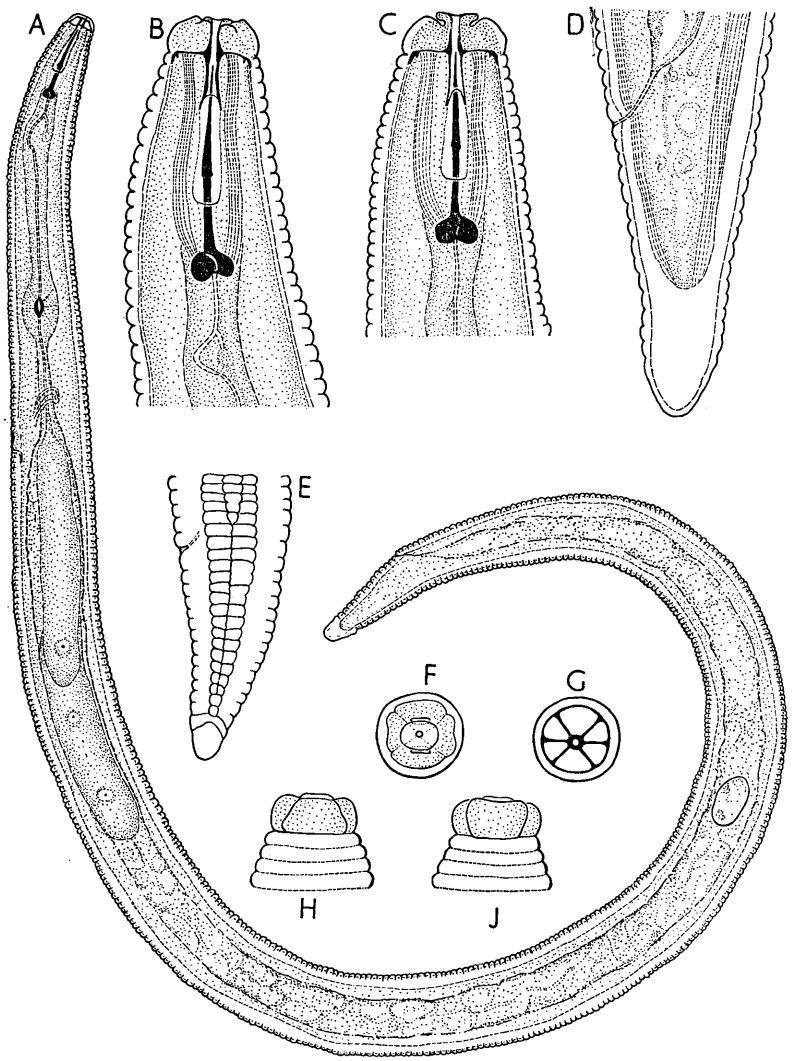

Fig. 3.—Second stage larvae *Meloidogyne brevicauda*. A—Lateral view, X 630; B—Head, X 1580; C—Head, dorsal view, X 1580; D—Tail, X 1580; E—Tail showing lateral field, X 1230; F—Face view showing labial cap, amphids and lips, X 1580; G—Transverse section through cephalic basal plate, X 1580; H—Lip region, lateral view, X 1580; J—Lip region, dorsal view; showing fused pair of lips, X 1580.

followed for a considerable distance down the neck (Fig. 2-A). Allen (1952) has pointed out the extreme forward position of the excretory pore in females of *Meloidogyne* species and has included this characteristic in the generic diagnosis. In this species the position varies somewhat between individuals but it general location is in the region of the spear, 20-28 annules from the head. In all cases observed the pore is located in front of the median bulb (Fig. 2-A). In ventral view the opening of the excretory pore lies in a pit or depression into which 3 or 4 striae converge (Fig. 2-H). The single lateral incisure is clearly seen on the neck where the transverse striae cross or converge to it (Fig. 2-J). Occasionally a transverse stria may not completely encircle the neck but may stop abruptly at any position. In the perineal region the striae form a pattern typical of *Meloidogyne* (Fig. 2-G). The pattern is made up of transverse striae which are interrupted, close to the perineal region, by the lateral field. A few of the striae circle the anal region where they are more closely spaced than around the vulva (Fig. 2-G). Phasmids are present (Fig. 2-G).

The female head in face view shows the hexaradial circumoral framework similar to that of the male though less heavily sclerotised (Fig. 2-F). The lateral lips (Fig. 2-C) are only slightly larger than the sub-ventral or the sub-dorsal ones (Fig. 2-D) which are not fused as they are in the male. The labial cap is about half the size of the lip region and the amphid openings are located in a position similar to that of the male (Fig. 2-E).

The spear is 22.1μ long with basal knobs 4-5μ wide. Due to the generally up-turned position of the head and anterior part of the neck the spear is seldom seen in a straight position. The median bulb is abnormally large and well developed (Fig. 2-A).

EGGS. The eggs are 108.5-133μ long and 41.7-59.5μ wide. The first larval moult takes place in the egg.

LARVAE. Length = 0.46-0.59 mm.; width = 14.7-20.6μ. Tail = 17.5-28μ. a = 23-33; *b = 6.2-7.3; c = 21-29.

The wormshaped body tapers anteriorly to a conoid neck and posteriorly to a conoid bluntly rounded tail (Fig. 3-A). The tail, shortest as yet described in *Meloidogyne* species, is about one and one-fifth times as long as the greatest body width and slightly more than twice the anal body width (Fig. 3-D). A conspicuous hemizonid is located immediately in front of the excretory pore, about 87-98μ from the head. (Fig. 3-A). Chitwood (1949) draws what is apparently the hemizonid structure at a similar position in many of his drawings of larvae.

The cuticle is fairly coarsely annulated with distinct striae 1.7μ apart. Annules do not occur on the head. The lateral field, as in the male, is mainly made up of four distinct incisures which are crossed by the transverse striae. The incisures commence, as in the male, half way down the spear. The third central incisure commences about one spear length behind the spear and divides into two incisures almost opposite the hemizonid. At about anal level the two field limiting incisures converge to end in a rounded terminus just before the end of the tail while the two central incisures meet and continue, as a single incisure, ending abruptly two annules before the end of the lateral field (Fig. 3-E). Deirids or phasmids were not observed.

The hexaradial circumoral sclerotised framework of the head and the

*Total body length/length from anterior end to base of median bulb.

sclerotised structure surrounding the entrance to the pharynx are similar to that in the male (Fig. 3-B, C, and G). The lateral lips are slightly smaller than the fused pairs of subventral and subdorsal lips (Fig. 3-F, H, & J). The labial cap is smaller, in relation to lip size, than that of the male being only about half as wide as the lip region as in the female (Fig. 3-F). The position of the amphid, at the juncture between the labial cap and the lateral lip is similar to that described for the male (Fig. 3-F).

The rather slender but well defined spear is 14.3-14.5μ long with basal knobs about 2.7μ wide. The anterior more heavily cuticularised tapering portion is slightly longer than the posterior parallel-sided but knobbed region (Fig. 3-B and C). The ovoid median bulb with its valvular appartus is about 15μ long by 10μ wide. The nerve ring encircles the esophageal isthmus between the median bulb and the hemizonid. The intestine with its granular contents overlaps the oesophageal gland region for almost its entire length making accurate determination of the oesophageal gland neclei difficult. The ovoid genital primordium is located about two-thirds down the body (Fig. 3-A).

DIAGNOSIS

Chitwood (1949) revived the generic name *Meloidogyne* Goeldi, 1887, erecting four combinations, one variety and a new species in place of *Heterodera marioni* (Cornu, 1879) Goodey, 1932, and later (1952) described a subspecies. He based the identification of species on the pattern of the cuticular annulations in the female perineal region. Allen (1952) showed that variations occur in the perineal pattern of females originating from a single egg mass and suggested that variation within a species was greater than previously indicated.

All previously described species within the genus have larvae difficult to separate on morphological characters—length of stylet, length of tail in relation to total body length and shape of tail are almost identical. Egg sizes too show no marked variation between species. On larvae alone the 5 species described by Chitwood (1949) are to all intents and purposes the same species morphologically. The most striking features which at once separate the new species from all other previously described species of *Meloidogyne* are the coarse annulations and the short rather blunt tail of the larvae together with the readily distinguishable lateral fields, and the unusually characteristic lip forms of larvae, males and females. Further detailed points of difference are set out below.

EGG. Larger than any so far described (108-133x41.7-59.5μ).

LARVAE. Shape of tail bluntly rounded, short (c = 21-29). Other described species have longer pointed tails (c = 5.8-13). Stylet 14.3-14.5μ compared with 10μ in other species.

FEMALE. Stylet 22.1μ compared with 14-16μ (*M. arenaria*), 16-17μ (*M. javanica*), VE-VFμ (*M. incognita*), 16μ (*M. incognita* var. *acrita*) and 12-14μ (*M. hapla*). The range in body size (0.68-1.86 mm.) is larger than any of the previously described species.

MALE. Sub-digitate tail terminus as opposed to the more conoid tails of other species.

TYPE LOCALITY. From roots of tea (*Camellia sinensis*) (L) O. Kuntze. Nuwara Eliya, Ceylon. (Elevation approximately 6500 ft.)

Acknowledgments. Preparation of face views of the male and larva were made by Dr. Basil Goodey. I wish to tender my thanks to him and to Dr. T.

Goodey and Dr. Mary Franklin, all of the Rothamsted Experimental Station, for helpful criticism and encouragement during the preparation of this paper. This work was conducted at the Nematology Department of the Rothamsted Experimental Station on a fellowship from the Technical Co-operation Scheme, Colombo Plan.

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Experimental Cross Transmission of *Strongyloides Papillosus* in Ruminants

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The intestinal threadworm, *Strongyloides papillosus,* a potential pathogen of young calves (Vegors and Porter, 1950, Jour. Parasitol. 36 (6-Sec 2):33) is parasitic in sheep and goats as well as cattle. Records of natural and experimental cross transmission, however, are few in number. Roberts (1942, Austral. Vet. Jour. 18:19-27) observed that three-months-old calves grazing on a sheep pasture became heavily infected with Strongyloides. The parasites were rapidly eliminated and the animals subsequently remained "free" despite attempts to reinfect them experimentally. Woodhouse (1948, Jour. Am. Vet. Med. Assoc. 113:354-356) described rather severe symptoms in a steer following cutaneous exposure to *S. papillosus* of ovine origin, although eggs of the parasite were never recovered from the feces.

In the experiments described here, parasite-free, grade Southdown lambs, grade Saanen kids, and a grade Jersey calf, were exposed to infective larvae of *S. papillosus,* cultured from feces of naturally infected calves or lambs. The larvae were cultured to the infective, filariform stage in a moist mixture of feces and animal charcoal. All animals were exposed to infection by placing larvae, their number having been determined by dilution count, on areas of skin that were clipped free of hair or wool.

The experimental data are compiled in Table 1. On the basis of egg counts, Lamb 1, exposed to 1,300 larvae of ovine origin, and Kid 1, exposed

TABLE 1. Data on Experimental Infections of Ruminants with *Strongyloides papillosus* from Cattle and from Sheep

Host		Age (days)	Larvae Given (number)	(source)	Interval Between Exposure and Appearance of Eggs in the Feces (days)	Maximum e.p.g. Observed (number)	(day after exposure)
Lamb	1	30	1,300	Sheep	10	21,000	23
	R	57	10,000	Sheep		92,600	17
Lamb	2	30	1,300	Cattle	15	54	18
	R	57	10,000	Sheep		57,400	14
Calf	1	19	1,300	Sheep	13	18	16
	R	46	10,000	Cattle		1,400	70
Kid	1	18	1,300	Cattle	11	10,600	15
	R	45	10,000	Sheep		4,200	17
Kid	2	45	10,000	Sheep	9	6,200	17

R—Signifies reexposure.

to a like number of larvae of bovine origin, both developed fairly large infections, with prepatent periods of 10 and 11 days, respectively. However, Calf 1, exposed to larvae from sheep, and Lamb 2, exposed at the same time to larvae from cattle, developed low grade infections, with somewhat longer prepatent periods of 13 and 15 days, respectively.

Twenty-seven days after the first exposures, both lambs, Kid I, and a previously uninfected kid (Kid 2) were all exposed to 10,000 larvae of ovine origin. Calf 1 was given the same number of larvae of bovine origin at the same time. On the day of reexposure, the counts of eggs per gram of feces (e.p.g.) were as follows: Lamb 1, 1,200; Lamb 2, 16; Calf 1, 4; Kid 1, 2,600. The number of eggs from Kid 1 dropped to 4 e.p.g. a week after reexposure. All previously parasitized animals eventually showed moderate to large increases in egg counts, indicative of successful superinfection (Table 1). Kid 2 developed a fairly high infection from the applications of larvae of sheep origin, with a prepatent period of 9 days. The fact that higher peaks were reached earlier in lambs reexposed to ovine Strongyloides would indicate that their infections were more successful than that obtained in the calf reexposed to bovine threadworms.

When the final observations were made 38 days after the last exposure, Lamb 2 was passing 11,600 e.p.g.; Kid 1, 3,200, and Kid 2, 800. Calf 1 and Lamb 1 were observed for 99 days after their last exposure. At the end of this time, the calf was passing 88 e.p.g. and the lamb, 3,200. During the infections, none of the animals evidenced any ill effects ascribable to parasitism.

SUMMARY

Although cross-transmission of *S. papillosus* from one domestic ruminant to another was possible under the conditions of these experiments, it appears that calves and lambs may not be as susceptible to Strongyloides from the opposite species as to those from their own.

On the other hand, goats were readily infected with larvae from either sheep or cattle.

On The Occurrence of Tapeworms, *Moniezia expansa* and *Moniezia benedeni*, in Cattle and Sheep

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The tapeworms *Móniezia expansa* and *M. benedeni* have been reported from both cattle and sheep. Stoll (1937, Proc. Helm. Soc. Wash., 4:32) and Roberts (1942, Austral. Vet. Jour., 18:19-27) have reported on calves acquiring *M. expansa* as a result of grazing pastures contaminated by sheep. The latter was of the opinion that *M. expansa* was not well adapted to cattle and that *M. benedeni* occurred only in sheep that grazed on cattle pastures. The writer (in press) observed infections with *M. expansa* in calves grazing pastures contaminated by sheep, but observed no cases of transfer of *M. benedeni* from calves to lambs.

Since 1946, the writer has identified 669 tapeworms from sheep and cattle. Superficially the two species are practically indistinguishable but are easily differentiated by microscopical examination of the interproglottid glands (Cameron, T.W.M., 1934, Internal Parasites of Domestic Animals, London, 292 pp. illus.).

In examining tapeworms, it was found that pressed preparations of fresh specimens were satisfactory; however, it was often necessary to stain and clear specimens that had been preserved in alcohol or formalin for several months. Approximately 300 additional specimens were not identified to species because of their immaturity or because of technical difficulties.

TABLE 1.—The occurrence of *Moniezia expansa* and *M. benedeni* in cattle and sheep from various localities.

Host	Number	*M. expansa*	*M. benedeni*	Total	Locality	Collector
Cattle	87	59	349	408	Alabama-Georgia	D. A. Porter
	1		1	1	Colorado	R. S. Jackson
	3		8	8	Florida	L. E. Swanson
	2	8	6	14	Illinois	N. D. Levine
	1		2	2	Louisiana	D. A. Porter
	14		68	68	N. Carolina	C. D. Grinnels
	2		3	3	Ohio	F. R. Koutz
	9		26	26	Texas	G. E. Cauthen
						R. D. Turk
Totals	119	67	463	530		
Sheep	9	85		85	Alabama	D. A. Porter
	1		1	1	Colorado	R. S. Jackson
	2	7		7	Florida	L. E. Swanson
	1	2		2	Idaho	L. E. Swanson
	2	5	4	9	Illinois	N. D. Levine
	4	17		17	Kentucky	A. C. Todd
	1	4		4	N. Dakota	D. F. Eveleth
	3	6	1	7	Ohio	F. R. Koutz
	2	7		7	Texas	R. D. Turk
Totals	25	133	6	139		

The numbers of each species identified from cattle and sheep, as well as the locality and collector, are shown in Table 1. Of the 67 specimens of *M. expansa* from cattle, 41 were from three calves which followed sheep on pasture; two of these calves harbored both species. Seventeen specimens of *M. expansa* were found together with *M. benedeni* in five out of eight calves from one dairy farm where there were no sheep. Histories of the other cases of *M. expansa* in cattle and of *M. benedeni* in sheep are not known. Link *et al.* (1950, Jour. Am. Vet. Med. Asso., 117:52-53) have reported parasitism of calves with *M. expansa* on a farm where sheep had never been kept, and Stoll (1938, Jour. Parasitol., 24:527-545) observed both species occurring in a flock of sheep separated from cattle.

From the data presented, it appears that *M. benedeni* is the common species of cattle while *M. expansa* is the common species of sheep, but that either host may be parasitized by either species.

Pathogenicity of some plant-parasitic nematodes from Florida soils. III. Growth of Chinese waterchestnut, *Eleocharis dulcis* (Burm. f.) Henschel inoculated with *Dolichodorus heterocephalus* Cobb (Tylenchinae)

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Chinese waterchestnuts (also known as matai) are grown in several localities in the southern and Pacific states for their edible corms. Despite the comparatively recent introduction of this plant as a cash crop in the United States, reports of a new disease affecting them have already been received. One grower who went into intensive but small-scale cultivation reported that sections of his planting were unhealthy and that undersized corms were produced. Among the soil micro-flora and -fauna associated with corms from diseased plants, specimens of awl nematodes, *Dolichodorus heterocephalus* Cobb, were isolated. These nematodes have also been found associated with a disease of celery in Florida known as "red root" (Taylor, 1943), and have been shown to cause a serious decline of celery seedlings in greenhouse experiments (Tarjan et. al., 1952). This report concerns a greenhouse test in which waterchestnuts were grown after being inoculated with awl nematodes.

MATERIALS AND METHODS**

Two units of 12 replicate gallon crocks, each containing three plants growing in a water-saturated sandy loam, were inoculated with nematode suspensions containing 92 per cent *D. heterocephalus,* 2 per cent *Hoplolaimus coronatus* Cobb, and 6 percent saprophytic forms of the genera *Eucephalobus, Rhabditis,* and *Acrobeles.* The volume of stirred nematode suspension pipetted was regulated so that each crock in the first lot received approximately 750 nematodes of the species *Dolichodorus heterocephalus* while each crock in the second lot received 1500 nematodes of the same species. A third lot of 12 replicate crocks was left uninoculated and used as control. All crocks were completely randomized and watered immediately after inoculation.

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**The author appreciates the help rendered by Drs. J. R. Christie and B. F. Lownsbery. The former supplied the propagative material as well as the inoculum, while the latter assisted in setting up this experiment.

After 3½ months at an average soil temperature of 24° C. data were obtained on total weight of plants, weight of roots, and number and weight of corms produced. Nematodes were recovered from the soil in which each plant had been growing by means of the Cobb (1918) screening-gravity method. Nematodes retained by 60- and 150-mesh screens were placed in water suspension and numbers in aliquots pipetted from the stirred suspensions were counted. The total recovered from each pot was computed.

RESULTS

Readily apparent differences between uninoculated controls and inoculated plants were in evidence only in comparisons of root systems. Whereas uninoculated plants had a number of whitish, fleshy feeding roots and larger root systems, inoculated plants had root systems containing discolored, sickly roots which were somewhat reduced. As indicated in Table I, total plant weight and root weight of uninoculated controls were significantly higher than corresponding weights of inoculated plants. There seemed to be little differences on subsequent growth of plants whether they were inoculated with 750 or 1500 nematodes. There were no significant differences between treatments, in number and weight of corms produced.

TABLE I.—Effect of awl nematodes on growth of Chinese waterchestnuts 3½ months after inoculation.

Treatment	Total plant weight in grams	Root weight in grams
750 nemas	121.8*	94.7
1500 nemas	121.5	96.4
Controls	184.0	154.7
LSD .05	20.2	20.6
LSD .01	27.0	27.6

*Each value represents the mean of 12 replicate crocks, each containing three plants.

Inspection of living corms and those stained by the lactophenol-acid fuchsin method (McBeth, et al., 1941) revealed the presence of *D. heterocephalus* around young shoots arising from these corms. In a few cases, all stages of *Hoplolaimus coronatus* were found within the cortex of secondary roots, apparently parasitizing these roots.

Counts of nematodes recovered from the soil in which plants had grown indicated that awl nematodes reproduced in greater numbers on plants receiving 750 nematodes than on plants receiving 1500 nematodes. Whereas for the former an average of 1758 nematodes per crock was obtained, representing a population increase of 134 per cent, an average of 1300 nematodes per crock was obtained for the latter, representing a population decrease of 13 percent. It is believed that plants receiving an inoculum of 1500 nematodes were unable to support this number and many of the nematodes perished. *Hoplolaimus coronatus,* the only other plant-parasitic nematode species present, was found in only a few crocks in numbers too small to be of significance.

CONCLUSIONS AND SUMMARY

An association of awl nematodes, *Dolichodorus heterocephalus,* with growing Chinese waterchestnuts has been shown to be detrimental to the host

plants. These nematodes were observed around shoots arising from corms but were not seen within or feeding on root tissues. Awl nematodes are believed to be ectoparasitic in feeding habit and are probably dislodged from their feeding sites once roots are disturbed.

Whereas symptoms of decline exhibited by some waterchestnut plantings may not be entirely due to the feeding of these parasites, it is quite likely that serious damage may result if populations of awl nematodes reach large proportions around the roots of plants.

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The Occurrence of the Fringed Tapeworm, *Thysanosoma actinioides*, in the Pronghorn Antelope

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An inspection of the host records of the Index-Catalogue of Medical and Veterinary Zoology shows that there is a single report, that of Baer (1927, Bull. Biol. France et Belgique, Suppl. 10:127), of the occurrence of the fringed tapeworm, *Thysanosoma actinioides,* in *Antilocapra americana,* the pronghorn antelope. However, an examination of Baer's paper and of the references cited by him fails to reveal the source of the information. It appears therefore that the report lacks authenticity. There are no specimens of the parasite in question from the pronghorn antelope in the Helminthological Collection of the U. S. National Museum. Also, Dikmans (1939, Proc. Helminthol. Soc. Wash. 6:97-101) does not list *T. actinioides* among the parasites reported from this host animal.

It seems appropriate therefore to record our recent finding of this cestode on two occasions in pronghorn antelope in New Mexico.

The first instance occurred in October, 1949, when the viscera of 10 antelope killed during the hunting season in central New Mexico became available for examination. The small intestines were intact in all of these cases but the livers were missing from four of the sets of viscera. Only one specimen of *T. actinioides* was found in these examinations, and it occurred in the small intestine.

*Report of a study made under the Research and Marketing Act of 1946.

In October, 1950, five additional sets of viscera from antelope were examined, and in this instance also only one of the antelope harbored *T. actinioides,* but the number of parasites found was 38. These also were found in the small intestine, the extrahepatic bile ducts being free from tapeworms as well as the pathologic changes which are frequently found in domestic sheep that harbor the parasites [Allen and Kyles (1950. J. Parasitol. 36:45, Sect. 2)].

A point of interest in these infections was that the adult specimens of the tapeworm were considerably smaller than those encountered in the domestic sheep. Longest specimens were only 4 to 5 cm. That some of these were adult tapeworms is evidenced by the fact that detached segments containing egg capsules were found in the intestinal contents, and at least one egg capsule contained a motile onchosphere. The detached segments were only about one-half the size of those usually encountered in the intestinal contents and feces of the domestic sheep, and in the antelope in question, this small size of the segments could be correlated with the small size of the adult tapeworms found.

Curtice [1889. 4. and 5. Ann. Rep., Bureau Animal Indust., U. S. Dept. Agric. (1887-88) pp. 167-186] stated that adult specimens of the fringed tapeworm from sheep range from 15 to 30 cm. in length. We have observed some adult specimens from sheep that were only 8 to 11 cm. in length, but even making allowance for variability in the state of contraction of the tapeforms when measured, there is a striking size difference between those found in sheep and those found in the two antelope mentioned in this report. Further observations may throw a different light on the matter but we mention it here because it is obviously difficult to obtain material from the antelope.

Since the fringed tapeworm is of such common occurrence in domestic sheep in some of the Western States (unpublished data in our possession show an incidence of 54 percent in central New Mexico), a question often arises as to whether wild ruminants play any part in maintaining infection in sheep. In central New Mexico at least, where most of the range is fenced, antelope and sheep are in close contact. There are many instances in which the two species are confined in the same pasture. It should be mentioned also that the geographical distribution of the pronghorn antelope as shown by Anthony (1928. The Field Book of North American Mammals.) corresponds very closely to the geographical distribution of the fringed tapeworm in the domestic sheep as we now know it.

The present report covers the examination of 15 antelope for the fringed tapeworm, of which only two harbored the parasite. In addition, Dr. N. G. Cobbett (Personal communication) of the Zoological Division, U. S. Bureau of Animal Industry, recently examined 4 antelope in east central New Mexico and did not find any of them infected. These data, then show an incidence of only 10.5 percent in the 19 antelope examined, a percentage incidence much lower than that mentioned above for sheep. It is of interest that of the 19 antelope examined, 3 were in pastures occupied by sheep, 7 were in pastures adjacent to those occupied by sheep, and 9 were in pastures that appeared to be some distance from those occupied by sheep. Of the first group, one was infected and of the last group, one was infected.

Thus, in these results, there was no correlation between incidence of the fringed tapeworm in the antelope and the contact these antelope had with domestic sheep.

Monogenetic Trematodes of Westhampton Lake Fishes. III. Part 2. A Discussion of Host-specificity.[1]

WILLIAM J. HARGIS, JR.
Florida State University

The present paper considers the host-parasite relations of the 24 identified species of monogenetic trematodes (Platyhelminthes: Trematoda) which were recovered from a series of 110 fishes of three famiiles (Cyprinidae, Ameiuridae, and Centrarchidae), with a view to further clarification of the problem of host-specificity in the group. The hosts were taken from Westhampton Lake, Richmond, Va.

Although considerable study and speculation concerning host-specificity among the monogenetic trematodes has been reported in the literature, insufficient information on the phenomenon is available for drawing conclusions. However, certain bits of information having a bearing on the specificity question have been made known both from a study of the material in the present collection and of the literature dealing with the species under consideration herein. In reporting these findings the writer has drawn freely from the works of other authors.

RESULTS

Table I lists the hosts and parasites and presents data concerning the numbers of hosts examined and the numbers and species of parasites encountered per host.

The flukes involved belong to the genera *Actinocleidus* Mueller, 1937, *Cleidodiscus* Mueller, 1934, *Dactylogyrus* Diesing, 1850, *Gyrodactylus* v. Nordmann, 1832, *Haplocleidus* Mueller, 1937, *Octomacrum* Mueller, 1934, and *Urocleidus* Mueller, 1934.

The hosts employed are: *Notemigonus crysoleucas crysoleucas* (Mitchill), eastern golden shiner, family Cyprinidae; *Ameiurus nebulosus nebulosus* (LeSueur), northern brown bullhead, family Ameiuridae; *Chaenobryttus coronarius* (Bartram), warmouth, *Lepomis gibbosus* (Linn.), pumpkinseed sunfish, *L. macrochirus macrochirus* (Raf.), bluegill sunfish, and *Pomoxis nigromaculatus* (LeSueur), black crappie, (subfamily Lepominae) family Centrarchidae; and *Micropterus* (*Huro*) *salmoides salmoides* (Lacépède),[2] northern large-mouth bass, (subfamily Micropterinae) family Centrarchidae.

[1]From the Dept. of Biology, Univ. of Richmond, Va., and the Dept. of Zoology, Florida State Univ., Tallahassee.

The writer wishes to thank Dr. Emmett W. Price, Zool. Div., U.S. Bureau of Animal Industry, for valued aid and opinions; Dr. Robert F. Smart, Dept. of Biology, Univ. of Richmond, for aid in securing equipment; Messrs. Marvin Patteson, Shelton Applegate, Ralph Turner, Ray Oglesby, and other of his colleagues at the Univ. of Richmond for aid in the securing of host material for study; and to Drs. Robert Short, Florida State Univ., and H. W. Stunkard, New York Univ., for reading and critisizing earlier drafts of this manuscript.

In an earlier paper (Hargis, W. J., Jr., 1952; A Revision of the genera of the subfamily Tetraonchinae. Proc. Helm. Soc. Wash., 19: 40-4) treating the taxonomy of some of the present species, the title of the key, p. 41, should be amended to read—*A Revised Key to the Genera of North American Fresh-water Tetraonchinae*—because it omits, purposely, the five marine genera found in our Continental waters, and is, therefore, not a "complete key" as claimed.

[2]This is the presently accepted name for the northern large-mouth bass and takes the place of *Huro salmoides salmoides* (Lacépède) as given in previous papers of this series.

DISCUSSION

Host-specificity in the Monogenea is very striking and several authors have discussed it. This present paper serves to further point up this characteristic. Baer (1951) treated some aspects of monogenetic host-specificity and gave a fairly extensive list of refences on the subject. Other references are given below.

It is possible that the direct mode of development is a major factor in this phenomenon. It must be noted, however, that few life histories of monogenetic trematodes are thoroughly known, and that the intimate details of the relations between the hosts and parasites and their physiological reactions to one another must be obtained before a true understanding of the phenomenon under discussion will be obtained. It has also been suggested by Nigrelli (1935) and Nigrelli and Breder (1934) that a physiological mechanism in the host is triggered by the parasite, thus setting up unfavorable conditions to foreign trematodes. Due to the fact that these authors studied captive material in aquaria it may be that some of the host records obtained in their studies are not natural because some of the barriers to "unnatural infections" are broken down when the fish are placed in restricted aquaria or crowded hatchery breeding tanks. However this may be, they discovered that even under these abnormal conditions there are certain unknown boundaries

TABLE I.—Host Distribution of the Present Collection of Monogenea.

Hosts Parasites	*N. c. crysoleucas* (24)	*A. n. nebulosis* (11)	*C. coronarius* (18)	*L. gibbosus* (7)	*L. m. macrochirus* (34)	*P. nigro-maculatus* (10)	*M. s. salmoides* (6)	Total 110
G. elegans					1			1
D. aureus	140		1*		1*			142
D. parvicirrus	15							15
A. fergusoni			1		546			547
A. flagellatus			210		1*			211
A. fusiformis							54	54
A. oculatus				95				95
A. okeechobeensis			33					33
A. recurvatus				46				46
A. sigmoideus				34				34
A. unguis							7	7
C. capax						5		5
C. pricei		6						6
C. robustus			1		64			65
C. stentor						23		23
C. vancleavei						195		195
H. dispar			11	186	171			368
H. furcatus							218	218
U. abaenobryttus			253		3**			256
U. doloresae			14					14
U. ferox			1	515	1,148			1,664
U. principalis							623	623
U. procax				68				68
O. microconfibula	10							10
Total number per host	165	6	525	944	1,935	223	902	4,700

*Considered accidental infection (i.e. carried over in the carrying containers, pipettes or vials or transferred while in the confines of holding tanks, etc.).

**One out of the three accidental, as above.

of specificity which the parasites apparently cannot transcend. The observations of the present study may be somewhat interesting in this respect due to the fact that the hosts came from a relatively restricted impoundment, 12 acres, and that all of the hosts were transported from the lake to the laboratory in small containers and some were held together for short periods (several hours to several days) in a 500 gallon holding tank.

Each of the 24 species in this collection is discussed separately below. Reference may be made to Table I for information necessary in reading the following. For the previously reported hosts and localities of these worms see Hargis (1953).

Gyrodactylus elegans v. Nordmann, 1832. This form is normally a parasite on the skin and fins of its hosts, and, having such an exposed habitat, may not naturally exhibit the same degree of host-specificity as that shown by species that live on the gills of their hosts. The occurrence of a single specimen of this species on the gills of *Lepomis m. macrochirus,* a fish not previously reported as a host, leads the author to believe the infection to be an unnatural or accidental one.

Dactylogyrus aureus Seamster, 1948, previously reported from the gills of *Notemigonus crysoleucas auratus* (Raf.), western golden shiner, by Seamster (1948) is herein recorded from *N. c. crysoleucas,* eastern golden shiner. Even though the species was recovered from the gills of *Chaenobryttus coronarius* and *Lepomis m. macrochirus,* it is probably more normally a parasite of the "shiners" because so few were encountered on the centrarchids. Also, most of those that did occur on these hosts are thought to result from collecting techniques (see table for explanation). Of course, it is possible that strays occur in nature on hosts other than the "shiners."

Dactylogyrus parvicirrus Seamster, 1948. This worm, also, was previously reported from *Notemigonus crysoleucas auratus.* As in the case of *D. aureus,* this species was taken from *N. c. crysoleucas* in the present study. It, therefore, appears to be a parasite of the "shiners."

It is quite possible that the similarity of the ectoparasite fauna in the two subspecies of "shiners," eastern and western, further substantiates the close natural relationships of the hosts, and that the occurrence of the polyopisthocotyleid fluke, *Octomacrum microconfibula* Hargis, 1952, on the eastern subspecies indicates a difference between them. Dr. Aaron Seamster, in a recent communication to the author, states that he did not encounter *Octomacrum* on the western golden shiner in his 1948 study of the ectoparasites of that host in Oklahoma.

Actinocleidus fergusoni Mizelle, 1938, has been reported from *Lepomis m. macrochirus* by several workers, and appears to have a natural predilection for this host. Seamster (1938) recovered it from *L. humilis* (Girard), orange-spotted sunfish, in Oklahoma, but makes no mention of numbers. It is herein reported from *Chaenobryttus coronarius,* but is considered accidental on this host because only one was recovered. In any case it has never been reported on hosts outside of the subfamily Lepominae, family Centrarchidae.

Actinocleidus flagellatus Mizelle and Seamster, 1939. This species is herein reported from both *Lepomis m. macrochirus* and *Chaenobryttus coronarius* which are in the same subfamily. Since only one was recovered from the former, it is considered as normally specific for the latter, and its appearance on fishes other than this host is probably accidental. This conclusion is supported by the work of Mizelle and Seamster (1939).

Actinocleidus fusiformis (Mueller, 1934) Mueller, 1937. According to Mizelle and Regensberger (1945) *A. fusiformis* has been recovered from *Micropterus dolomieu* Lacépède, smallmouth bass, *M. punctulatus* (Raf.), Kentucky bass, and *M. (Huro) s. salmoides.* The numbers in which it occurs on the first two of these hosts are unknown, and attempts to decide the limits of specificity among these hosts are futile. The present specimens were recovered only from the last host. Though this worm occurs on several different species of fish, all of the hosts belong to the same subfamily, Micropterinae, of the family Centrarchidae.

Actinocleidus oculatus (Mueller, 1934) Mueller, 1937, is probably primarily a parasite of *Lepomis gibbosus,* and is considered as such here. Mueller (1936) reports it from the "Sunfish" which has been interpreted by others to mean *L. m. macrochirus,* but gives no indication of numbers. The present study reports it only from *L. gibbosus* although there was ample opportunity for cross-infection in the nests and holding tanks. Whether it is chiefly an *L. gibbosus* parasite or not, both fish are in the same subfamily, Lepominae, of the family Centrarchidae.

Actinocleidus okeechobeensis Mizelle and Seamster, 1944. This fluke has not been reported from any other host and is probably specific for *Chaenobryttus coronarius.* Its occurrence is not rare, but there are usually few per host.

Actinocleidus recurvatus Mizelle and Donahue, 1944. This worm is probably specific for *Lepomis gibbosus:* It has been reported only from this host.

Actinocleidus sigmoideus Mizelle and Donahue, 1944. As far as is known, this fluke is specific for *Lepomis gibbosus.* It occurs only on this host in Westhampton Lake, and the results of Mizelle and Donahue (1944) bear out this conclusion.

Actinocleidus unguis Mizelle and Cronin, 1943, is probably specific for *Micropterus (Huro) s. salmoides,* and the studies of Mizelle and Cronin (1943) and Mizelle and Regensberger (1945) support this conclusion.

Cleidodiscus capax Mizelle, 1936, has been previously reported from the gills of *Pomoxis nigromaculatus* and *P. annularis* (Raf.), white crappie, by Mizelle, LaGrave and O'Shaughnessy (1943). The present investigation supports the observation of these authors that it is probably strictly a parasite of the genus *Pomoxis.*

Cleidodiscus pricei Mueller, 1936, is herein reported from *Ameiurus n. nebulosus.* Other authors (see Mizelle and Regensberger, 1945) have reported it from *A. n. natalis* (LeSueur), yellow bullhead, *A. m. melas* (Raf.), black bullhead, *Ictalurus furcatus* (LeSueur), Fulton cat, and *I. lacustris punctatus* (Walb.), channel cat. Therefore, this worm is unusually adaptable to many different host species, but all of them belong to the family Ameiuridae. This is the usual situation for those Monogenea that are not strictly species-specific.

Cleidodiscus robustus Mueller, 1934. This worm is reported herein from *Lepomis m. macrochirus* and *Chaenobryttus coronarius,* but only one was recovered from the latter. Previous authors (see Mizelle and Regensberger, 1945) have reported it from *L. m. macrochirus, L. cyanellis* (Raf.), green sunfish, *L. gibbosus* and "Bass." *C. robustus* is probably primarily a parasite of *L. m. macrochirus,* or at least of the genus *Lepomis.* In any event all of the hosts, except the "Bass" are in the same subfamily. All of them are in the family Centrarchidae.

Cleidodiscus stentor Mueller, 1937. This is the first instance of the recovery of this worm from any host other than *Ambloplites r. rupestris* (Raf.), rock bass. During the present study it was recovered in numbers from *Pomoxis nigromaculatus* but may be primarily a parasite of the former host; however, both fish are in the subfamily Lepominae.

Cleidodiscus vancleavei Mizelle, 1936, is herein recorded from *Pomoxis nigromaculatus*. Mizelle, LaGrave, and O'Shaughnessy (1943) stress the fact that *C. capax*, see above, and *C. vancleavei* regularly occur together on the gills of individuals of this host species whereas they, plus *Cleidodiscus longus* Mizelle, 1936 and *C. uniformis* Mizelle, 1936, are found together on the gills of individual specimens of *P. annularis*. This pattern of infestation was characteristic and constant in these hosts and led Dr. Mizelle, the senior author, to contend that a diagnosis of the host species—either *P. nigromaculatus* or *P. annularis*—could be made from an analysis of the parasite fauna of the gills. This present study partially supports Dr. Mizelle's contention, but it would have been interesting to have had specimens of *P. annularis* for a more complete comparison.

Haplocleidus dispar (Mueller, 1936) Mueller, 1937, is not as highly specific as some of its relatives. It has been reported from *Lepomis gibbosus, L. m. macrochirus, L. humilis,* and *Micropterus* (*Huro*) *s. salmoides,* see Mizelle and Regensberger, (1945). The present study reports it from *Chaenobryttus coronarius:* It was also recovered in numbers from the first two fish above. The regularity with which it has been taken from the several hosts is such that it cannot be considered specific for either host; however, in no case has *H. dispar* been reported on hosts outside of the subfamilies Lepominae and Micropterinae, family Centrarchidae.

Haplocleidus furcatus Mueller, 1937, has been previously reported from *Micropterus* (*Huro*) *s. salmoides* and *M. punctulatus* (see Mizelle and Regensberger, 1945). The results of the present study establish the fact that it is commonly a parasite of the former host. The numbers in which it occurs on the latter fish are unknown; therefore, the limits of specificity between the two hosts cannot be estimated. Both hosts are members of the subfamily Micropterinae.

The Monogenea which frequent hosts belonging to this piscine subfamily commonly infest several different species of the group. This is somewhat unusual. It is possible that this similarity of ectoparasitic infestation between the species of this subfamily further demonstrates their close taxonomic relationship. It is also possible that it is an indication of the recent evolutionary delimitation of species in this group.

Urocleidus chaenobryttus Mizelle and Seamster, 1939, has never been reported from any host other than *Chaenobryttus coronarius* in sufficient numbers to cause it not to be regarded as principally a parasite of this fish. Mizelle and Jaskoski (1942) reported it from *Lepomis miniatus* Jordan, stumpknocker sunfish, but recovered only one which may easily have been a case of accidental infestation. It is recorded herein from *L. m. macrochirus,* but only three flukes were recovered (one of which is probably accidental).

Urocleidus doloresae Hargis, 1952, is apparently not a commonly recovered form and has never been reported from any other body of water. It may be specific for *Chaenobryttus coronarius* since none of the other hosts from Westhampton Lake harbored it.

Urocleidus ferox Mueller, 1934. The present study reports *U. ferox* in numbers from *Lepomis gibbosus* and *L. m. macrochirus.* One worm was recovered from *Chaenobryttus coronarius,* which case is regarded as accidental. Other studies (see Mizelle and Regensberger, 1945) have reported it from *L. m. macrochirus, L. gibbosus, L. humilis,* and hybrids between *L. m. macrochirus* and *L. humilis,* and *L. gibbosus* and *L. humilis.* The numbers of worms encountered during these previous studies are unknown; therefore, the limits of specificity are impossible to define. All of these hosts belong to the subfamily Lepominae of the family Centrarchidae.

Urocleidus principalis (Mizelle, 1936) Mizelle and Hughes, 1938. This worm is a parasite of the subfamily Micropterinae, family Centrarchidae. During this study it was encountered only on *Micropterus* (*Huro*) *s. salmoides,* but it has been reported by Mizelle and Regensberger (1945) from *M. punctulatus* and *M. dolomieu. U. principalis* possesses the characteristic of parasitizing a variety of hosts within the same piscine subfamily as does *Haplocleidus furcatus,* above.

Urocleidus procax Mizelle and Donahue, 1944, has never been reported, herein or elsewhere, from any host other than *Lepomis gibbosus* for which it is probably specific.

Octomacrum microconfibula Hargis, 1952. Too little is known concerning this fluke to establish the limits of specificity. Dr. Aaron Seamster states, personal communication, that he did not recover any species of *Octomacrum* Mueller, 1934 from *N. crysoleucas auratus* in his Oklahoma studies.

CONCLUSIONS

The results and discussion of the present paper indicate that a fairly high degree of host-specificity exists among the monogenetic trematodes studied. The work of other authors on the same species supports this conclusion. Even those that occur on more than one host species are, with the single exception of *Dactylogyrus aureus,* confined to a subfamily or, at most, a family. The occurrence of *D. aureus* on a fish which is not a species of its normal host family, Cyprinidae, was probably accidental.

Indeed, the specificity as reported herein and elsewhere (Mizelle, LaGrave and O'Shaughnessy, 1943) is so marked that it is possible to make a fairly accurate identification of the hosts studied above by an analysis of the ectoparasite fauna of the branchial material, provided sufficient material is checked.

With this degree of host-specificity existing in the Monogenea it is probable that more extensive study of them, especially their life-histories, physiology and ecology, with the aim of better understanding the natural taxonomy and phylogeny of both the hosts and parasites will prove profitable. It is certain that it will prove interesting.

SUMMARY

The host-parasite relationships of twenty-four species of Monogenea representing the genera *Actinocleidus* Mueller, 1937 (8), *Cleidodiscus* Mueller, 1934 (5), *Dactylogyrus* Diesing, 1850 (2), *Gyrodactylus* v. Nordmann, 1832 (1), *Haplocleidus* Mueller, 1937 (2), *Octomacrum* Mueller, 1934 (1), and *Urocleidus* Mueller, 1934 (5) have been discussed, and the following conclusions made: 1—The monogenetic trematodes studied, and probably most Monogenea, exhibit a very high degree of host-specificity. Many of them

studied are confined to a single host species, most of them to a subfamily, and nearly all to a piscine family. 2—Existing knowledge concerning the life-histories, physiology, and ecology of the Monogenea must be greatly augmented before more extensive and definite conclusions can be made concerning host-specificity in this group. 3—It is possible that additional knowledge of the ectoparasitic trematodes may throw light on the phylogeny and natural taxonomy of the hosts and vice versa.

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Some Monogenetic Trematodes of Marine Fishes from Fiji*

H. W. MANTER AND DONALD F. PRINCE

This paper describes three new species of monogenetic trematodes collected by the senior author from fishes at Suva, Fiji, in 1951. A new genus is named for one of these species. A fourth species, previously known from India, is identified. Parasites of the numerous fishes of the South Pacific have been studied very little and, so far as we can learn, no Monogenea has been reported from this region. One difficulty met in collecting there was lack of adequate identification of the hosts. In these particular cases, only the common or native names of the fishes were obtained. Often, however, the general type of family of the fish was evident.

The specimens were killed in formol-alcohol-acetic solution under slight pressure with a cover glass, and preserved in 70% alcohol. They were stained in Delafield's hematoxylin and mounted in Permount.

I. Suborder POLYOPISTHOCOTYLEA Odhner, 1912

A. Family Discotylidae Price, 1936

a. Subfamily Vallisiinae Price, 1943

1. *Lethacotyle fijiensis* n. gen., n.s.p. (Rigs. 1-6).

DESCRIPTION (based on two specimens): Body elongate, pointed anteriorly, widest about 2/3 body length from anterior end. Total length 3.156 to 3.759 mm., greatest width 0.663 to 0.770 mm. The small haptor, at the posterior end, is a transversely extended lobe bearing three pairs of hooks. Another lobe with ventrally curved anterior and posterior ends lies on the right side of the body adjacent to the haptor. There is no trace of clamps. Largest hooks (Fig. 2) are most lateral. Anterior root of this hook thick and apparently double with fused halves; posterior root short, the blade sharply recurved; length from base of anterior root to curvature of blade, 0.024 mm. Just median to these hooks is a pair of very slender, needle-like hooks with long anterior root and short blade (Fig. 4). These are 0.014 mm. long. Third pair of hooks median, 0.016 mm. long, with single roots (Fig. 3). Dorsal surface of haptor with fine transverse striations. Left side of the body from base of haptor for a distance of about 0.710 mm. provided with small scales pointed anteriorly. MacCallum (1918) described similar spines for *Protomicrocotyle mirabilis* (MacCallum, 1918) Johnston and Tiegs, 1922.

Oral suckers 0.049 to 0.052 mm. in diameter; pharynx 0.064 by 0.050 mm.; esophagus 0.670 mm. long, bifurcating 0.871 mm. from anterior end of body; ceca with both median and lateral branches; each cecum ends blindly at base of haptor.

Ovary posttesticular, near posterior end of body, consisting of a longitudinally coiled tube with lobed proximal end and both ends posterior. Vitellaria beginning a short distance posterior to intestinal bifurcation and extending not quite to posterior end of body. No eggs present. Atrial pore median and ventral about 1/5 body length from anterior end. The thick-walled atrium is armed with a circle of 24 or 25 conspicuous, sickle-shaped spines 0.024 mm. long, all of approximately equal length (Fig. 5). Vagina a conspicuous, muscular and glandular organ lying near but to the left and rear of

*Studies from the Department of Zoology, University of Nebraska, No. 260.

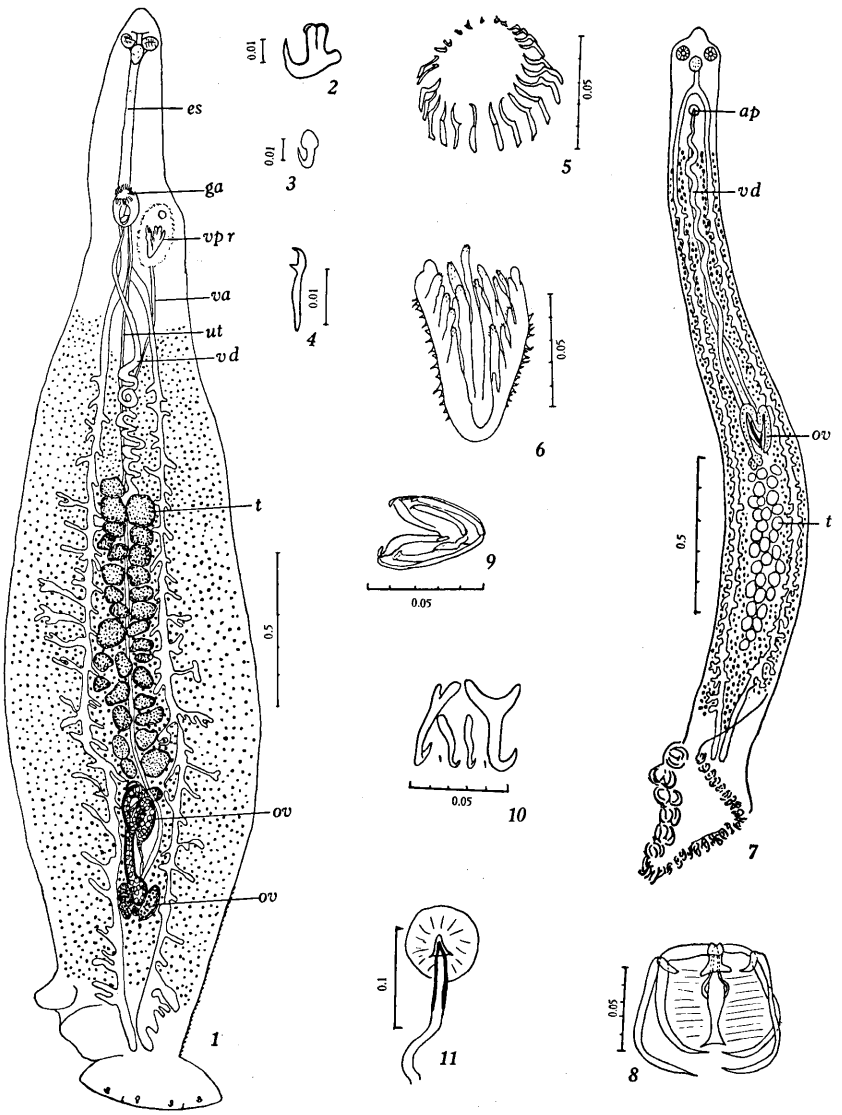

PLATE I

Fig. 1. *Lethacotyle fijiensis.* Entire worm, ventral view.
Figs. 2-4. Haptoral hooks of *L. fijiensis.*
Fig. 5. Hooks of genital atrium of *L. fijiensis.*
Fig. 6. Vaginal processes of *L. fijiensis.*
Fig. 7. *Cemocotyle saquae.* Ventral view.
Fig. 8. Large clamp of *C. saqae.* Ventral view.
Fig. 9. "Muzzle" type clamp of *C. saqae.* Lateral view.
Fig. 10. Haptoral hooks of *C. saqae.*
Fig. 11. Genital atrium and cirrus of *C. saqae.*

genital atrium. It contains a basal cluster of thinly chitinized processes of varying length most of which are long and slender rather than cone-shaped as in *P. pacifica.* An odd feature is the presence of minute spines on the tips and in some cases along the sides of these vaginal processes (Fig. 6). These fine spines can be seen only by high magnification but they do not occur in *P. pacifica.*

Testes 30, in two rows filling midbody region between ceca anterior to ovary. Vas deferens sinuous between ceca, then straightening near bifurcation of ceca.

HOST: yellow jack (Family Carangidae)

LOCATION: Gills

LOCALITY: Suva, Fiji Islands

TYPE SPECIMENS U. S. Nat. Mus. Helminth. Collection No. 48718.

GENERIC DIAGNOSIS OF LETHACOTYLE. Characters of the subfamily Vallisiinae Price, 1943 as emended by Sproston, 1946. Clamps lacking. Three pairs of hooks on the small, lobe-like haptor. Atrial hooks of one type. Vagina muscular and glandular, with chitinized processes. Type species: *L. fijiensis.*

The name *Lethacotyle* is from *letha* = forgetting, and *cotyle* = cup, and refers to the absence of clamps.

DISCUSSION: This trematode is most closely related to *Protomicrocotyle* Johnston & Tiegs, 1922, differing chiefly in the entire absence of all clamps. While there is a possibility that these could have been lost, both specimens were in excellent condition and had retained even the minute hooks of the haptor. Two species of *Protomicrocotyle* have been named, *P. mirabilis* (MacCallum, 1918) Johnston & Tiegs, 1922 and *P. pacifica* Meserve, 1938.

P. pacifica differs, in addition to having four clamps, in the number, size, and shape of the haptoral hooks; in very different atrial spines; in shape of the vaginal processes. *P. mirabilis* is still inadequately described. It resembles *L. fijiensis* in the shape of the largest haptoral hooks, location of the vagina, and general shape of the ovary. *P. mirabilis,* however, has four clamps; the structure of the vagina is different; and there are two sizes of spines in the genital atrium.

Another related genus is *Bilaterocotyle* Chauhan, 1945 based on a species (*B. chirocentrosus*) from the gills of *Sciaena belengeri* and *Chirocentrua dorab* at Bombay, India. It was distinguished from *Protomicrocotyle* by the presence of six rather than four clamps. It differs from *Lethacotyle* in possessing the six clamps, an unarmed vagina, and the ovary is at midbody.

EXPLANATION OF PLATES

All the drawings were made with the aid of a camera lucida. The value of the projected scale is indicated in millimeters in each figure.

ABBREVIATIONS USED IN FIGURES

ap	atrial pore	*sr*	seminal receptacle
c	cirrus	*t*	testis
ch	cirrus hooks	*ut*	uterus
es	esophagus	*va*	vagina
ga	genital atrium	*vc*	vaginal cone
lp	lateral plate of atrial cavity	*vd*	vas deferens
mp	median plate of atrial cavity	*vp*	vaginal pore
ov	ovary	*vpr*	vaginal processes

B. Family Microcotylidae Taschenberg, 1879
b. Subfamily Gastrocotylinae Sproston, 1946

2. *Pseudaxine indicana* Chauhan, 1945

HOST: "salala" or mackerel (Family Scombridae)
LOCATION: Gills
LOCALITY: Suva, Fiji Islands

This report constitutes a new host and geographical record. Chauhan records it from *Chrysophrys berda* from the coast of India. A single specimen collected in Fiji is deposited at U. S. Nat. Mus. Helminth. Collection, No. 48721.

C. Subfamily Microcotylinae Monticelli, 1892

3. *Cemocotyle sagae* n.sp. (Figs. 7-11)

DESCRIPTION (Based on three specimens. Measurements are of the holotype): Length 3.095 mm., greatest width 0.272 mm. Anterior end conical, widening abruptly at level of oral suckers. Body width increasing gradually to a maximum near midbody, then remaining about the same. Haptor asymmetrical, V-shaped, bearing three pairs of hooks near its posterior end (Fig. 10). The lateral, largest hooks have bifid roots, strongly curved tips, and measure 0.040 mm. in length. The mesial, medium-sized hooks have unforked roots, slightly curved points and measure 0.027 mm. in length. The smallest hooks are only 0.006 mm. long, with sharp point and root region showing three fine parallel lines. Eighteen to 23 small clamps occur on the longer (left) side of the haptor; six larger clamps are on the shorter (right) side. Larger clamps, of typical microcotylid structure, measure 0.060 mm. long and 0.068 mm. wide (Fig. 8). The smaller clamps measure 0.034 to 0.147 mm. in length by 0.045 to 0.181 mm. in width. Many of these smaller clamps have the typical microcotylid structure but a varying number are of the "muzzle" type (Fig. 9) described by MacCallum (1913). These are seen in side view and have anterior and posterior halves gaping apart like clam shells rather than like a muzzle. In these clamps, the single median sclerite is replaced by a median pair, and each half of the clamp has an anterior and a posterior pair of sclerites. Each of the four latter has a minute, spine-like piece at its tip, and the anterior-median sclerite is also sharply pointed. Of 20 small clamps in the holotype specimen, only two were of the "muzzle" type. The number and location of such clamps can be indicated for each specimen by enumerating the numbers beginning at the anterior end of the haptor and placing the "muzzle" types in parentheses. Holotype: 5 + (2) + 13. Paratypes: (10) + 4 + (2) + 3 + (3); (7) + 2 + (9).

Oral suckers 0.037 mm. in diameter; pharynx 0.040 by 0.037 mm.; esophagus 0.188 mm. long, bifurcating about 0.335 mm. from anterior end of body. Ceca with both lateral and median side branches except that the anterior 0.355 mm. and the posterior 0.268 mm. are without branches. Ceca end at anterior edge of the short side of the haptor.

Ovary approximately at midbody; poorly defined; it is tubular and seems to show two longitudinal coils. Neither a seminal receptacle nor a vagina could be seen. Uterus a straight tube leading to the ventral atrial pore just anterior to intestinal bifurcation. A conspicuous atrial or genital bulb present with weak radial striations and a chitinous, triradiate lumen into which the chitinous cirrus opens. Vitellaria lateral along the branched extent of the ceca and median to ceca posterior to testes; not extending posterior to ceca. A

single collapsed egg measured 0.075 by 0.017 mm. It had a short twisted filament at its anterior end but none at its more pointed posterior end. It may have been abnormal. Testes 32 to 35, rounded, postovarian, extending posteriorly from ovary about 0.084 mm., not reaching ends of ceca. Vas deferens almost straight until about halfway to genital pore where it becomes slightly sinuous. Cirrus a straight, unspined, chitinous tube about 0.045 mm. long and 0.008 mm. wide at its base. Its distal end is abruptly pointed.

HOST: "Saqa" (Caranx sp.)

LOCATION: Gills

LOCALITY: Suva, Fiji

TYPE SPECIMEN: U. S. Nat. Mus. Helminth. Collection, No. 48719

DISCUSSION: The genus *Cemocotyle* was named by Sproston (1946:450) for *Microcotyle carangis* MacCallum, 1913 from *Caranx crysos* from the North American Atlantic. The genus resembles *Microcotyle* except for the haptor which is asymmetrical with typical clamps on the short side but with some peculiar "muzzle" type clamps on the longer side. Two pairs of hooks were described at the posterior tip of the haptor.

The excellent type specimen of *C. carangis,* mounted with a paratype, was kindly loaned by Dr. E. W. Price. It shows the third pair of very small haptoral hooks (about 5 μ long) which resemble those of *C. saqae.* The "muzzle" type clamp is very clearly distinct consisting essentially of six sclerites in three rows each of two, end to end sclerites. The conspicuous vagina possesses pointed chitinous processes suggestive of *Protomicrocotyle.* The vaginal pore is ventral. Although the pores are not distinct, it is believed the uterus opens with the cirrus rather than separately.

C. saqae is very different from *C. carangis* in its unspined genital atrium, simple chitinous cirrus, distribution of vitellaria, and much smaller size. The chitinous cirrus is somewhat similar to that in *Microcotyloides incisa* (Linton, 1940) Fujii, 1944 which, however, has in addition a prostatic bulb as well as other differences.

4. *Lintaxine microcotyla* n.sp. (Figs. 12-17)

DESCRIPTION (Based on 9 specimens; with measurements of the holotype): Total length 4.134 mm., greatest width 0.302 mm. near midbody; almost equally wide along entire length. Left side of haptor 1.3 mm. long, bearing 33 large, stalked clamps; right side of haptor 0.838 mm. long, bearing about 61 exceedingly minute clamps. Large clamps 50 to 60 μ in greatest diameter; small clamps almost incredibly small, only 8 to 9 μ in diameter. In spite of this minute size, the delicate armature of the small clamps has sclerites similar to those of the large clamps. Large clamps (Fig. 13) with the following sclerites: median sclerite with bifid posterior end; directly at its tip ventrally is a pair of small, curved pieces curving apart medianly but meeting at each end; a pair of outer, lateral sclerites with sharp bifid tips almost meeting at distal end of clamp; an inner, shorter, lateral pair of sclerites, each with a short transverse piece at the distal end. At the proximal junction of the lateral sclerites on each side, a short piece with hook-like point extends diagonally inward. Inconspicuous, transverse striations occur within the wall of the clamp.

Mouth subterminal; oral sucker ovoid, 0.044 to 0.049 by 0.061 to 0.070 mm. Pharynx 0.036 by 0.034 mm.; esophagus 0.275 mm. long; ceca un-

branched for about 0.194 mm., then with lateral branches until shortly anterior to haptor where they become unbranched, extending into haptor about 2/3 its length.

Testes with somewhat indistinct outline, probable number 26 to 31; postovarian; intercecal; from level of ovary to slightly less than halfway to posterior end of body. Vas deferens sinuous, leading to an armed, thin-walled sac, the cirrus, just posterior to the genital atrium. Cirrus hooks in right and left sets, each set consisting of a dorso-ventral row of 7 hooks (Fig. 15) with broad bases and recurved tips. In these rows the three middle hooks are largest, the next one on each side somewhat smaller, while the most dorsal and ventral hooks are smallest.

Ovary near midbody, tubular, extending forward about 0.817 mm., then backward a short distance. Uterus a straight tube. Genital atrium large; ventral to intestinal bifurcation; encircled with small gland cells; provided with three spiny plates or pads, one median with a transverse anterior edge measuring 0.070 mm. across and armed with rather long spines, two lateral and more dorsal (Fig. 14). Vagina very short but conspicuous, opening on median dorsal surface opposite distal end of ovary, approximately at midbody; it consists of an ovoid, unarmed sac containing a ventral, cone-shaped papilla. Gland cells surround the vaginal sac (Fig. 17). A small, spherical seminal receptacle lies just posterior to the vagina. Vitellaria begin shortly posterior to genital atrium and extend to a level about opposite hindmost testis. Only one normal egg was seen (Fig. 16); it measured 0.121 by 0.040 mm. and had a filament at each end.

HOST: "ribbon fish"

LOCATION: gills

LOCALITY: Suva, Fiji Islands

TYPE SPECIMEN: U. S. Nat. Mus. Helminth. Collection, No. 48720

DISCUSSION: The only other species in the genus *Lintaxine* Sproston, 1946 is *L. cokeri* (Linton, 1940) Sproston, 1946 (= *Heteraxine cokeri* Linton, 1940) from the gills of the freshwater drum, *Aplodinotus grunniens,* at Fairport, Iowa. This rather surprising relationship prompted an examination of the type material of that species. The slide, containing the holotype and 5 paratypes, was kindly loaned by Dr. E. W. Price. Since Linton's description is rather incomplete a few details are added here. The clamps, all sessile, are of typical *Axine* structure as suspected by Sproston. The "flange of unarmed haptor tissue" noted by Sproston is not very evident and probably represents only the edge of the body. The large clamps have rather strong sclerites (Fig. 18). At the extreme posterior tip of the body is a single clamp about half the size of the large clamps and twice the size of the small clamps. There is a single, median, unarmed, dorsal vaginal pore midway between the anterior edge of the ovary and the atrial pore. The armature of the cirrus (Fig. 19) is complex although consisting of a single ring or spines or hooks, not two rings as described by Linton. Ventrally and laterally there is a partial circle of slightly curved, thorn-shaped hooks, 12 in number, all about equal in size. Dorsally in the ring, these hooks are replaced by very long, needle-like spines, 7 in number. The three middle spines are longest and are very slightly knobbed at the end. To each side of these three, the spines are progressively shorter, pointed, and provided with curious, hair-like processes on the outer side of their distal halves. All these spines project into the atrial cavity from the anterior side.

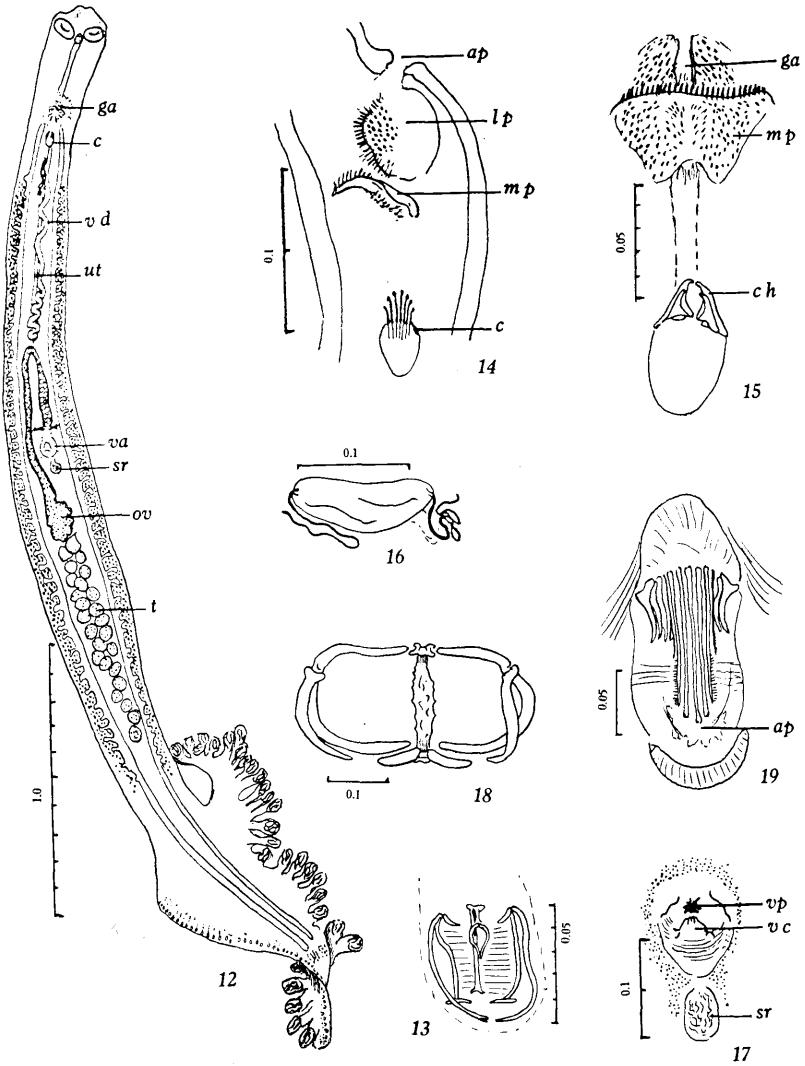

PLATE II

Fig. 12. *Lintaxine microcotyla.* Ventral view.
Fig. 13. Large haptoral clamp of *L. microcotyla.* Ventral view.
Fig. 14. Genital atrium and cirrus of *L. microcotyla.* Lateral view.
Fig. 15. Genital atrium and cirrus of *L. microcotyla.* Ventral view.
Fig. 16. Egg of *L. microcotyle.*
Fig. 17. Dorsal view of vaginal region of *L. microcotyla.*
Fig. 18. Large clamp of *Lintaxine cokeri* (Linton, 1940). Ventral view.
Fig. 19. Cirrus and cirrus spines of *L. cokeri.* Ventral view.

This study of the type specimens of *Lintaxine cokeri* confirmed relationship to the Fijian material. Generic criteria now used in taxonomy of Monogenea would seem to permit the two species in one genus. There are, however, a number of distinct differences. Perhaps the most conspicuous of these, other than sizes, are the stalked large clamps and the very minute and numerous small clamps of *L. microcotyla.* The armature of the cirrus and atrium is also different, particularly the numerous small atrial spines and the bilateral rows of cirrus hooks in *L. microcotyla.* However, the thorn-like shape of the cirrus hooks is similar in both species. *L. microcotyla* has a more posterior vaginal pore and a smaller egg with a filament at each end rather than at one end only.

In view of the considerable host specificity shown by Monogenea, the resemblance of this species from a marine fish in the Fiji Islands to a species from a freshwater fish in Iowa is surprising. It is unfortunate that the "ribbon fish" was not more exactly identified. It might be noted that the drum, *Aplodinotus grunniens,* is one of the few freshwater species in the family Sciaenidae. Its marine affinities are shown by the fact it is host to two species of *Microcotyle,* a large genus of marine Monogenea. The family Sciaenidae is not known to occur in the islands of the South Pacific.

SUMMARY

The following monogenetic trematodes from marine fishes of Fiji are described: *Lethacotyle fijiensis* n.gen., n.sp. (Family Discotylidae) from a "yellow jack" (Family Carangidae); *Cemocotyle saqae* n.sp. (Family Microcotylidae) from a "saqa" (*Caranx* sp.); and *Lintaxine microcotyla* n.sp. (Family Microcotylidae) from a "ribbon fish." *Pseudaxine indicana* Chauhan, 1945 (Family Microcotylidae), formerly known from India, is reported from a "salala" (Family Scombridae) in Fiji.

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Hypocaryophyllaeus gilae n.sp. (Cestoda: Caryophyllaeidae) from the Utah Chub, *Gila straria*, in Wyoming*

JACOB H. FISCHTHAL

In a study of the parasites of fishes in the upper Snake river drainage of Wyoming, Bangham (1951) reported two species of Caryophyllaeidae from a cyprinid fish, the Utah chub, *Gila straria* (Girard). These were listed as "*Glaridacris laruei* and 2nd spp.," occurring in 86 (12.8 per cent) of 670 hosts examined during July and part of August in 1949 and 1950. He stated that the second species was similar to *Caryophyllaeus terebrans* (which he found in the rosyside sucker, *Catostomus fecundus*), but "size and other differences have prevented positive identification as yet."

Dr. Ralph V. Bangham, College of Wooster, Ohio, kindly contributed to the author an abundant supply of the caryophyllaeid tapeworms from the Utah chubs. After consideration of whole mounts, and serial cross and sagittal sections, the author finds that only one species of caroyphyllaeid is represented, instead of two. This species is a new form in the genus *Hypocaryophyllaeus* Hunter, 1927, for which the name *H. gilae* is proposed.

According to Bangham (1951), the worms sent to the author were fixed in hot 10 percent formalin and stored in a 5 percent solution. Some of the specimens received were already mounted in balsam after having been stained in Delafield's haematoxylin. The unmounted worms were stained by the author in Mayer's paracarmine (whole mounts) or Harris' haematoxylin (sections) and also mounted in balsam.

Sincere appreciation is due to Dr. R. V. Bangham for furnishing the specimens for study. Appreciation is also due to Dr. E. W. Price, Zoological Division, Bureau of Animal Industry, U. S. Department of Agriculture, for the loan of the type specimen of *Hypocaryophyllaeus paratarius* Hunter, 1927 (two slides of serial sections in the U. S. Nat. Mus. Helm. Coll. No. 51150; formerly slides No. 29.45 a-b in the collection of the late Dr. Henry B. Ward), and to Dr. F. G. Wallace, University of Minnesota, for the loan of a whole mount cotype specimen from the Department of Zoology collection (slide No. G-40-13, from *Carpiodes carpio*, Rock River, Illinois).

Hypocaryophyllaeus gilae n.sp.

DIAGNOSIS: With characters of genus. Body elongate, oval in cross section; widest at level of anterior testes, tapering only slightly to blunt point posteriorly (fig. 1). Scolex indistinctly defined from neck, bearing a flattened terminal disc and three pairs of distinct loculi; relationship of disc and loculi are such as to form the Greek letter "II." Cuticula thick. Subcuticular layer, between cuticula and outer longitudinal muscles, approximately of same thickness or thicker than cuticula. Inner and outer longitudinal muscles, originating in scolex and extending length of body, delineate medullary and cortical parenchyma, respectively; dorso-ventral muscles present. Terminal excretory bladder present.

Testes approximately 68-96 in number; usually transversely elongated; occur in one or two layers and in two or three longitudinal rows; entirely

*Contribution No. 5 from the Department of Biological Sciences, Harpur College, State University of New York, Endicott, New York.

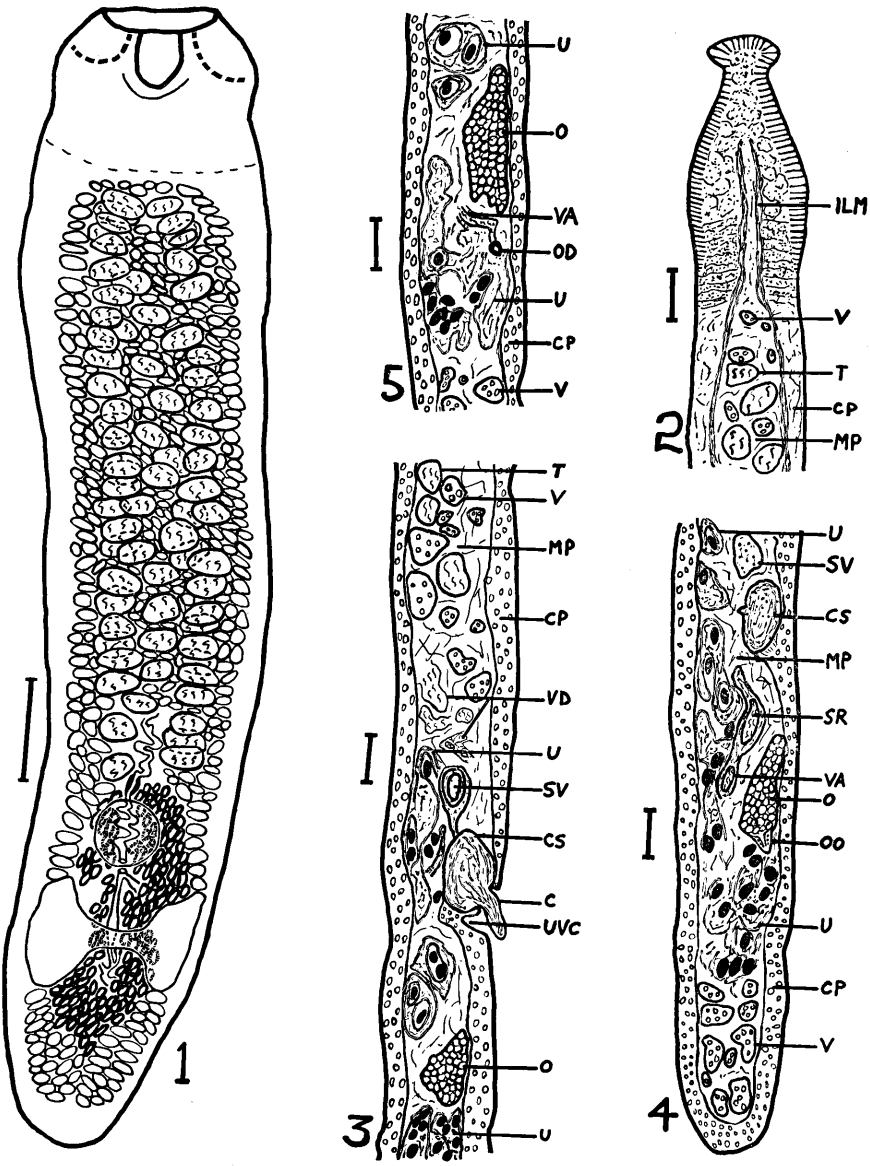
medullary (figs. 1-3). Vas deferens in middle portion of medullary parenchyma, surrounded by testes and vitellaria; anterior to and outside of cirrus sac the vas deferens is much convoluted, enlarging into a relatively thick-walled seminal vesicle; short ejaculatory duct leaves vesicle to enter cirrus sac. Cirrus sac in ventral portion of body, large, nearly circular, and surrounded by layers of circular muscles (figs. 1, 3 and 4). Cirrus convoluted, opening on ventral surface anterior to and separate from the utero-vaginal canal opening; cirrus protrusible (figs. 1 and 3).

Ovary H-shaped, distinctly lobate; wings connected by narrow ventral commissure; entirely medullary; just behind cirrus sac. Oöcapt muscular, located at postero-medial margin of ovarian commissure; surrounded by single layer of unicellular glands (figs. 1 and 4). Oviduct short, thick-walled, surrounded on outside by gland cells; passes posteriorly a short distance before being joined by vagina (figs. 1 and 5). Junction point of oviduct and vagina forms vaginal-oviducal canal; latter passes posteriorly a short distance before being joined by common vitelline duct; other traces of vitelline duct not discernible. Uterus continues from this point through highly glandular oötype to beyond posterior ends of ovarian wings and up to area of post-ovarian vitellaria; much convoluted uterus extends anteriorly dorsal to ovarian commissure, cirrus sac, and seminal vesicle, to posterior portion of testicular field; uterus joins vagina immediately behind cirrus sac, forming short utero-vaginal canal which opens on ventral surface posterior to and separate from cirrus; uterus surrounded by glands throughout entire length, being especially numerous anterior to ovarian commissure (figs. 1, and 3-5). Vagina thick-walled, surrounded by unicellular glands along its length; becomes distended into seminal receptacle antero-dorsal to ovarian commissure and between anterior arms of ovary; vagina joins oviduct a short distance posterior to ovarian commissure (figs. 1, 4 and 5). Vitellaria extensively developed; entirely medullary; follicles small compared to testes; extend posteriorly throughout medullary parenchyma from anterior to testes, along each side and between testes, to just below testicular field, disappearing in center but continuing laterally in narrow row to anterior tips of ovarian wings, occasionally overlapping them slightly; just behind posterior tip of each ovarian wing and occasionally overlapping them, a row of vitellaria extends posteriorly for short distance before merging in median line; these continue posteriorly throughout medullary parenchyma almost to posterior tip of body (figs. 1-5). Intrauterine eggs oviform; numerous in fully matured specimens.

Mean measurements in millimeters (with minima and maxima in parentheses) of 12 whole mount specimens containing eggs in the uterus are: Body, length 3.12 (2.24-4.43), width 0.62 (0.54-0.78); distance from anterior end of body to beginning of vitelline field, 0.41 (0.31-0.50); pre-ovarian field of

ABBREVIATIONS

C	cirrus	SR	seminal receptacle
CP	cortical parenchyma	SV	seminal vesicle
CS	cirrus sac	T	testis
ILM	inner longitudinal muscles	U	uterus
O	ovary	UVC	utero-vaginal canal
OD	oviduct	V	vitellaria
OO	oöcapt	VA	vagina
MP	medullary parenchyma	VD	vas deferens



All figures refer to *Hypocaryophyllaeus gilae*, and were drawn with the aid of a microprojector. The value of the scale is 0.3 mm. for Fig. 1 and 0.1 mm. for Figs. 2-5.

Fig. 1. Adult worm, ventral view.

Fig. 2. Sagittal section through middle pair of loculi.

Fig. 3. Sagittal section through middle of cirrus sac and utero-vaginal canal; note the protruded cirrus.

Fig. 4. Sagittal section through oöcapt region.

Fig. 5. Sagittal section at junction point of oviduct and vagina.

vitellaria, length 2.02 (1.39-2.88); post-ovarian field of vitellaria, length 0.36 (0.23-0.44); longitudinal extent of testicular field, 1.69 (1.20-2.47); longitudinal extent of uterine coils, 0.73 (0.52-1.02); ratio of length of testicular field to that of uterine coils, 1:0.43 (1:0.37-1:0.47); distance from anterior margin of cirrus sac to posterior end of body, 0.90 (0.61-1.26); distance from anterior margin of ovarian wings to posterior end of body, 0.74 (0.48-1.08); right ovarian wing, length 0.32 (0.17-0.50); left ovarian wing, length 0.30 (0.21-0.53); testes, length 0.070 (0.051-0.083), width 0.093 (0.075-0.108); intrauterine eggs, length 0.045 (0.041-0.051), width 0.030 (0.029-0.033).

HOST: *Gila straria* (Girard).

LOCALITIES: Emma Matilda lake, Snake river, Two Ocean lake, Pacific creek, Moran creek, and Kelly Warm Springs, in Teton County, Wyoming, U.S.A.

HABITAT: Small intestine.

TYPE: U. S. Nat. Mus. Helm. Coll. No. 48701 (type) and No. 48702 (paratypes). (Consists of 1 whole mount slide of the type specimen, 1 slide with 6 whole mount paratypes, and 2 slides of serial sagittal sections of 1 worm.)

The genus *Hypocaryophyllaeus* was created by Hunter (1927) to receive the type and only recorded species, *H. paratarius,* found in catostomid fishes in Illinois and Iowa.

The new species *gilae* may be readily separated from *paratarius* as follows:

H. paratarius Hunter, 1927	*H. gilae* n. sp.
1. Scolex wedge-shaped	1. Scolex uniform in shape
2. Scolex without terminal disc	2. Scolex with terminal disc
3. Loculi poorly defined	3. Loculi well-defined
4. Cirrus not protrusible	4. Cirrus protrusible
5. Vitellaria surround testes in an irregular annular ring.	5. Vitellaria around as well as between testes.
6. Uterine coils reach a maximum longitudinal extent of one-fourth or less that of testicular field.	6. Uterine coils reach a maximum longitudinal extent of one-half or less that of testicular field.
7. Eggs small, 0.026-0.032 x 0.018-0.021 mm.	7. Eggs large, 0.041-0.051 x 0.029-0.033 mm.
8. Hosts are catostomid fishes.	8. Host a cyprinid fish.

The genus *Hypocaryophyllaeus* as characterized by Wardle and McLeod (1952) needs slight emendation in order to receive *H. gilae.*

Hypocaryophyllaeus Hunter, 1927 char. emend.

DIAGNOSIS: Caryophyllaeidae with three pairs of shallow depressions (loculi) on the holdfast end (scolex). Cirrus opens independently on ventral surface or into a shallow genital atrium just anterior to the utero-vaginal canal opening. Ovary H-shaped. Uterine coils extend anteriorly beyond the cirrus sac.

TYPE: *H. paratarius* Hunter, 1927.

The generic diagnosis by Wardle and McLeod (1952) differs in stating that the shallow depressions on the holdfast are "poorly defined," and that the cirrus opens into a "non-eversible atrium." The presence in *gilae* of well-

defined depressions on the holdfast and a protrusible cirrus makes this emendation necessary.

In possessing a "II" type scolex *Hypocaryophyllaeus gilae* very closely resembles *Glaridacris laruei* (Lamont, 1921), and thus the two tapeworms could readily be confused. However, Hunter (1930), in commenting on the creation of the genus *Hypocaryophyllaeus,* stated that *H. paratarius* "cannot be classed as a member of the genus *Glaridacris* (or *Caryophyllaeus*) on account of the uterine coils . . . ." He was referring to the fact that in both *Glaridacris* and *Caryophyllaeus* the uterine coils *never* extend anteriorly beyond the cirrus sac, whereas they do in *Hypocaryophyllaeus.* Wardle and McLeod (1952) concurred in the above by using the relationship of the uterine coils to the cirrus sac in their diagnoses of the above and other genera in the family Caryophyllaeidae.

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A Comparison of the Pathogenicity and Course of Infection of Two Nematodes of Sheep, *Nematodirus spathiger* and *Trichostrongylus colubriformis,* in Pure and Mixed Infections

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The reports of Andrews (1939, 1944), Franklin *et al.* (1946), Gordon (1943, 1950), and the writers (1953), and other relevant literature summarized by these authors, show that although the parasitic diseases trichostrongylosis and nematodirosis, associated, respectively, with *Trichostrongylus* spp.* and *Nematodirus* spp.,* have much in common, they show certain dissimilarities. Heavy infections of young sheep are capable of causing an enteritis followed by diarrhea, anorexia, and other sequelae. However, trichostrongylosis appears generally to be more acute in all respects than nematodirosis. Of the aforementioned authors, Andrews, Franklin *et al.,* and Gordon reported deaths of lambs following administration of large, single or multiple, doses of *Trichostrongylus colubriformis* larvae, whereas the writers (*loc. cit.*) were unable to produce experimentally a fatal case of nematodirosis in lambs following administration of large doses of *Nematodirus spathiger* larvae. Clinical nematodirosis, when induced experimentally in lambs, invariably was followed by recovery of the affected animals, the only after effect of the infections being a retardation in growth.

It is an obvious fact that sheep on pasture are seldom, if ever, parasitized by pure infections of one parasite species. Parasitic disease in sheep usually

*The two common species parasitizing sheep are indicated in the title of this paper, but in some reports the species are identified only as *Nematodirus* spp. and *Trichostrongylus* spp.

results from the combined assault of several parasite species occurring in varying numbers, each contributing to the disease syndrome according to its proportionate numbers and pathogenic potential. Therefore, it is often difficult to evaluate the relative importance of the various degrees of infection of mixed species of internal parasites found in naturally infected sheep. At times, however, the diagnostic difficulties are lessened by the predominance of one or two species in mixed infections. As the pathogenic potentials of individual species become better known as a result of experimental work on pure infections, studies, such as the one reported herein, on the pathogenic interaction of species in mixed infections, should produce additional information useful in diagnosis and control.

Because under natural conditions mixed infections of young sheep with *N. spathiger* and *T. colubriformis* occur quite commonly, and information based on experiments of mixed infections is unavailable, an experiment was carried out to obtain (1) information on the effect on lambs of mixed infections of these two species and (2) supplemental information on their comparative pathogenicity.

EXPERIMENTAL PROCEDURE

Ten pure-bred, female, Shropshire lambs, two to three months old, raised parasite-free except for minor infections of coccidia and Strongyloides, were employed in this experiment. Five lambs were fed nematode larvae, as subsequently described, and five served as controls. Throughout the experiment the infected lambs as a group and the controls as a group were maintained in separate, concrete-floored pens, and rack-fed three pounds of good grade alfalfa hay per lamb per day; the uneaten portion of hay was removed and weighed daily before the next ration was given. For each lamb in the infected group there was a lamb of comparable weight in the control group.

Large enough doses of nematode larvae were administered to induce typical clinical effects, but were presumed to be insufficient to cause death of any of the lambs as a result of action of one or the other species alone. Single doses of 500,000 *N. spathiger* larvae, and/or 50,000 and 100,000 *T. colubriformis* larvae, were administered to the five lambs as indicated in table 1. Only one of the five lambs was fed *N. spathiger* larvae alone, as the writers (1953) had already obtained considerable information on the effect of similar, pure infections of this parasite on lambs.

TABLE 1.—Summary of larval doses, and selected data obtained from the experiment. (N—*N. spathiger*; T—*T. colubriformis*)

Lamb No.	Estimated Number of Larvae Fed (May 23)		Estimated Number of Worms at Autopsy (June 26)		Abnormal Stools* Duration in Days		Weight Gain (+) or Loss (—) Lbs. Infected Lambs	Excess Gain of Uninfected over Infected Lambs Lbs.
	N	T	N	T	Fluid	Mushy	May 23-June 26	May 23-June 26
1	500,000		5,600		3	6	+9.5	3.5
2	500,000	100,000	13,200	90,400	8	15	—2.0	10.5
3**	500,000	50,000	589,000	52,700	4	2	—3.5	9.5
4		100,000		92,000	2	13	+1.0	8.5
5		50,000		55,000	0	1	+7.5	4.5

*The five uninfected, control lambs had normal stools throughout the experiment.

**This lamb died on June 9, 17 days after administration of larvae; all data given for this lamb and its control for this period only.

Infected lambs were observed daily for clinical effects, and at weekly intervals they and their controls were weighed, fecal samples obtained for egg counts, and blood samples for haematological study. At the termination of the experiment, the infected lambs were autopsied, their parasites recovered, and the numbers estimated by dilution counts. The total counts of larvae administered and adults recovered at autopsy, which are recorded in table 1, were obtained from counts of duplicate samples, each count being subject to a variation of not more than ten percent. Egg counts (table 2) were done by the direct centrifugal flotation (DCF) method, saturated saline solution being used as the flotation medium.

RESULTS

The data in table 1 show that all five parasitized lambs developed some clinical signs of infection. They made poorer weight gains than their uninfected controls, and all but one developed an enteritis, as evidenced by mushy or fluid stools. There was, however, variation in the severity of the clinical effects in the different lambs. Lambs 2 and 3, which were fed larvae of both species of nematodes, were the most severely affected, both losing rather than gaining weight during the experiment. They also became diarrheic about 14 days after being dosed with larvae, which was several days before the bulk of their parasites reached the egg-laying stage. Lamb 3 died on the 17th day, after a four-day period of continuous diarrhea accompanied by progressive anorexia and weakness. Lamb 2 became very weak and was near death at the end of the third week of the experiment, after suffering from

TABLE 2.—Results of fecal egg counts eggs per gram) of infected lambs. (N—*N. spathiger;* T—*T. colubriformis*)

Days After Infection	Parasite Eggs	Infected Lambs				
		1 (N)	2 (N & T)	3 (N & T)	4 (T)	5 (T)
16	N					
	T		5		6	18
20	N		12			
	T		1,527	*	2,742	1,278
23	N		21			
	T		2,784		No Sample	48
27	N	45	0			
	T		5,373		4,143	3
30	N	213	15			
	T		672		4,200	285
34	N	52	6			
	T		3,642		3,537	930

*Last fecal examination at time of death, 17 days after infection, was negative for nematode eggs.

diarrhea for eight days; although it was very weak and listless, it appeared to be recovering slightly during the fifth week. Lamb 4, fed 100,000 *T. colubriformis* larvae only, was more severely affected than lambs 1 and 5, particularly in regard to weight, as it gained only one pound during the experimental period, or 8.5 pounds less than its control. Lamb 1, fed 500,000 *N. spathiger* larvae and lamb 5, fed 50,000 *T. colubriformis* larvae, were the least affected. However, both showed some anorexia and retardation in weight gain in comparison with their controls. Lamb 5 made slightly poorer weight gain than lamb 1, but did not develop diarrhea, whereas lamb 1 had diarrhea for three days. The fluid feces in the four lambs with diarrhea was consistently light brown in color and contained no blood.

Although marked clinical effects were observed in some of the infected lambs, including one death, haemoglobin and haematocrit values, as well as total leucocyte counts, were normal in all the lambs throughout the experiment.

The pathological changes observed at postmortem examination of lamb 3, which died, were similar to those reported by Andrews (1939) for fatal cases of experimental trichostrongylosis in lambs. This lamb was emaciated, and its small and large intestines contained only fluid material in which was suspended much cellular debris; although some food material was present in the rumen, it was evident that the lamb had eaten little for several days. Upon inspection, the duodenum and jejunum showed evidence of a catarrhal enteritis with some erosion of the mucosal surface. Sections from the jejunum showed some necrosis of the superficial surfaces of the villi and erosion of the epithelium; the intestinal lumen contained much cellular debris. Typical sections of the small intestine of this lamb are shown in fig. 1. Because this lamb prior to death had suffered from diarrhea and anorexia for several days, it is presumed that the changes noted resulted only indirectly from the activity of the parasites. It is apparent, however, that the induced parasitic infections in this lamb interrupted the normal physiological processes of the alimentary tract, particularly those of the small intestine, to such an extent that death ensued. No marked pathological effects of a similar nature were noted in the other lambs when autopsied, as they were recovering at this time from the acute phase of their infections. Lamb 2, however, was more emaciated than the other three surviving lambs.

The recovery of worms at autopsy (table 1) in relation to the number of larvae fed differed in regard to the two species studied, except in lamb 3, which died before its worm population reached maturity (see table 2). Lambs 3 and 5, fed 50,000 *T. colubriformis* larvae each, retained infections of this species at autopsy in numbers approximately equal, for all practical purposes, to the numbers of larvae fed, and lambs 2 and 4, fed 100,000 larvae of the same species, retained about 90 percent of the larvae fed. The fact that some of the estimated worm counts were slightly higher than the estimated numbers of larvae fed is not of much significance in the interpretation of the results, because of normal variation in the dilution counts. However, only in the case of lamb 3, which died 17 days after larvae were fed, did the *N. spathiger* worm count approximate the number of larvae fed, whereas in lambs 1 and 2, examined 34 days after infection, the *N. spathiger* worm count was only a small percentage of the number of larvae fed. Although the terminal retention of *N. spathiger* infections in these latter two lambs was small, evidence in table 1 shows that adequate infections were retained long enough

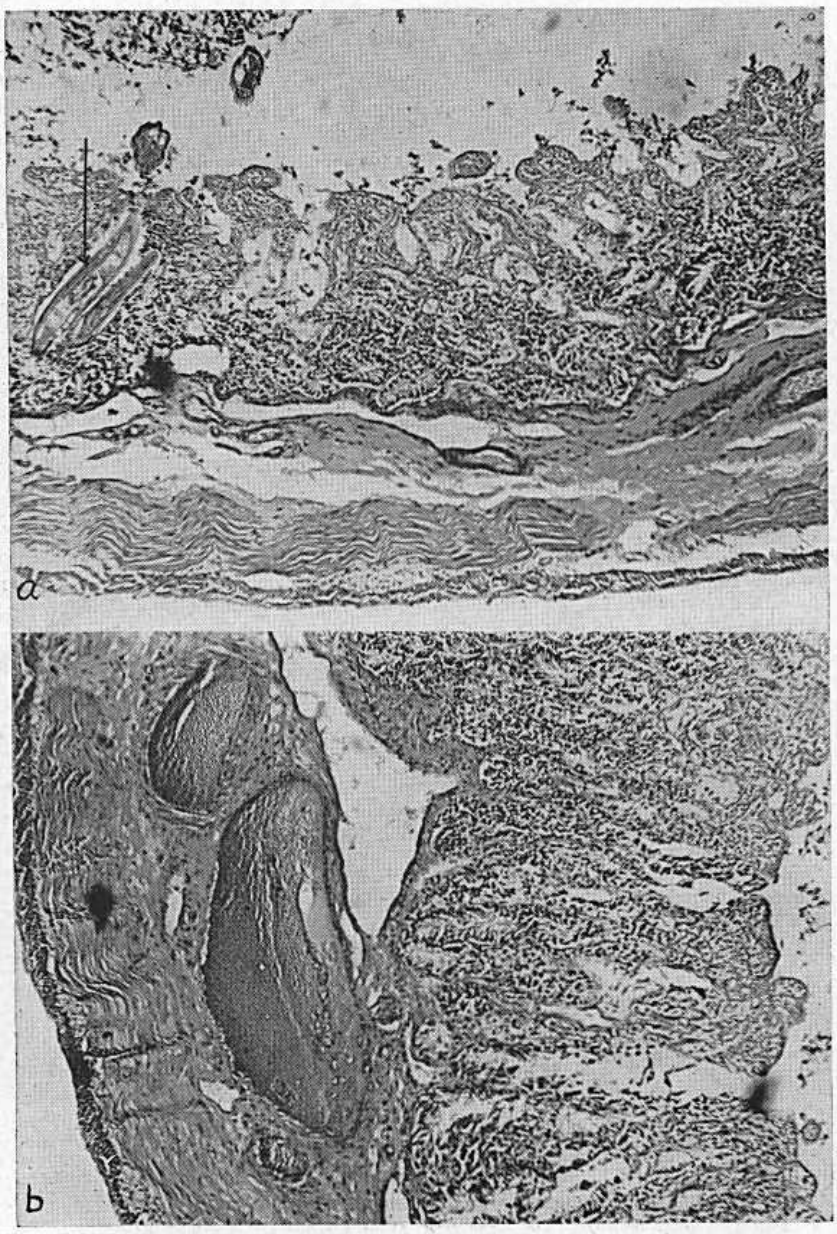

Fig. 1. Histological sections of the jejunum of lamb 3, which died 17 days after being fed larvae of *N. spathiger* and *T. colubriformis*. Note the superficial necrosis and erosion of the villi in (a) and (b), and the enlarged blood vessels of the submucosa in (b). Although on postmortem examination most of the parasites recovered were free in the lumen of the small intestine of this lamb, in (a) the anterior part of one nematode (species not determined) is shown penetrating deep into the partially destroyed mucosa.

to produce typical clinical effects; lamb 1 had fluid or mushy stools for 9 days and was retarded in growth compared to its control, and lamb 2 was more severely affected than lamb 4, which was fed an equal number of *T. colubriformis* only. These observations show that the induced *T. colubriformis* infections persisted longer than the *N. spathiger* infections, which has an important bearing on the pathogenicity and dissemination of these parasites.

Another difference in behavior of the two nematodes studied is shown by the egg counts (table 2). *N. spathiger* egg counts were small in lambs 1 and 2, although 5,600 and 13,600 worms were recovered at autopsy. In contrast, the *T. colubriformis* egg counts obtained from lambs 2, 4 and 5, which survived the experimental period, were relatively high. When these egg counts are compared with the postmortem worm counts in table 1, it may be concluded that most of the *N. spathiger* larvae fed to lambs 1 and 2 did not attain the egg-laying stage, and those worms of this species still retained in these lambs at autopsy, or shortly before, were producing eggs at a very low rate. In contrast, however, most of the *T. colubriformis* larvae fed were retained by the lambs during the course of the experiment, and in lambs 2, 4, and 5, which survived, the parasites were producing eggs at a relatively high rate compared to the *N. spathiger* infections in lambs 1 and 2. There is little indication from the data in table 2 that the presence of *N. spathiger* in the same lamb had any effect on the egg production of *T. colubriformis,* when the egg counts of lambs 2, 4, and 5 are compared, or that the reverse was true when egg counts of lambs 1 and 2 are compared. Although these data are admittedly limited in this respect, it does not appear that egg production of one species of parasite was influenced to any great extent by the presence of the other species in the same lamb.

DISCUSSION

On the basis of larval intake necessary to produce moderate to severe symptoms of enteritis, the evidence in the reports already cited and in the present report shows that *T. colubriformis* is more pathogenic than *N. spathiger* to lambs, and that severe uncomplicated, experimental trichostrongylosis may result in fatalities, whereas experimental nematodirosis is seldom fatal. Furthermore, the writers have shown that heavy infections of *N. spathiger* in lambs, which usually are not fatal, may be a contributory cause of death when substantial, but sublethal, infections of *T. colubriformis* are also present. It is evident, therefore, that species of these two genera, which live in the anterior portion of the small intestines of sheep, are threats to the health of their hosts whether they occur in almost pure or mixed infections.

An interesting problem is posed by the fact that Australian workers already cited induced experimentally fatal cases of trichostrongylosis in lambs by administration of larval doses only half, or less than half, as large as those employed by American workers with the same result. Lucker (personal communication) and the writers observed only minor effects on lambs fed approximately 50,000 *T. colubriformis* larvae, a larval dose reported by the Australian workers sufficient at times to produce some fatalities. At present it is not possible to account fully for the apparently greater susceptibility of the experimental lambs employed by the Australian workers. It is possible, however, that these variable results may have been due to differences in breed and/or size of the lambs employed. The Australian workers apparently em-

ployed small type Merino lambs in their trichostrongylosis experiments, whereas Andrews (1939) and Lucker (*loc. cit.*) employed Southdown-Hampshire cross-bred lambs, and the writers purebred Shropshire lambs.

The doses of larvae employed in this experiment may seem large in relation to the pathogenic effects produced in the experimental lambs. It should be noted, however, that our experimental lambs were necessarily maintained in pens under excellent sanitary conditions, had limited opportunity for exercise, were fed a good ration in quantity, and were not exposed to constant reinfection. Under these circumstances, it is believed that the clinical effects of the parasite burdens were not so marked as they may have been if the lambs had been maintained under less favorable nutritional and sanitary conditions.

That the experimental use of such large doses of larvae is justified, is shown by the published and unpublished record to the effect that an intake by lambs on pasture of 50,000 to 100,000 *T. colubriformis* larvae over a period of time is not uncommon, and an intake of 500,000 *N. spathiger* larvae not impossible. Another important point to consider in this respect, which is illustrated to some extent by the data reported herein, is that postmortem worm counts seldom reflect more than a small proportion of the total larval assault on sheep. A recent demonstration of this is found in the unique study of Tetley (1949), which showed that during a period of six months over 18,000 nematodes of various species were recovered from the feces of a lamb, which, upon postmortem examination at the end of the study period, retained only a little over 7,000 parasites. Because of obvious difficulties in the recovery of all worms passed in the feces, especially small species, immature specimens, and those distintegrated beyond recognition, Tetley's total recovery of worms from the manure of this lamb was minimal. If this ratio of 3 or more to 1 of worms expelled to worms recovered postmortem can be generally applied, then a lamb having about 20,000 worms at autopsy, a not uncommon occurrence, could have been exposed to over 100,000 larvae over a period of a few months or less. Adding to this reasonable assumption the known fact that immature nematodes are often more pathogenic than adults, it is reasonable to assume that the doses of larvae employed in this experiment, and in others reported in the literature, are not excessive compared to possible larval exposure of lambs on pasture, and serve the intended experimental purposes very well.

SUMMARY

A small-scale experiment was carried out in which sublethal doses of larvae of *N. spathiger* and/or *T. colubriformis* were administered to purebred, parasite-free, Shropshire lambs. The results of this experiment confirm and extend those previously reported by other workers and the writers, to the effect that *T. colubriformis* is a more pathogenic and more persistent trichostrongylid parasite of sheep than *N. spathiger* and demonstrate that the two species in mixed infections apparently have additive pathogenic effects.

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On the Migratory Behavior of the Larvae of *Neoascaris vitulorum* (Goeze, 1782) Travassos, 1927 in White Mice

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Sprent (1952) reported that various species of ascaris which he investigated could be grouped into two main categories, tracheal or somatic, depending on the type of larval migration following the ingestion of embryonated eggs. Due to an insufficient number of eggs of *Neoascaris vitulorum*, the large roundworm of cattle, Sprent was unable to include this species in his study. When the present writer obtained a supply of eggs of this parasite, it was considered desirable, in the light of Sprent's work, to investigate the migratory behavior of the larvae of *N. vitulorum* in experimentally infected white mice.

MATERIALS AND METHODS

Eggs used in this study were recovered from fecal material collected from young calves at the Iberia Livestock Experiment Station, Jeanerette, Louisiana. The eggs were cultured in tap water at 28° C. for 30 days. White mice fed 5,000 embryonated eggs succumbed to pneumonia within four to six days after ingesting the eggs. Consequently, 3,000 eggs mixed with a little powdered meal was fed to each of eight male, five-week-old mice. One mouse was killed daily 1, 2, 3, 4, 6, 8, 12 and 21 days after infection. Each mouse was examined in the manner described by Sprent (loc. cit.) with two modifications: (1) In view of the fact that Schwartz (1922) found larvae in the kidneys as well as the livers and lungs of guinea pigs which had been fed *N. vitulorum* eggs, kidneys of the mice were removed and examined in the same manner as liver and lungs, and (2) the carcass was washed prior to blendorization and digestion, and the washings were then examined for larvae.

In addition, embryonated eggs were fed to four pregnant mice to determine whether the larvae could reach the fetuses. A single dose of 3,000 eggs was fed to each of three mice which were in the first, second and third weeks of pregnancy, respectively. The fourth mouse was fed a total of 6,000 eggs in three equal, weekly doses. At parturition, the entire litter of each mouse was blendorized and digested, and the material was then examined for larvae.

RESULTS

Figure 1, showing number of larvae recovered from mice at various intervals after infection, indicates that the larvae passed from the intestinal tract to the liver, lungs and kidneys one day after infection. Nearly all larvae had reached the liver by the fourth day and they appeared in great numbers in the lungs from the fourth to the sixth day. Lungs of mice killed four and six days after ingestion of eggs showed severe congestion and extensive petechiation. After 12 days, a few larvae were still in the lungs, but all had disappeared from the liver. Larvae were found in the kidneys during the first six days of infection and larvae also appeared in the intestinal wall and intestinal contents from the second to the eighth day. No larvae were recovered from the carcass as observed in the cases of *A. columnaris, A. mustelarum, T. leonina,* and *T. transfuga* (Sprent, 1952). However, larvae were found in the washings of the carcass from the third to the twelfth day after infection. Twenty-one days following exposure to the embryonated eggs all larvae had disappeared, indicating that they had either been destroyed or had escaped to the outside.

No larvae were recovered from the litters of the four mice fed eggs during pregnancy.

DISCUSSION

Sprent (loc. cit.) showed that the larvae of *Parascaris equorum* and of the human and porcine strains of *Ascaris lumbricoides* displayed a tracheal type of migratory behavior essentially similar to that manifested by the larvae in

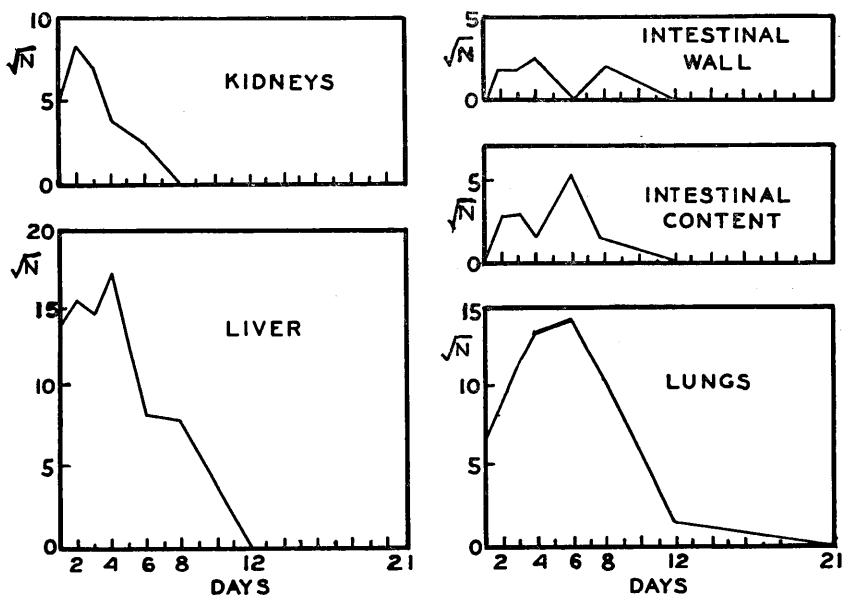

Fig. 1. Showing the number ($\sqrt{N}$) of larvae of *N. vitulorum* recovered from various locations in mice on various days after infection.

their respective definitive hosts which have been shown to become infected in nature by the direct ingestion of embryonated eggs. The present investigation shows that the migratory behavior of *N. vitulorum* larvae in white mice closely resembles that of *P. equorum* and *A. lumbricoides,* indicating that infection of the bovine may also follow direct ingestion of embryonated *N. vitulorum* eggs.

It has been suggested (Griffiths, 1922; Boulenger, 1922; Schwartz, 1925) that calves may become prenatally infected with this parasite. Failure of the larvae of *N. vitulorum* to reach the mouse fetus when embryonated eggs were fed to pregnant mice does not negate nor detract from this hypothesis as it must be borne in mind that these trials involved an abnormal host.

SUMMARY

Mice were infected by feeding them embryonated eggs of *Neoascaris vitulorum*. The larvae manifested a tracheal type of migration similar to that of *Parascaris equorum* and *Ascaris lumbricoides*. Attempts to infect mice prenatally failed.

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Report of the Brayton H. Ransom Memorial Trust Fund

FUNDS ON HAND, Jan. 1, 1952	$1737.72
RECEIPTS: Interest received in 1952	59.64
DISBURSEMENTS: Expenses and grant to Helminthological Society of Washington	46.00
BALANCE ON HAND, Dec. 31, 1952	1751.36

ELOISE B. CRAM,
Secretary-Treasurer

Observations on the Maturation of Gametocytes of *Leucocytozoon simondi**

JUNE RAWLEY**

The gametocytes of *Leucocytozoon simondi* are known to occur in two shapes in the dried stained preparations, the characteristic elongate forms in fusiform cells and the less common large round forms in round host cells. The differences in shape might be accounted for in terms of the type of host cell invaded (Fallis et al, 1951). According to Huff (1942), Mathis and Leger thought that the forms in round host cells had entered "mononuclear leucocytes" while those "species with gametocytes in fusiform cells preferred erythroblasts or erythrocytes." On the other hand, Huff concludes that all gametocytes develop in lymphocytes or monocytes and seems to imply that the round forms are immature and only the elongate forms in fusiform cells are mature. Cook (1953) was able to demonstrate developing gametocytes only in the red blood cell series in the peripheral blood. Fallis et al (1951) reported exflagellation of both round and elongate microgametocytes of *L. simondi.*

In view of the conflicting reports, the activity of these gametocytes was studied in wet mounts. At the time wet mounts were prepared dried giemsa-stained smears were made for comparison. The wet mounts were made from birds in which the dried giemsa-stained slides (1) showed only the large round gametocytes and (2) those which showed both round and the more characteristic elongate forms in fusiform host cells. Wet mounts were prepared by placing a drop of freshly drawn blood in 0.85% NaCl on a slide and protecting it with a vaseline sealed cover glass.

Although the blood was handled as rapidly as possible, some of the elongate gametocytes had become rounded by the time the preparation could be observed under the microscope. The newly rounded macrogametocytes can be readily distinguished from the originally round forms. The newly rounded forms are smaller and may be seen in fusiform cells. Even if the host cell has secondarily become rounded there is a large clear unstained area between the parasite and the host cell membrane in contrast to the very narrow, often indiscernible, band of host cell cytoplasm around the originally round parasite. In a number of cases the rapid rounding of these elongate macrogametocytes was observed. The sequence of events is a change first in the parasite and later on in the host cell. A rounding of the spindle shaped ends of the parasite occurs. This is accompanied by activation of the cytoplasmic granules, which for the most part tend to concentrate at each end of the elongate cell. Very few forms were observed which did not have this clumping of the granules. The gametocyte then in a swift steady movement changes from the elongate form into a rounded one. The host cell has been inactive up to this time. After the parasite has completed its phase of rounding, the nucleus and extended portions of the cytoplasm of the hose cell become rounded. The nucleus of the host cell becomes a compact circular object adjacent to the parasite nucleus. The entire process occurs in less than a minute but not all of them begin to round at the same time. However, not all the

*From the University of Michigan Biological Station. This work was supported by a grant from the Joseph Henry Fund of the National Academy of Sciences to Dr. G. F. Otto.

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elongate forms became round; some remained elongate and apparently failed to undergo maturation. After the host cell has become rounded, the rounded macrogametocyte continues to show activity. The cytoplasmic granules, which were evenly distributed, were seen to follow one of two courses. In one sequence, the entire rounded macrogametocyte became vacuolated and the active granules were scattered around the vacuoles; granular activity in these forms ceased in about eighteen minutes. In other cases, the granules were observed to clump together in four or five places and remain active in these clumps. The entire process occurred while the parasite was either within the host cell at at least associated with the host cell membrane.

The process was not followed in detail in the normally round macrogametocytes but the activity of cytoplasmic granules suggest that it was essentially the same except that maturation was not accompanied by any basic change in shape.

In the case of the microgametocyte, there is even greater activity apparent on the part of both the parasite and the host cell than in the case of the macrogametocyte. The process was observed in both the normally round and the normally elongate forms.

In blood in which only round gametocytes were found, the parasitized cells show a quivering action, as the exflagellation process occurs and the microgametes were released while the round microgametocyte was still within the round host cell. In another instance a round parasite in a round cell was seen to elongate into a fusiform shape after which exflagellation began. Thereafter the parasite again rounded while still containing unescaped microgametes. The gametocyte elongated a second time and again became rounded whereupon exflagellation was resumed. This was not merely an optical distortion due to the turning of the parasitized cell but the actual movement of the parasite was seen.

The fusiform microgametocytes usually become rounded and thereafter undergo exflagellation. However, in one case a fusiform microgametocyte was seen to exflagellate while still elongate and then became round after release of the microgametes.

Fertilization was not observed even though microgametes migrated to the surface of the rounded macrogametes and were active at the surface of these forms for some time.

SUMMARY AND CONCLUSIONS

Apparently both the large round and large fusiform gametocytes of *L. simondi* seen in the dried giemsa-stained blood smears are mature. Some evidence of the maturation process of both the round and elongate macrogametocytes was seen as well as maturation and exflagellation of both the round and elongate microgametocytes.

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Harley Jones Van Cleave
1886-1953

Harley Jones Van Cleave, research professor of zoology and parasitology at the University of Illinois, died suddenly January 2, 1953. Although he had been in poor health for several years, his sudden demise came as a shock to his colleagues and friends.

Dr. Van Cleave was born in Knoxville, Illinois, October 5, 1886. He received his B. S. degree at Knox College in 1909, and the M. S. and Ph. D. degrees from the University of Illinois in 1910 and 1913, respectively.

From 1913 until his death, Dr. Van Cleave served, in different capacities, as a member of the faculty of Zoology at the University of Illinois. He was made Research Professor in 1949, a position which relieved him of teaching and administrative duties and permitted him to devote his entire time to research.

Dr. Van Cleave was a member of many scinetific societies, including the American Association for the Advancement of Science, American Society of Naturalists, American Society of Parasitologists (Pres. 1947), American Society of Zoologists, Ecological Society of America, American Microscopical Society (Pres. 1931), and the Illinois Academy of Sciences (Pres. 1928). He was elected a corresponding member of the Helminthological Society of Washington in 1922 and became an active member in 1947.

Dr. Van Cleave was a tireless worker and made many important contributions to parasitology. His specialty was the Acanthocephala and he was one of the world's outstanding authorities on this group of invertebrate animals. His publications in the field of parasitology and related subjects comprised about 150 papers, the majority of which dealt with his specialty.

In his passing, parasitology has lost a distinguished devotee, and his fellow workers, a friend. The Helminthological Society extends to his family its sincerest expression of sympathy.

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Minutes

Three Hundred Ninth to Three Hundred Sixteenth Meetings

309th meeting: McMahon Hall, Catholic University of America, October 15, 1952. THE FOLLOWING PAPERS WERE PRESENTED: Effect of antibiotics on the mites of pigs by Shorb, A review of nematology in the past twenty years by B. Chitwood, Methods of screening drugs for their effect against multiple helminthic infections in mice by Tiner, Parasitic castration by Reinhard, Possible correlation of trichomonads in the nasal passage and dystrophic rhinitis of swine by Spindler, Observations on the infective rate of the F_2 generation of *Australorbis glabratus,* from a cross of one parent from a refractory Brazilian strain with one from a susceptible Puerto Rican strain by Newton.

310th meeting: Library of the Zoological Division of the B.A.I., Beltsville, Md., November 21, 1952. PAPERS PRESENTED: *Haemoproteus* in Anatidae by Herman, Parasites of Canada geese by Wehr, and Viability studies on infective larvae of *Nematodirus spathiger* by Turner.

311th meeting: McMahon Hall, Catholic University of America, December 17, 1952. The following recommendations of the Executive Committee were ratified: Ten dollars ($10.00) be given to Science Fair. The price of volumes of the Proceedings be revised to three dollars ($3.00) domestic and three and a quarter dollars ($3.25) foreign. Officers elected for the year 1953 were Dr. E. G. Reinhard, President; Dr. Paul Weinstein, Vice President; Miss Edna Buhrer, Corresponding Secretary and Treasurer; Dr. C. G. Durbin, Recording Secretary. PAPERS PRESENTED: Treatment of lungworms of sheep and goats by Durbin, Collecting trypanosomes from the stomach of a foetal calf by Dikmans, A general discussion of toxoplasma infection in man and animals by Jacobs.

312th meeting: McMahon Hall, Catholic University of America, January 21, 1953. Dr. Lutermoser was elected member-at-large of the Executive Committee. (Dr. von Brand's term expired). Dr. Spindler was re-elected as representative of the Society to the Washington Academy of Science. PAPERS PRESENTED: A film with comments on epidemiology and control of *Ascaris* by McMullen, On the use of artificially altered oocysts of *E. tenella* to establish immunity to cecal coccidiosis in chickens by Uricchio, Observations on the life history of *Trichinella spiralis* by Mengoli, Unsuccessful attempts to establish *Eustronglides* in the black crowned night heron by Elsea.

313th meeting: Naval Medical Research Institute, Naval Medical Center, Bethesda, Md., February 18, 1953. A motion that an obituary for Dr. Van Cleave be published in a forthcoming issue of the Proceedings was carried. PAPERS PRESENTED: Recent studies on *Oncomelania* sp. the intermediate host of *S. japonicum* by Hubendick, On the mating behavior of mosquitoes by Stahler, On the role of the stomach wall in the exogenous malarial cycle by Weathersby, On malarial infected mosquitoes by Terzian, On the growth of galls in root knot infections by Dropkin.

314th meeting: National Institutes of Health, Bethesda, Md., March 18, 1953. PAPERS PRESENTED: On experimental chemotherapy of Schistosomiasis by Luttermoser, Helmenthic disease of West Africa by Berry, Effects of dry season on *Australorbis glabratus* by Barbosa, Respiratory metabolism of *Balantidium coli* by Agosin, Some interactions in vitro of *Endamoeba histolytica* and single species of microbial symbionts by Rees.

315th meeting: School of Hygiene and Public Health, Johns Hopkins University, Baltimore, Md., April 17, 1953. Dr. Steiner was elected repre-

sentative of the Society at the International Congress of Zoology to be held in Copenhagen, Denmark during the summer of 1953. Dr. Spindler read a telegram which the Washington Academy of Science sent to President Eisenhower protesting the dismissal of the Director of the National Bureau of Standards. PAPER PRESENTED: Gametocytes of *Leucocytozoon* by Cook, Morphological variations of larvae of the scutellaris group of *Aedes* in Polynesia by Rosen, Chemoreceptors in oviposition of mosquitoes by Wallis, Genetics of autogeny in the *Culex pipiens* complex by Spielman, Antigenic activity of secretions and excretions of trichina larvae by Chute, Absorption of protective antibodies from anti-*Nippostrongylus* serum by Thorson.

316th meeting: Log Lodge—Cafeteria, Agricultural Research Center, Beltsville, Maryland, May 16, 1953.

THE FOLLOWING WERE ELECTED TO MEMBERSHIP DURING THE YEAR: 309th meeting George E. Cauthen, Jacob H. Fischthal, James K. Gilford; 310th meeting Thomas W. Graham, Robert B. Short, David J. Doran, Francis Huber; 311th meeting Winfield H. Hart, Quintin L. Holdeman, Ralph W. Macy, Donald B. McMullen, Richard N. Rossan, Joseph N. Sasser, R. T. Young; 313th meeting Bryce C. Walton, Robert E. Freytag; 314th meeting Morgan Golden, Eldon J. Cairns, A. F. Schindler, George Swank, Jr., W. B. Mountain, Daniel O. Betz; 315th meeting M. S. Briscoe, William H. Coil, Leslie C. Costello, John T. Maloney, L. D. Newsom, Morris Schneider, Conrad E. Yunker; 316th meeting Virginia G. Blankemeyer, Emmanuel Franco, Lloyd A. Lider; G. Dikmans was elected a Life Member.

C. G. DURBIN
Recording Secretary

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ERRATA

In the paper by Tarjan and Sasser, 1953, Known and Suspected Plant-parasitic Nematodes of Rhode Island, I, 20(1) :49-54, the explanation and identification for figures 1 and 2 (pages 51 and 53) were reversed. *Discomyctus brevicaudatus* is illustrated on page 51 and the legend, fig. 2, etc., on page 53 applies to it. *Longidorella parva* is illustrated on page 53 and the legend, fig. 1, etc., on page 51 applies to it.

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