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PROCEEDINGS OF THE HELMINTHOLOGICAL SOCIETY OF WASHINGTON

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Control of cattle-parasitic and free-living nematodes by soil fumigation with methyl bromide. LEONARD E. SWANSON, University of Florida Experiment Station, Gainesville, Florida, and A. L. TAYLOR, U. S. Bureau of Plant Industry, Tifton, Georgia.

The extraordinary efficacy of methyl bromide when used as a soil fumigant against the root-knot nematode, *Heterodera marioni* (Cornu) Goodey, as reported by Taylor and McBeth (1940, Proc. Helm. Soc. Wash. 7(2): 94–96, and 1941, Ibid. 8(1): 26–28) and Gingrich and Haenseler (1941, Ibid. 8(2): 50–53) led to the consideration of this chemical when it was desired to eliminate free-living nematodes and the larvae of nematode parasites of cattle from some plots to be used for experimental work. Since no information as to its effect on these forms was available, a preliminary experiment was performed to test its action against the stages of these nematodes usually found in soil.

MATERIALS AND METHODS

Three similar plots of soil, each 4 by 5 feet, were isolated from the surrounding area by tight wooden frames extending 10 inches below and 2 inches above the soil surface. The soil so enclosed was a light sandy loam, moderately moist and rather loose at the time of fumigation. Soil temperature was about 25° C. The plot soil was examined by the Cobb sifting and gravity method and found to contain about 20,000 free-living nematodes to the cubic foot, most of which were species of the genera *Hoplolaimus, Pratylenchus, Mononchus,* and *Criconemoides.* In addition, cattle-parasitic and free-living nematodes were placed in the plots. These additions came from two sources as follows:

1. From cultures of calf feces. The genera represented were *Haemonchus, Ostertagia, Trichostrongylus, Oesophagostomum, Cooperia, Bunostomum,* and *Nematodirus.* They were extracted from the culture by the Baermann method and placed with a small quantity of wet sphagnum moss in wide mouth glass bottles of about 60 cc capacity.

2. From pasture grass. These additions were extracted by the Baermann method from grass cut from a pasture regularly used for nematode-infected cattle. The extracts contained representatives of the cattle-parasitic genera mentioned above and in addition, free-living nematodes of the genera *Dorylaimus, Cephalobus, Rhabditis,* and *Acrobeles.* These were also placed in small bottles with sphagnum moss.

Further portions of the same lot of grass from which the nematodes mentioned in the preceding paragraph were extracted were placed in cylindrical glass tubes 6 to 8 inches long and 1¼ inches in diameter and in several wire cages about 4 inches on each side.

All of the above mentioned material was examined and found to contain numerous living nematodes, with a minimum of approximately 200 nematodes to each bottle, tube or cage. Openings of the containers were covered with cheese cloth and all material was kept cool and moist until it was placed in the plots.

Twenty-six of the containers of prepared samples were placed in each of the 3 experimental plots. Of these, 10 came from feces, 7 from grass extracts and

TABLE 1.—*Condition of nematodes from plots fumigated with methyl bromide and from control plot*

Origin of sample	Plot No. 1 (Fumigated)				Plot No. 2 (Fumigated)				Plot No. 3 (Control)			
	Depth of sample				Depth of sample				Depth of sample			
	Surface	2 in.	6 in.	12 in.	Surface	2 in.	6 in.	12 in.	Surface	2 in.	6 in.	12 in.
Grass[b]	Dead				Dead				Alive[a]			
Feces	Dead				Dead				Alive[a]			
Feces		Dead				Dead				Alive		
Feces		Dead				Dead				Alive		
Feces		Dead				Dead				Alive		
Grass[b]		Dead				Dead				Alive		
Grass[b]		Dead				Dead				Alive		
Grass		Dead				Dead				Alive		
Grass		Dead				Dead				Alive		
Grass		Dead				Dead				Alive		
Feces			Dead				Dead				Alive	
Feces			Dead				Dead				Alive	
Feces			Dead				Dead				Alive	
Grass[b]			Dead				Dead				Alive	
Grass[b]			Dead				Dead				Alive	
Grass			Dead				Dead				Alive	
Grass			Dead				Dead				Alive	
Grass			Dead				Dead				Alive	
Feces				Dead				Dead				Alive
Feces				Dead				Dead				Alive
Feces				Dead				Dead				Alive
Grass[b]				Dead				Dead				Alive
Grass[b]				Dead				Dead				Alive
Grass				Dead				Dead				Alive
Grass				Dead				Dead				Alive
Grass				Dead				Dead				Alive
Soil			Dead	Dead			Dead	Dead			Alive	Alive
Soil			Dead	Dead			Dead	Dead			Alive	Alive
Soil			Dead	Dead			Dead	Dead			Alive	Alive
Soil			Dead	Dead			Dead	Dead			Alive	Alive

[a] 50% of the nematodes dead. Water in bottle warm from heat of sun.
[b] Nematodes extracted from grass by Baermann method.

9 were pasture grass. One feces and one grass extract sample was placed on the surface of each plot and the remainder buried at random locations 2 inches, 6 inches, or 12 inches deep in the plot soil, with the samples of each origin evenly divided between the 3 depths.

Two adjacent plots with a 2½-foot alley between them were then covered with a single sheet of glue-coated kraft paper large enough that about 1 foot of paper on each edge could be buried in the soil outside the plots. The remaining control plot was covered similarly. Covers were carefully checked to make sure that they were as gas tight as possible.

A rubber hose was pushed through a small hole in the cover over the 2 plots and attached to a methyl bromide applicator. Fumigation was accomplished in less than 1 minute by merely placing a 1-pound can of methyl bromide in the applicator, and discharging the full contents into the space between the paper and the soil surface. The tube was withdrawn and the hole sealed with a large piece of glued paper. The single plot was designated as the control and received no methyl bromide.

Twenty-two hours after the methyl bromide was applied, the covers were removed from the plots and the samples again examined. Nematodes from the feces and grass extract samples were washed out with a small quantity of water and concentrated by centrifuging. Pasture grass samples were placed in the Baermann apparatus. Soil samples were collected from all the plots at the 6- and 12-inch levels, washed, and examined. All nematodes found were examined microscopically. Lots of material containing moving nematodes were counted as alive, those containing no moving nematodes were counted as dead. Some of the samples were reexamined after 24 hours to check on possible revival of the apparently dead nematodes.

The results of this experiment are given in detail in table 1. Living nematodes (free-living and parasitic) were numerous in all samples from the control plot. No free-living or parasitic nematodes were found alive in the 2 treated plots. Actively moving Protozoa were often observed in the samples from the control plot, but none were seen in the samples from the fumigated plots.

DISCUSSIONS AND CONCLUSIONS

It is apparent that, under the conditions of the experiment, methyl bromide fumigation is extremely effective, at least against adults and larvae of the free-living forms and larvae of the parasitic forms. Eggs were not included in the experiment.[1]

The area fumigated was 63 square feet and the amount of methyl bromide used was 1 pound. Cost of treatment for the chemical alone would be approximately 1¼ cents per square foot of soil at the present price of methyl bromide, which is about 75 cents per pound in small lots. The method of application is simple and easy. Much larger areas can easily be fumigated under a single cover or by treating successive sections. Other gas impervious covers than glue-coated kraft paper may be used.

If further experiments corroborate the results of the present one, a number of uses for methyl bromide might be found in experimental or commercial work. While the cost is rather high, the expenditure would often be justified where valuable animals are kept on small plots of soil.

SUMMARY

Fumigation of the soil with methyl bromide under a paper cover killed all of the cattle-parasitic and free-living nematodes to a depth of at least 12 inches. Rate of application was one pound of the chemical to 63 square feet of soil.

[1] A second experiment including eggs is in progress and will be reported later.

Fumigation of soil with methyl bromide as a means of destroying infective stages and intermediate hosts of some internal parasites of mammals.

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INTRODUCTION

Methyl bromide[1] is used chiefly as a refrigerant, as a fire extinguisher, and as an insecticide. When employed as an insecticide, it is generally used to destroy insect pests at quarantine stations, in grain elevators, and in fur storage vaults where fumigation can be carried out in closed gas-tight chambers.

In 1940, Livingstone reported that methyl bromide fumigation of bulk soil containing the larvae of the white fringed beetle, *Pantomorus leucoloma* (Boh.), would completely control this insect. In the same year, Taylor and McBeth demonstrated that methyl bromide was effective as a soil nematocide for use against the root-knot nematode, *Heterodera marioni* (Cornu) Goodey, and the innumerable species of free-living nematodes constantly present in the soil. Their investigations showed that 1½ pounds of methyl bromide released under a gas-tight cover placed over the soil to be treated would completely control both root-knot and free-living nematodes in 200 square feet of sandy loam soil in the field. In the summer of 1942, Swanson and Taylor, using the same technique, were able to destroy all the free-living nematodes as well as the infective larvae of nematodes parasitic in cattle which were present in soil and on grass, by fumigation with methyl bromide. The results of these experiments suggested that this chemical might be useful in destroying the preparasitic stages of parasites of other animals and their intermediate hosts in infested soil. The present paper presents the results of an experiment to test this possibility.

PROCEDURE AND RESULTS

Swine feces containing eggs of *Ascaris lumbricoides* Linn., *Strongyloides ransomi* Schwartz and Alicata, *Oesophagostomum* spp. and oöcysts of coccidia, were mixed with animal charcoal and cultured at room temperature in large covered glass dishes. The cultures were made at intervals so that at the beginning of the experiment developmental stages of these parasites aged 0, 4, 7, 11, 18, 25 and 28 days would be available. On the day before the experimental plots were fumigated, kidney worm eggs were obtained from female specimens of *Stephanurus dentatus* Dies. secured at the local slaughter house, and specimens of dung beetles and earthworms, the intermediate hosts of the spirurid stomach worms and lungworms, resepectively, were collected, so that the experiment might be made as complete as possible. (Specimens of the white grub, the intermediate host of the thorn-headed worm, were not available.) Material from swine was used in this experiment because of availability and because infective stages of some swine parasites are among the most resistant to chemical agents.

Four plots, each 8 feet square and separated by alleyways 4 feet wide, were marked off on newly plowed sandy loam soil. Representative samples of the fecal cultures, of the kidney worm eggs, and of the dung beetles and earthworms, were placed in glass containers closed by pieces of gauze held in place by rubber bands. The containers were placed in random positions in all the plots at 2, 6 and 12 inches below the soil surface and also on the surface. Distribution of the material

[1] Methyl bromide (bromomethane, CH_3Br) is a liquid which boils at 4.5° C. at atmospheric pressure and is, therefore, a gas at ordinary temperatures. It is obtainable commercially in one-pound cans to which inexpensive applicators can be attached. The applicators make it possible to discharge the contents of the can in gaseous form quickly, easily and safely. Methyl bromide is poisonous and extreme care should be taken not to breathe the gas.

was so arranged that a sample from each culture and a sample from each of the other sources was at each level in each of the four plots. After the containers had been placed, a ditch 1 foot deep was dug around each plot. A gas-tight cover of glue-coated kraft paper was placed over each plot and its edges buried in the ditch. Immediately afterwards, the contents of a 1-pound can of methyl bromide was released into the air space beneath the cover of plots 1, 2 and 4, by means of a methyl bromide applicator attached to a rubber tube inserted through a small hole in the paper cover. The hole in the paper was immediately sealed with a gummed paper patch. Plot No. 3 was left untreated as a control. After about 20 hours, the covers were removed, the containers recovered and their contents examined macroscopically and microscopically for living organisms. The cultures of unembryonated ascarid eggs were kept until sufficient time had elapsed for development to take place. Lack of movement, absence of further development on continued observation, and general state of decomposition were criteria used to determine whether the organisms were killed by the gas.

No evidence of life was found in the cultures or organisms on any of the fumigated plots, while the larger portion of those from the unfumigated control plot were viable.

DISCUSSION AND CONCLUSIONS

The results obtained in the present experiment are most significant in connection with the destruction of the embryos in the eggs of the large roundworm, *Ascaris lumbricoides*. These eggs are very resistant to ordinary chemicals and may remain viable in soil for several years. The killing of earthworms, the intermediate hosts of the swine lungworm, also appears significant since infected earthworms may survive in soil for as long as 4 years. The destruction of infective larvae of *Strongyloides* and *Oesophagostomum*, as well as immature larvae within the kidney-worm eggs, suggests that the infective larvae of strongylid parasites of other animals may also be killed by methyl bromide. Destruction of coccidia indicates that methyl bromide may also be useful for control of protozoan parasites.

At the present price of methyl bromide in small quantities (about 75¢ per pound), cost of fumigating at the rate used in this experiment (1 pound to 64 square feet) would be 1.17¢ per square foot for the chemical alone. No attempt was made to ascertain the minimum quantity necessary to produce a complete kill, and it is possible that a smaller amount would have been equally effective.

It is apparent that methyl bromide when used as a soil fumigant is highly effective against many of the forms of animal parasites found in soil, and further tests of its efficacy are suggested, since several possibilities for practical use are apparent. While the trouble and expense would not be justified under ordinary circumstances, fumigation might be used to advantage where small areas are involved. This is especially true where valuable breeding stock or small animals, such as foxes and mink, are raised or valuable dogs are kept in restricted areas. In such cases the cost of soil fumigation would often be only a small fraction of the value of the animals or the investment in permanent runs. Methyl bromide fumigation might be useful in the control of coccidiosis in poultry and as a means of sterilization of animal cages in zoos, menageries and laboratories, where this gas would have a considerable advantage over steam or hot water because it penetrates cracks and crevices without loss of efficiency. In control of human parasites, fumigation with methyl bromide might occasionally be found useful to clean up small centers of infection.

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The occurrence of *Viannaia viannai* Travassos (Nematoda: Heligmosomidae) in opossums in North America. G. DIKMANS, U. S. Bureau of Animal Industry.

Travassos (1937, Revisão da Familia Trichostrongylidae Leiper, 1912) lists four species in the genus *Viannaia,* namely, *V. viannai, V. conspicua, V. hamata* and *V. pusilla.* These are all parasites of different species of opossum and all are reported from South America. Recently the writer had an opportunity to examine the viscera of two opossums, *Didelphys virginiana,* found dead along a road leading to the U. S. Department of Agriculture Beltsville Research Center, Beltsville, Maryland. The small intestines of both of these animals contained, among other helminths, two species of nematodes, one being identified as *Longistriata didelphis* (Travassos, 1914) and the other as *Viannaia viannai* Travassos, 1914. Since, so far as the writer has been able to determine, no member of the genus *Viannaia* has previously been reported as a parasite of the opossum in North America, and since no description of the genus and its members is available in North American parasitological literature, a brief description of the genus and of its type species, together with some figures illustrating the salient morphological characters of this nematode, are presented herewith.

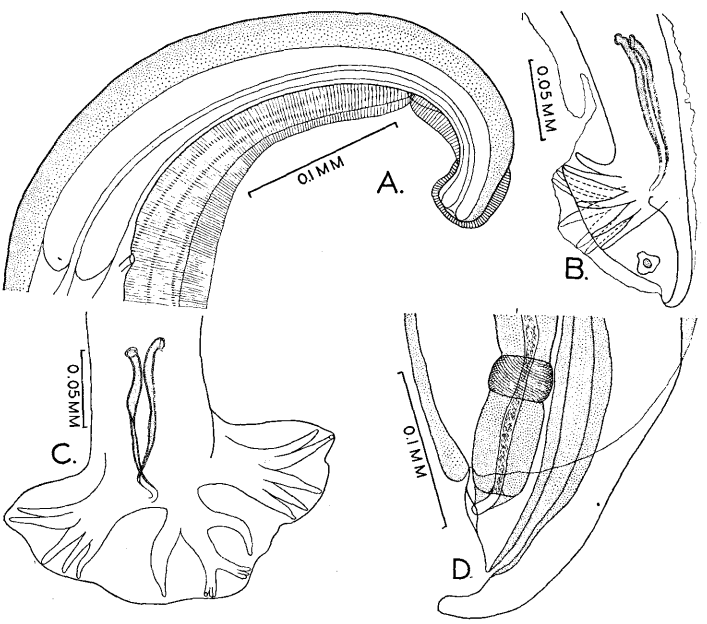

FIG. 1. *Viannaia viannai.* A—Anterior portion of body. B—Terminal portion of male, lateral view. C—Bursa and spicules. D—Terminal portion of female.

The description of the genus and species are taken in part from Travassos (1921, Mem. Inst. Oswaldo Cruz. 13(1): 5–135; 1937, *loc. cit.*).

Genus *Viannaia* Travassos, 1914

Diagnosis.—Viannaiinae: Small nematodes, reddish in color when freshly collected. Body rolled in a tight spiral; cuticle marked with light transverse striations and generally without longitudinal striae, markedly dilated and expanded, especially in the males. Cuticle of the head dilated and transversely striated, separated from that of the rest of the body by an annular constriction. Bursal lobes slightly asymmetrical. Ventral rays separated for their entire length. Lateral rays diverging from each other; externo-dorsal rays arising from a common trunk with the dorsal, sometimes asymmetrically; dorsal ray divided into 2 branches in its distal third, each branch terminating in 3 points. Spicules short and slender; gubernaculum present or absent. Vulva close to anus, ovejector well developed, uterus and ovary single.

Type species.—*Viannaia viannai* Travassos, 1914.

Viannaia viannai Travassos, 1914

Description.—Viannaia: Males about 3 mm long, females from 3.75 to 5 mm long. Body spirally rolled; cuticle markedly inflated, with fine transverse and longitudinal striae. Cervical inflation about 0.05 mm long and 0.025 mm wide. Cervical inflation marked with coarser striations than the inflated cuticle of the remainder of the body.

Male with small caudal bursa. Ventral rays close together for the greater part of their length and diverging at the tips; ventro-lateral ray the longest of the bursal rays, extending to margin of bursa. Lateral rays divergent, externo-lateral arising independently and separated from the other laterals; medio- and postero-laterals arising from a common trunk; all laterals reaching margin of bursa. Externo-dorsal rays arising from a common trunk with the dorsal, sometimes asymmetrically; they narrow abruptly near the tip and terminate in blunt points which are turned outward. Dorsal ray dividing into 2 branches in its distal portion, each branch ending in 3 points. Spicules equal, slender, 0.1 to 0.11 mm long, ending in finely drawn out curved points. Gubernaculum absent.

Female with posterior portion of body markedly thickened. Uterus and ovejector single. Anus about 0.05 mm and vulva about 0.085 mm from the tail end. Body narrowing abruptly behind anus and ending in a blunt point. Eggs 0.055 to 0.060 mm long by 0.040 to 0.045 mm wide.

Host.—Opossum, *Didelphys virginiana.*

Location.—Small intestine.

Distribution in the United States.—Maryland and Georgia.

There are a few minor points in which our specimens of *Viannaia viannai* differ from those described by Travassos. In 1937 (*loc. cit.*) he mentioned the presence of long, fine, prebursal papillae and in 1921 (*loc. cit.*) he reported that the vulva was located 0.14 mm and the anus 0.13 mm from the tail end. In our specimens no prebursal papillae were observed and the anus and vulva were located 0.050 and 0.085 mm, respectively from the tail end. Travassos further stated (1921, *loc. cit.*) that the cuticle of *V. viannai* was without longitudinal striations; our specimens show longitudinal striae. In other respects the North and South American specimens are in agreement, and the differences enumerated are regarded as insignificant.

The lungworm, *Protostrongylus rushi* Dikmans, 1937, of the mountain sheep, *Ovis canadensis*. G DIKMANS, U. S. Bureau of Animal Industry.

A description of this lungworm was published in 1937 (Rabot. Gelmint. (Skrjabin) pp. 126–128) but as the volume in which this paper appeared was published in Moscow, the description and the figures illustrating the characters differentiating it from other members of the genus are not generally available in North America. Since no reprints of the article were received it has been difficult to point out to interested persons the difference between this and other species of the genus *Protostrongylus*. Consequently a slightly revised description of the species, including figures which illustrate the morphological characters that serve to differentiate it from other members of the genus, is reproduced herewith.

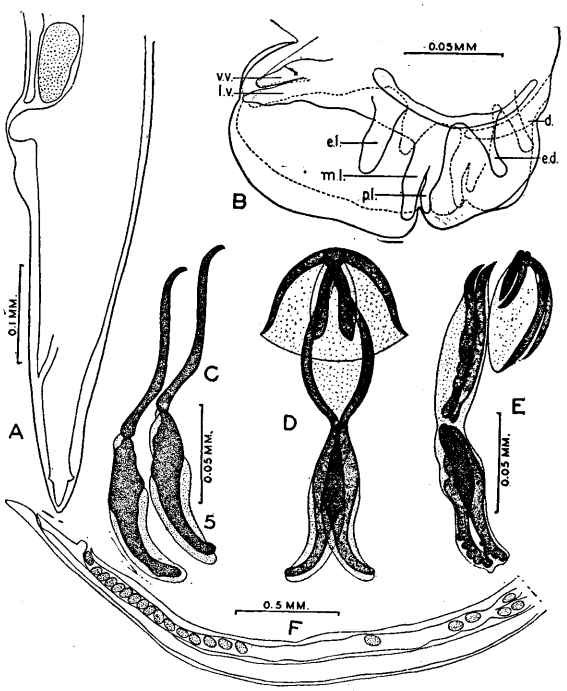

FIG. 1. *Protostrongylus rushi*. A—Tail end of female. B—Bursa of male. C—Paired accessory pieces. D—"En face" view of unpaired and paired accessory pieces. E—Lateral view of unpaired and paired accessory pieces. F—Posterior portion of female.

Protostrongylus rushi Dikmans, 1937

Description.—Male 45 to 50 mm long by 0.2 mm wide in region of proximal ends of spicules and 0.16 to 0.17 mm wide in region of bursa. Bursa (Fig. 1, B) with rays disposed in pattern characteristic of genus: Ventral rays united except in distal third, in this part separate and distinct, ventro-ventrals somewhat shorter than latero-ventrals; externo-lateral rays separate, widely separated from ventrals; medio-lateral and postero-lateral rays originating from a common stem, separated in their distal portions, postero-laterals somewhat shorter than medio-laterals; externo-dorsals relatively short; dorsal ray in available material folded underneath body, hence impossible to study in detail. Spicules characteristic of genus, 0.300 to 0.325 mm long, with slightly expanded and knob-like terminations. Spicular

sheaths supported by the usual series of digitations, these digitations beginning about 0.04 mm from proximal ends of spicules and extending to within 0.02 mm from distal ends, sheath continuing beyond ends of spicules for about 0.02 mm. Structure of unpaired accessory piece (Fig. 1, D and E) resembling that in other species of genus, of a rather elaborate nature, its exact morphology difficult to determine, but apparently consisting of the following parts: A central triangular head with apex directed caudally, 2 arms directed laterally and posteriorly, and 2 boat-like structures directed posteriorly with the keels directed laterally, a thin colorless sheath stretching between the 2 arms. Paired accessory pieces (Fig. 1, C, D, and E) consisting of slender, lightly pigmented and slightly cuticularized proximal portions about 0.095 mm long, and more deeply pigmented and more heavily cuticularized distal portions about 0.085 mm long with a maximum width of 0.015 mm, the terminations of the distal portions bluntly rounded and smooth in some cases, and presenting a broken appearance in other cases. Viewed *en face* (Fig. 1, D), slender proximal portions of paired accessory pieces approach each other at their tips, and the paired and unpaired accessory pieces appear to form a single structure such as is figured by Schulz and co-workers (1933, Zool. Anz. **102**: 303–310); viewed laterally (Fig. 1, E), however, these structures appear to be separated, the slender portions of the paired accessory pieces lying dorsal to the unpaired accessory piece.

Female 50 to 60 mm long by 130 μ wide in region of vulva. Vagina (Fig. 1, F) about 2 mm long. Posterior lip of vulva (Fig. 1, A) markedly enlarged; provagina absent. Distance from vulva to tip of tail, 0.3 to 0.325 mm; from anus to tip of tail, 0.140 mm. Two papillae, not protruding beyond cuticle, about 0.025 mm from tip of tail. Eggs in vagina 0.085 to 0.090 mm long by 0.035 to 0.04 mm wide.

Hosts.—Mountain sheep, *Ovis canadensis,* and mountain goat, *Oreamnos americanus.*

Location.—Bronchi.

Type locality.—Yellowstone National Park, Wyoming, U. S. A.

This species of *Protostrongylus* may be distinguished from *Protostrongylus rufescens* (Leuckart) and *P. stilesi* Dikmans by the absence of teeth on the distal portions of the paired accessory pieces; from *P. ocreatus* (Railliet and Henry) by the shape of the terminations of the distal portions of the paired accessory pieces; from *P. macrotis* Dikmans by the presence of an unpaired accessory piece; from *P. austriacus* Gebauer by the length of the spicules, from *P. rupricaprae* Gebauer and *P. coburni* Dikmans by the absence of a provagina; and from all species of *Protostrongylus,* so far described, by the length of the vagina.

Judging from the lungworm material collected from these hosts and submitted for examination, this worm appears to be the most frequently encountered parasite in the respiratory tract of these animals. The greater frequency with which these worms are collected may be due to their greater abundance or it may be due to their location within the lungs. *Protostrongylus stilesi,* the first member of this genus described as a parasite of the lungs of the mountain sheep in North America, occurs in the tissue of the lungs. It is a very fine, hairlike nematode and it is extremely difficult to obtain sufficient material upon which to base an accurate identification. When pieces of infested lung are fixed in formalin and then sent to the laboratory for examination it is almost impossible to secure sufficient material for identification. Judging from the reports of various collectors, who have been kind enough to send specimens for identification, *Protostrongylus rushi* occurs, like *Dictyocaulus filaria* of the sheep, in the bronchi and bronchioles and is, therefore, more easily found than *P. stilesi.*

North American monogenetic trematodes. VII. The family Discocotylidae (Diclidophoroidea). EMMETT W. PRICE, U. S. Bureau of Animal Industry.

This paper is a continuation of the series dealing with the North American monogenetic trematodes and of a general revision of the Monogenea. The purpose and organization of this installment are the same as for previous sections (Price, 1937, 1938, 1939a, 1939b, 1942, and 1943).

Family DISCOCOTYLIDAE Price, 1936

Diagnosis.—Haptor terminal, usually linguiform, with 4 pairs of clamp-like suckers, and with 1 to 3 pairs of terminal hooks. Genital atrium small, unarmed. Vaginae, when present, with marginal openings located in anterior part of body a short distance posterior to level of genital aperture.

Type genus.—*Discocotyle* Diesing, 1850.

Key to subfamilies of Discocotylidae

1. Cirrus armed ..... Anthocotylinae Price
   Cirrus unarmed 2
2. Haptor armed with a single pair of hooks of the type shown in fig. 1, C; testes postovarial ..... Discocotylinae Price
   Haptor armed with 1 to 3 pairs of hooks unlike those of Discocotylinae; testes preovarial ..... Vallisinae, n. sf.

Subfamily DISCOCOTYLINAE Price, 1936

Diagnosis.—Mature individuals sometimes fused in form of letter X (*Diplozoon*). Haptor with 4 pairs of clamp-like suckers of the type shown in figure 1, B, and with 1 pair of hooks near posterior tip of haptor. Cirrus, when present, unarmed; testes lobed or follicular, postovarial. Vaginae present or absent.

Type genus.—*Discocotyle* Diesing, 1850.

Key to genera of Discocotylinae

1. Mature individuals fused in form of letter X ..... *Diplozoon* Nordmann
   Mature individuals not fused as above 2
2. Genital sucker present; single testis, extensively lobed ..... *Octomacrum* Mueller
   Genital sucker absent; follicular testes ..... *Discocotyle* Diesing

Genus *Discocotyle* Diesing, 1850[1]

Synonyms.—*Octobothrium* Leuckart, 1827, in part; *Placoplectanum* Diesing, 1858.

Diagnosis.—Haptor more or less rectangular, set off from body proper by slight constriction, bearing 4 pairs of sessile, clamp-like suckers, and with 1 pair of small hooks near posterior end. Intestine bifurcate, each branch with numerous median and lateral diverticula. Genital sucker absent. Testes follicular. Ovary pretesticular; vagina Y-shaped, openings lateral, at or near level of genital aperture. Vitellaria pre- and postovarial.

Type species.—*Discocotyle sagittatum* (Leuckart, 1842) Diesing, 1850.

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[1] *Discocotyle dorosomatis* Yamaguti (1938) cannot be included in the genus *Discocotyle* even though the presence of a double vagina with lateral openings superficially suggests such an affinity. However, the nature of the haptoral suckers and hooks, and the peculiarly armed cirrus definitely place the species in the family Mazocraeidae. In view of the double vagina, a character which differs from that in any other member of the Mazocraeidae, the new genus *Neomazocraes* is proposed, the type and only species being *N. dorosomatis* (Yamaguti, 1938), n. comb.

Discocotyle salmonis Shaffer, 1916

Fig. 1, A–E

Description.—Body lancet-shaped, 5.5 to 7 mm long by 1.7 mm wide, with distinct constriction at level of vaginal apertures. Anterior haptor in form of 2 suckers, 95 to 102 μ in diameter, opening into oral cavity. Posterior haptor rectangular, 680 to 850 μ long by about 1 mm wide, set off from body proper by a somewhat elongate constriction, bearing 4 pairs of clamp-like suckers arranged in 2 more or less parallel rows, and with 1 pair of small hooks between posterior pair

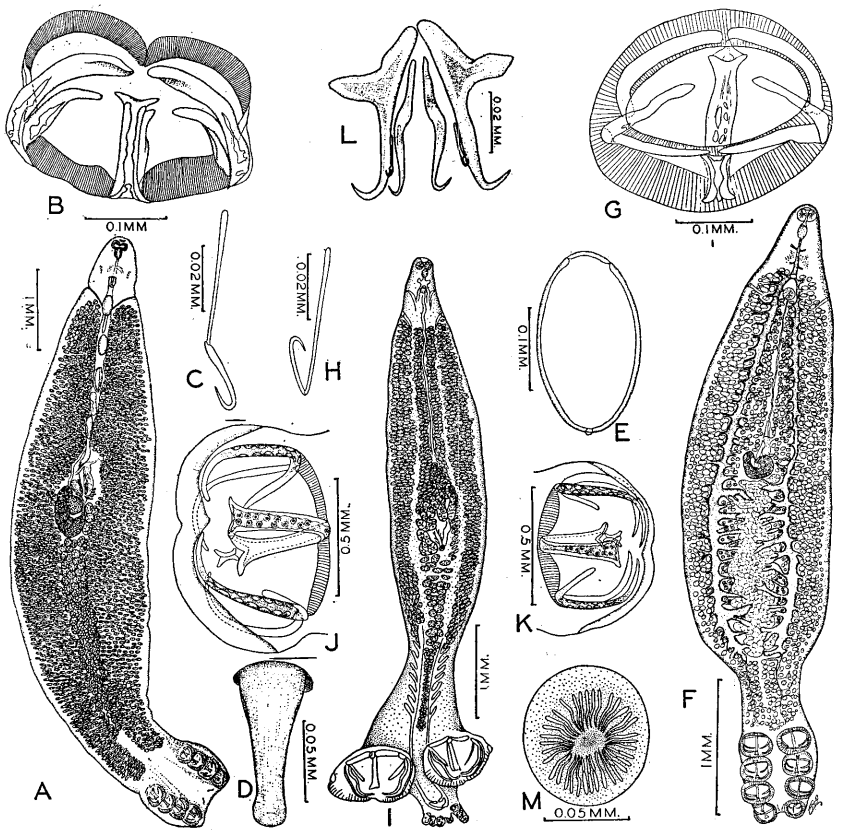

FIG. 1. A–E—*Discocotyle salmonis*. A—Complete worm, ventral view. B—Haptoral sucker. C—Haptoral hook. D—Cirrus. E—Egg. F–H—*Octomacrum lanceatum*. F—Complete worm, ventral view. G—Haptoral sucker. H—Haptoral hook. I–M—*Anthocotyle americanus*. I—Complete worm, ventral view. J—Right haptoral sucker of anterior pair. K—Left haptoral sucker of anterior pair. L—Haptoral hooks. M—Genital coronet.

of suckers. Suckers of anterior 3 pairs about equal in size, 235 to 247 μ wide, those of posterior pair usually slightly smaller, about 200 to 247 μ wide; hooks about 38 μ long, consisting of a strongly curved blade and a slender, delicate shank. Oral aperture slightly subterminal; pharynx oval, 60 to 114 μ long by 65 μ wide; intestinal branches with lateral and median diverticula, extending into haptor. Genital aperture median, 510 μ from anterior end of body. Cirrus club-shaped, 76 to 115 μ, usually extruded. Testes numerous. Ovary elongate, folded, median, pre-

testicular. Vitelline follicles extending from level of vaginal apertures to near anterior margin of haptor. Vagina Y-shaped, openings dorso-lateral 595 to 680 μ from anterior end of body. Uterus long and slender. Eggs oval, 254 to 265 μ long by 133 μ wide, with button-like knob at antopercular pole.

Hosts.—*Salmo irideus* Gibbons, *S. salar* Linnaeus, *S. fario* Linnaeus, *Salvelinus fontinalis* (Mitchill) and *Leucichthys ontariensis* Jordan and Evermann.

Location.—Gills.

Distribution.—United States. (New York Aquarium and Cold Spring Harbor, L. I.).

Specimens.—U.S.N.M. Helm. Coll. 35083, 35097, 35109, 35131, 35132, 35154, 35171, 35201, 35278, 35330, and 35595–35606.

This species was originally described by Shaffer (1916) from material collected at Cold Spring Harbor, Long Island. A large number of specimens of this form were collected at the New York Aquarium by the late Dr. G. A. MacCallum during several years. The description given above is based on the MacCallum material.

It is extremely doubtful whether *Discocotyle salmonis* represents a species distinct from *D. sagittatum* (Leuckart)—syn. *Mazocraes sagittatum* (Leuckart) Southwell and Kirshner (1937)—from European salmonoid fishes. However, since there exists no accurate modern description of *D. sagittatum* the possible identity of the two forms must remain unsettled until an adequate comparison of American and European specimens can be made.

Genus *Octomacrum* Mueller, 1934

Diagnosis.—Genital sucker present. With single, lobulated testis. Vagina absent. Other characters as in *Discocotyle.*

Type species.—*Octomacrum lanceatum* Mueller, 1934.

Octomacrum lanceatum Mueller, 1934

Fig. 1, F–H

Synonym.—*Octobothrium sagittatum* Leuckart, of Wright, 1879.

Description.—Body lanceolate, 4.2 to 6 mm long by 1.2 to 2 mm wide. Anterior haptor in form of a pair of suckers, 90 to 100 μ in diameter, opening into oral cavity. Posterior haptor rectangular, 765 to 800 μ long by 680 to 714 μ wide, bearing 4 pairs of clamp-like suckers arranged as in *Discocotyle salmonis,* and with 1 pair of hooks between posterior pair of suckers. Suckers of anterior 3 pairs about equal in size, 255 to 340 μ wide, those of posterior pair distinctly smaller, 170 to 255 μ wide; hooks delicate, about 57 μ long, similar to those of *D. salmonis.* Oral aperture terminal or slightly subterminal; pharynx oval, 57 to 95 μ long by 64 to 76 μ wide; intestine as in *D. salmonis.* Excretory apertures dorso-lateral, at or near level of genital aperture, about 595 to 680 μ from anterior end of body. Cirrus heavily cuticularized, about 50 μ long, surrounded by a strongly muscular genital sucker about 115 μ in diameter. Testis multilobed but not broken up into distinct follicles, postovarial. Ovary elongate, folded, to right of median line. Vitelline follicles extending from a short distance anterior to genital aperture to anterior margin of haptor. Genito-intestinal canal present, opening into left branch of intestine a short distance anterior to ovary. No eggs present in available specimens.

Hosts.—*Catostomus commersonii* (Lacépède) and *Erimyzon oblongus* (Mitchill).

Location.—Gills.

Distribution.—United States (Oneida Lake, New York) and Canada.

Specimens.—U.S.N.M. Helm. Coll. No. 32570 (cotypes).

This species was first reported from North America by Wright (1879) as *Octobothrium sagittatum,* his specimens having been collected in Canada. While

Octomacrum lanceatum is very similar to *Discocotyle sagittatum*, the absence of vaginae and the presence of a single lobed testis instead of numerous or follicular testes exclude it from the genus *Discocotyle*. Mueller (1934) considered as another difference between the two genera the absence of hooks between the posterior pair of haptoral suckers. However, a restudy of the cotype specimens shows hooks similar to those in *D. sagittatum* to be present; they are very delicate and difficult to detect.

Genus *Diplozoon* Nordmann, 1832

Synonym.—*Diporpa* Dujardin, 1845.

Diagnosis.—Mature individuals fused in form of letter X. Haptor rectangular, concave ventrally, bearing 4 pairs of clamp-like suckers, and with 1 pair of hooks near posterior end. Intestine not bifurcate, with numerous lateral unbranched diverticula. Testis single, at posterior end of body proper. Ovary pretesticular. Vitellaria preovarial. Vagina absent.

Type species.—*Diplozoon paradoxum* Nordmann, 1832.

This genus is placed in the same subfamily as *Discocotyle* and *Octomacrum* because of the similarity of the clamp-like haptoral suckers and of the haptoral hooks. The genus at present contains two species, *D. paradoxum* Nordmann (1832) from European fresh-water fishes, and *D. nipponicum* Goto (1891) from Japanese fishes.

VALLISINAE, new subfamily

Diagnosis.—Body proper divided into 2 parts, transverse axis of anterior part at right angles to that of posterior part, which is curved laterad. Haptor linguiform, short, not distinctly set off from body proper, bearing 4 pairs of suckers arranged in 2 more or less converging rows, and with a pair of hooks near tip. Cirrus unarmed. Testes numerous, preovarial. Vaginae absent.

Type genus.—*Vallisia* Perugia and Parona, 1890.

Genus *Vallisia* Perugia and Parona, 1890

Diagnosis.—Characters of subfamily.

Type species.—*Vallisia striata* Perugia and Parona, 1890.

This genus contains only the type species which was described from *Lichia amia* by Perugia and Parona (1890); it was later described as *Octocotyle arcuata* by Sonsino (1890). This species is not known to occur on North American hosts.

Subfamily ANTHOCOTYLINAE Price, 1936

Synonym.—*Diaphorocotylinae* Monticelli, 1903, in part.

Diagnosis.—Haptor with 4 pairs of clamp-like suckers, those of anterior pair may be very large, and with terminal lobe armed with 2 or 3 pairs of hooks. Cirrus armed with a circle of hooks. Testes numerous, postovarial. Vaginae present, openings marginal.

Type genus.—*Anthocotyle* Beneden and Hesse, 1863.

Key to genera of Anthocotylinae

Suckers of anterior pair very large as compared with those of posterior three pairs ..... *Anthocotyle* Beneden and Hesse

Suckers of anterior pairs not larger than those of posterior 3 pairs. *Winkenthughesia*, n. g.

Genus *Anthocotyle* Beneden and Hesse, 1863

Diagnosis.—Suckers of anterior pair larger than those of posterior pairs; terminal lobe armed with 3 pairs of dissimilar hooks.

Type species.—*Anthocotyle merluccii* Beneden and Hesse, 1863.

This genus has been placed in a separate subfamily and included in the family Discocotylidae although some of its characters are distinctly those of the Microcotylidae. The supporting structures of the suckers are much more like those of the microcotylids than of the discocotylids. However, the vaginal openings are lateral and in the position characteristic for *Discocotyle*. The two genera comprising this subfamily represent an intermediate group and the assignment of them to the Discocotylidae is purely arbitrary.

Anthocotyle americanus (MacCallum, 1916), n. comb.

Fig. 1, I–M

Synonym.—*Anthocotyle merluccii americanus* MacCallum, 1916.

Description.—Body 6 to 6.9 mm long by 1.1 mm wide (9 to 12 mm long by 1.2 to 2 mm wide, according to MacCallum). Anterior haptor in form of 2 circular suckers, 79 to 95 μ in diameter, opening into oral cavity. Posterior haptor more or less triangular, bearing 4 pairs of clamp-like suckers and a terminal lobe armed with 3 pairs of hooks. Suckers of anterior pair unequal, right 935 μ to 1.2 mm wide, left 680 to 850 μ wide; those of posterior 3 pairs about equal in size, 68 to 85 μ wide. Posterior lobe of haptor truncate; hooks of outer pair 60 μ long, those of intermediate pair about 20 μ long, and those of inner pair about 45 μ long. Oral aperture slightly subterminal; pharynx oval, 120 μ long by 76 μ wide. Genital aperture median, 560 to 680 μ from anterior end of body. Genital coronet with approximately 35 to 38 hooks measuring 30 μ in length. Testes numerous, in median field of post equatorial region of body proper. Ovary elongate, folded, pretesticular. Vitelline follicles abundant, extending from slightly posterior to vaginal apertures to anterior portion of haptor. Vaginal apertures lateral, 765 to 850 μ from anterior end of body. No eggs present in available specimens.

Host.—*Merluccius bilinearis* (Mitchill).

Location.—Gills.

Distribution.—United States (Woods Hole, Mass.) and ? Canada.

Specimens.—U.S.N.M. Helm. Coll. Nos. 35133, 35607, 35608, and 8191.

This species is closely related to *A. merluccii* Beneden and Hesse as described by Cerfontaine (1895) and may possibly be the same. However, there appears to be less disparity in the size of the right and left clamp-like haptoral suckers of the anterior pair in *A. merluccii* than in *A. americanus*. The illustration given by Beneden and Hesse (1863) for *A. merluccii* shows the clamp-like suckers of the anterior pair to be equal in size, while that given by Cerfontaine (1895, 1896) shows the left to be slightly larger than the right; however, no mention of this inequality is given in Cerfontaine's description of the worm. In the specimens of *A. americanus* available to the writer the right sucker is strikingly larger than the left. In addition to the differences in the suckers as pointed out above there are 3 pairs of hooks on the terminal lobe of the haptor in *A. americanus*, while only 2 pairs are figured by Cerfontaine. This difference may not be significant, since the intermediate pair of hooks are difficult to observe because they are closely applied to the hooks of the outer pairs, and it is possible that they might have been overlooked by Cerfontaine.

Stafford (1904) reported *Anthocotyle merluccii* Beneden and Hesse from *Merluccius bilinearis* in Canada, but it is more likely that the form listed by him is the same as that described here as *A. americanus*, rather than the European *A. merluccii*, if the two species are actually distinct.

The specimens at the writer's disposal consisted of a few specimens collected by the late Dr. G. A. MacCallum on three dates, August 6, 1913, July 23, 1914, and

August 22, 1914, and 1 specimen collected by the late Dr. Edwin Linton, July 2, 1924, at Woods Hole, Mass.

Winkenthughesia, new genus

Diagnosis.—Haptoral suckers about equal in size; terminal lobe with 2 pairs of dissimilar hooks. Other characters as in *Anthocotyle.*

Type species.—Winkenthughesia thyrites (Hughes, 1928), n. comb.

The type and only species of this genus was described under the name *Octobothrium thyrites* by Hughes (1928) from specimens collected from the gills of *Thyrites atun* in Australia. This species cannot be included in the genus *Octobothrium* (= *Mazocraes*) because of the type of the haptoral suckers. The organization of the parasite suggests affinities with members of the Discocotylidae; it is placed in the subfamily Anthocotylinae because the armed cirrus and the type of haptoral hooks most closely resemble those of *Anthocotyle.*

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Notes on the lungworms of porpoises and their occurrence on the California coast. ELLSWORTH C. DOUGHERTY, Department of Zoology, University of California, Berkeley, California.

Mammalian lungworms of the nematode superfamily Metastrongyloidea Lane, 1917, have been found in mammals of the orders Marsupialia, Insectivora, Primates, Carnivora (including Fissipedia and Pinnipedia), Artiodactyla, Perissodactyla, Rodentia, Lagomorpha, and Cetacea (only Odontoceti). Probably they will be discovered by further investigation in mammals of other orders. The lungworms of odontocete cetaceans[1] are exceptionally interesting because of their close host-specificity, their remarkable abundance in an infected host, and the apparently very high incidence of their infection in porpoises. These characteristics must be related to the nature of the life cycle of the worms, but there are as yet only negative data on their mode of transmission; thus it is as yet unknown whether they employ an intermediate host for development as is the case with most lungworms of terrestrial mammals, or pass directly from one definitive host to the next.

In current study of the comparative morphology of the metastrongyloid nematodes I have directed considerable attention to the lungworms of porpoises. These parasites are generally placed in the family Pseudaliidae Railliet, 1916, and by some authors (Skrjabin, 1933, 1941; Chitwood, 1937) a subfamily Pseudaliinae Railliet and Henry, 1909, has been further recognized in order to segregate the porpoise lungworms from forms placed with them in the same family, but occurring in terrestrial mammals.

I wish to present here evidence for certain changes which seem to be necessary in the nomenclature of porpoise pseudaliids, to report their occurrence in porpoises of the California coast, for which there has been hitherto no record, and to list them with their hosts. Results of my study on details of morphology will be published later. It need only be mentioned that morphologically the porpoise pseudaliids are of particular interest in that there has occurred, in common with the *Filaroides*-group of lungworms in terrestrial carnivores and in primates, great reduction in the bursa of the male from the typical strongyline condition.

The most important study in the literature on the lungworms of porpoises is that of Baylis and Daubney (1925), whose excellent revision established a group of four well defined genera where only a confusion of species in the genus *Pseudalius* Dujardin, 1844, had existed. Schmidt-Ries (1939) in a recent paper on the lungworms of the Harbor porpoise (*Phocoena phocoena*) has introduced certain errors into the interpretation of their morphology, but his work has served to focus attention on the problems of life history in the group.

Six genera have been definitely instituted by one author or another for lungworms of porpoises, and I have encountered two other generic names in the literature without being able to discover descriptions or diagnoses for them. The six defined genera are: *Pseudalius* Dujardin, 1844[2]; *Stenurus* Dujardin, 1844[2]; *Pharurus* Leuckart, 1848; *Prosthecosacter* Deising, 1851; *Torynurus* Baylis and Daubney, 1925; and *Halocercus* Baylis and Daubney, 1925. *Prosthecosacter* is clearly a synonym of *Stenurus*, as the same species, now generally known as *Stenurus minor* (Kuhn, 1829) Baylis and Daubney, 1925, must be considered genotype of both genera. Thus *Stenurus inflexus* (= *S. minor*) was the only species originally included under *Stenurus*, and *Strongylus minor* (= *Stenurus minor*) was designated

[1] Only members of the family Delphinidae (providing the present boundaries of this family remain unchanged) have been reported as hosts for metastrongyloids.

[2] This date is usually given as 1845, but, according to Neave (*Nomenclator Zoologicus*) and Sherbourne (*Index Animalium*), the publication date was actually 1844 (November).

as type of *Prosthecosacter* by Stiles and Hassall (1905). *Pharurus* has been placed as a synonym of *Stenurus* by Baylis and Daubney (1925), but I am reconsidering here the status of this genus. In addition there appear to have been established, or there apparently will be established, two further genera which were mentioned in a recent paper by Skrîabĭn (1941). This author, however, included no bibliography, and despite diligent search I have been unable to find descriptions or further reference to these genera. They are *Otophocoenurus*, for which Skrîabĭn gave no authority, nor any information save that members of the genus inhabit the inner cavities of the ear, presumably of some porpoise, and *Skrjabinalius* with type species *S. cryptocephalus* from the lungs of *Delphinus delphis*, apparently to be credited to S. L. Delîamur of the Krymskiĭ Meditsînskiĭ Înstĭtut (Crimean Medical Institute).

Leuckart (1848) established the species *Strongylus alatus* for a pseudaliid which he indicated had been found in the venous sinus of the cranium of the Narwhal (*Mcnodon monoceros*). Later von Linstow (1888) redescribed this species more adequately under the name of *Pseudalius alatus*. In a footnote Leuckart made the following statement: "Fast fühle ich mich versucht, aus unserem Wurm ein neues. . . . Genus *Pharurus* (von φᾶρος, Lappen, und οὐρά, Schwanz) zu bilden." Leuckart in effect established a genus with *Strongylus alatus* as type in case this species should be found not to belong within the genus *Strongylus* Müller, 1780; that this would prove true, he fully expected. Since *Strongylus sensu stricto* with its very limited present use by comparison with that of the nineteenth century can no longer be applied to a metastrongyloid, the availability of *Pharurus* Leuckart, 1848, must be considered. From the standpoint of metastrongyloid morphology this generic name must compete only with *Pseudalius* Dujardin, 1844, and *Stenurus* Dujardin, 1844. *Strongylus alatus* is excluded, however, from both of these genera by the nature of its spicules, which are very long and thin, whereas in the other two genera the spicules are short and leaf-like. I believe that this difference in spicular morphology is of value in establishing generic status, and thus *Pharurus* must be employed for *Strongylus alatus;* since Stiles and Hassall (1905) have already indicated the association of *Pharurus* with *alatus*, this species is correctly designated as *Pharurus alatus* (Leuckart, 1848) Stiles and Hassall, 1905. Baylis and Daubney, despite the fact that they placed *Pharurus* as a synonym of *Stenurus*, referred to this species as *Strongylus alatus*. Yorke and Maplestone (1926) were first to make the combination *Stenurus alatus*. This designation must therefore also be considered a synonym of *Pharurus alatus*.

The other two porpoise pseudaliid genera which have been defined must now come under consideration as possible synonyms of *Pharurus*. *Halocercus* Baylis and Daubney, 1925, is excluded from *Pharurus* by the following facts: that in *Halocercus* the bursal rays are very much shortened and thick, more or less equal in size, whereas those of *P. alatus* are dissimilar in size and longer; that the bursa in *Halocercus* is vestigial with very little or no indication of lobes, whereas *P. alatus* has a well-developed, clearly lobed bursa; and that the spicules, although in most species of *Halocercus* as long as those of *P. alatus*, are somewhat thicker and more or less simply arcuate in the former and thinner, almost filiform in the latter. On the other hand *Torynurus convolutus* (Kuhn, 1829) Baylis and Daubney, 1925, although possessing a singular sucking disc not shared by *P. alatus*, has similar spicules, bursal rays (though the dorsal ray is longer in *P. alatus*), and caudal alae to those of *P. alatus*. *T. convolutus* is type and only species of the genus *Torynurus*. The similarities between the two species necessitate, I feel, their inclusion in a single genus. Thus *Torynurus* Baylis and Daubney, 1925, becomes a synonym of *Pharurus* Leuckart, 1848, and *T. convolutus* must be designated *Pharurus convolutus* (Kuhn, 1829), comb. nov. The discovery of additional species in *Pharurus* may serve to bridge such differences as do exist between *P. alatus* and *P. con-*

volutus; but, if the reverse should be true and the differences are accentuated, then *Torynurus* can always be reconstituted.

From the nature of the spicules, long and slender in both genera, *Pharurus* and *Halocercus* would seem to be more closely related to each other than either are to the other two generally known pseudaliid genera in porpoises.

My study of the morphology of *Pharurus convolutus* convinces me that the species *Torynurus bicostatus* (v. Linstow, 1906), which was reëstablished by Schmidt-Ries (1939) after having been referred to as a tentative synonym of *P. convolutus* by Baylis and Daubney, is not distinct from the latter species. It must therefore be considered a synonym thereof.

The name *Strongylus inflexus,* the specific part of which is preserved in the name of the pseudaliid *Pseudalius inflexus,* is generally attributed to the second volume of Rudolphi's *Entozoorum sive Vermium Intestinalium Historia Naturalis,* published in 1809. Therein appeared the Latin diagnosis of the composite species so named, Rudolphi actually dealing with two distinct species under one name. However, in volume 1 of this monumental early work (1808) he first employed the name *Strongylus inflexus* to refer to forms previously described by Klein (1740) and Camper (1794). Klein dealt with the form now generally known as *Stenurus minor.* I have not been able to check the original work of Camper, but, according to Diesing (1851), Camper dealt with both *S. minor* and *P. inflexus,* and the same conclusion may be drawn from reading Rudolphi's discussion of *Strongylus inflexus* (1809). If Diesing was correct in identifying the lungworms studied by Camper, then the specific name *inflexus* could have been restricted to either of these species by the first worker to recognize the composite nature of Rudolphi's species. Kuhn (1829a, b), who was actually this first worker, limited the name to the form now known as *Pseudalius inflexus.* Historically only Dujardin (1844) did not do this. He referred the name to the form now generally known as *Stenurus minor,* while *Pseudalius inflexus* he called *Pseudalius filum.* However, Kuhn's usage has priority and should be followed. Hence the form to which he restricted *inflexus* is correctly designated *Pseudalius inflexus* (Rudolphi, 1808) Schneider, 1866. However, his name *Strongylus minor* must be challenged. Ten years before Kuhn (1829a) named this species, Rudolphi (1819) had employed *minor* as a variety name for a form in the genus *Strongylus,* namely for *Strongylus retortaeformis minor* (as opposed to *S. r. major*). In general zoölogical usage variety names have been accepted as of equivalent nomenclatorial value with subspecific names and hence with specific names. Therefore, the designation *minor* was preoccupied at the time Kuhn first employed it. The next name which has been attributed to Kuhn's species is *Strongylus vagans,* employed by Eschricht (1841) for certain worms observed by him free in the bronchi of the Harbor porpoise. It is true that Eschricht identified as *Strongylus vagans* the worms which Klein (1740) had reported from the venous sinus of the cranium of this host—forms that were Kuhn's species. However, in specimens actually observed by him he described the spicules as "always protruding, and of a different shape" from those of *Pseudalius inflexus.* Therefore, *Strongylus vagans* must be considered a synonym of *Pharurus convolutus,* the only species, free in the bronchi of the harbor porpoise, which has spicules that ordinarily protrude. Diesing (1851) regarded *Strongylus vagans* as a synonym of "*Prosthecosacter minor.*" Baylis and Daubney (1925) erroneously considered that "*Strongylus vagans* Eschricht, 1841," might be a synonym of the form which they called *Halocercus inflexocaudatus.* No available specific name has been applied to Kuhn's original species, therefore, it must be renamed. In view of the close host-specificity among the porpoise pseudaliids, I consider it appropriate to name it after its host, *Phocoena phocoena* (Linné), and, therefore, designate it *Stenurus phocoenae,* nom. nov.

Among the present designations of the remaining species of porpoise pseudaliids two further names should be changed. Van Beneden (1870) recorded parasitic nematodes which he indicated had been described by Pallas from the White whale (*Delphinapterus leucas*). I have not yet been able to locate this original description by Pallas, but, as there is little reason to question van Beneden's accuracy, the latter's rendition thereof would seem acceptable for my purposes. This description was necessarily very superficial. Van Beneden, however, gave the name *Strongylus Pallasii* to the nematodes. Baylis and Daubney (1925) considered this name a *nomen nudum*, but also stated that there could be little doubt but that it referred to the same organism later and more accurately described by Cobb (1888, 1889) and von Linstow (1900) and now known as *Stenurus arcticus* (Cobb, 1888) Baylis and Daubney, 1925. After considering this evidence, I cannot agree that van Beneden's name is a true *nomen nudum*. Therefore I propose that the species which is now called *Stenurus arcticus* be redesignated *Stenurus pallasii* (v. Beneden, 1870), comb. nov. The name *Strongylus invaginatus* was indicated by Baylis and Daubney as possibly referring to the form which they designated *Halocercus inflexocaudatus* (v. Siebold, 1842). It was employed by Quekett (1841) as a new species name for a worm which he reported from cysts in the parenchyma of the lungs of the Harbor porpoise (*Phocoena phocoena*). Baylis and Daubney did not accept this name as they considered Quekett's account to be inadequate for recognition of a definite species. However, they overlooked a later paper by Quekett (1844) in which a form clearly the same as that now generally termed *H. inflexocaudatus* was well described and figured under the name *Strongylus invaginatus*. Therefore Quekett's name, since first employed previously to von Siebold's (1842), must be used for the species of *Halocercus* in the Harbor porpoise, and the form is therefore correctly designated *Halocercus invaginatus* (Quekett, 1841), comb. nov.

While there are the descriptions by Wu (1929) of *Halocercus pingi* and by Hsü and Hoeppli (1933) of *Stenurus auditivus*, both from the Chinese black finless porpoise (*Meomeris phocaenoides*) of the western Pacific coast and rivers, the occurrence of pseudaliids in porpoises of the Pacific coast of North America and, in fact, of the entire eastern Pacific has not been recorded heretofore. I wish to make here a preliminary report of examination of two species of porpoises caught in San Francisco Bay, California. From a Dall porpoise (*Phocoenoides dalli*), caught in October, 1940, I acquired specimens of a new nematode of the genus *Halocercus*. This find has already been mentioned by Benson and Groody (1942). The worms were embedded in the parenchyma of the lungs in nodules which marked the surfaces thereof with several dozen white patches about a centimeter in width. In a Harbor porpoise (*Phocoena phocoena*), caught in July, 1941, I found lungworms of three species: *Stenurus phocoenae*, nom. nov.; *Pharurus convolutus* (Kuhn, 1829), comb. nov.; and *Halocercus invaginatus* (Quekett, 1841), comb. nov. *H. invaginatus* occurred in tiny nodules or cysts which were scattered throughout the parenchyma of the lungs and resembled tubercles of miliary tuberculosis. The other two were found in the bronchi. *S. phocoenae* occurred as well in the nasal passages, mouth cavity, and oesophagus, and two specimens were even recovered from the first division of the stomach. In all probability the worms had migrated, to the last three locations at least, after the death of the host. Whether the fourth pseudaliid reported from this host, *Pseudalius inflexus* (Rudolphi, 1808) Schneider, 1866, occurs in the Harbor porpoise of the Pacific will have to be determined by further investigation.

A word about the status of the host of these last four parasites seems in order. It is apparently as yet not definitely established whether or not the species of

Phocoena[3] of the North Pacific is to be considered conspecific with that of the Atlantic. The Harbor porpoise of the North Pacific has been described as a separate species, *Phocaena*[3] *vomerina*. Because of the uncertain status of this species, however, I prefer to regard it as conspecific with the European and Atlantic Harbor porpoise (*Phocoena phocoena*). It is interesting, however, that the pseudaliids in the Pacific form seem to show slight differences from those in the Atlantic form of the host, primarily in size—in harmony with a concept of slight difference in their hosts.

Lungworms from the Odontoceti so far reported are enumerated in the following list. I wish to express my appreciation to Dr. Remington Kellogg of the United States National Museum for supplying me with the scientific and vernacular names in current good usage for the porpoise hosts, to Dr. E. W. Price of the Zoölogical Division, Bureau of Animal Industry, United States Department of Agriculture, for generously lending me for study, specimens of *Pseudalius inflexus*, *Pharurus alatus*, and *Stenurus globicephalae*, and *Pharurus convolutus* from an Atlantic Harbor porpoise, and to Dr. H. A. Baylis of the British Museum (Natural History) for kindly sending me specimens of *P. convolutus* from a Harbor porpoise of the British Isles.

Pseudalius Dujardin, 1844

P. inflexus (Rudolphi, 1808) Schneider, 1866—genotype
Host: Harbor porpoise, *Phocoena phocoena* (Linné)

Stenurus Dujardin, 1844

S. phocoenae, nom. nov.—genotype
Host: Harbor porpoise, *Phocoena phocoena* (Linné)

S. globicephalae Baylis and Daubney, 1925
Host: North Atlantic blackfish, *Globicephala ventricosa*[4] (Lacépède)

S. pallasii (v. Beneden, 1870), comb. nov.
Host: White whale, *Delphinapterus leucas* (Pallas)

S. ovatus (v. Linstow, 1910) Baylis and Daubney, 1925
Host: North Atlantic bottle-nosed porpoise, *Tursiops truncatus* (Montague)

S. auditivus Hsü and Hoeppli, 1933
Host: Chinese black finless porpoise, *Meomeris phocaenoides* (Cuvier)

Pharurus Leuckart, 1848

P. alatus (Leuckart, 1848) Stiles and Hassall, 1905—genotype
Host: Narwhal, *Monodon monoceros* Linné

P. convolutus (Kuhn, 1829), comb. nov.
Hosts: Harbor porpoise, *Phocoena phocoena* (Linné) (type); (?) North Atlantic blackfish, *Globicephala ventricosa* (Lacépède)[5]

Halocercus Baylis and Daubney, 1925

H. delphini Baylis and Daubney, 1925
Host: Common porpoise, *Delphinus delphis* Linné

H. lagenorhynchi Baylis and Daubney, 1925
Host: White-snouted porpoise, *Lagenorhynchus albirostris* Gray

H. invaginatus (Quekett, 1841), comb. nov.
Host: Harbor porpoise, *Phocoena phocoena* (Linné)

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[3] The spelling *Phocoena* has priority over the spelling *Phocaena*, as pointed out recently by Scheffer (1942).

[4] Specimens which I received from Dr. Price were collected from a host identified as *Globicephala brachyptera* (Cope). In the absence of an authoritative revision of the blackfishes, Dr. Kellogg is inclined, however, to regard all North Atlantic forms as a single species, *G. ventricosa* (syn. *G. melaena*).

[5] This record is a doubtful one inasmuch as von Linstow (1889), who made it, later (1906) described *Pseudalius bicostatus*, which is synonymous with *Pharurus convolutus* from the Harbor porpoise, as a new species.

H. pingi Wu, 1929

Host: Chinese black finless porpoise, *Meomeris phocaenoides* (Cuvier)

H. brasiliensis Lins de Almeida, 1933

Host: Brazilian fresh-water porpoise, *Sotalia brasiliensis* (v. Beneden)

H., sp. nov.

Host: Dall porpoise, *Procoenoides dalli* (True)

[*Skrjabinalius* Delîamur, *teste* Skrîabīn, 1941

S. cryptocephalus[6] Delîamur—genotype, *teste* Skrîabīn

Host: Common porpoise, *Delphinus delphis* Linné]

[*Otophocoenurus, teste* Skrîabīn, 1941]

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[6] This was given as *S. cryptocephala* by Skrîabīn, but I have emended the specific name to *cryptocephalus* in order that it should agree in gender with its genus.

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Overwinter loss of *Haemonchus contortus* larvae from a sheep pasture. MERRITT P. SARLES, U. S. Bureau of Animal Industry.

Mainly on the basis of the work of Ransom (1906, U. S. Bur. Anim. Indus., Circ. 93), on the development and resistance of the infective larvae of the sheep stomach worm, *Haemonchus contortus,* the ensheathed larvae of this pathogenic nematode are quite generally regarded as being able to survive for long periods on pasture, including overwinter, and to be unusually resistant to drying. However, the data in a number of recent papers indicate that the majority of the larvae of this species, as well as those of most other nematodes parasitic in sheep, die off rapidly on pasture, and that in the colder parts of the country the free-living stages of *H. contortus* are unable to survive overwinter. This is an important point in connection with the planning of measures for the control of sheep parasites, and additional information pertinent to specific local conditions and regions is needed.

The work reported herein was done at the U. S. Department of Agriculture, Beltsville Research Center, Beltsville, Md., under natural pasture conditions. The larvae had accumulated on a small 0.7-acre pasture during a period of nearly 6 months from May 10, 1940, to November 1, 1940, from 4 lambs having light stomach infections. Although reinfection obviously occurred during the summer, the infections were kept at a low level by the administration of phenothiazine to each animal during the summer. On account of the temporary disappearance of eggs from the feces for 3 weeks after each treatment, the seeding of the pasture with eggs was intermittent, but a calculated total of 820,000 stomach worm eggs was passed on the area during the 6-month period. All 4 sheep contributed to this total, as shown by weekly fecal examinations and by the finding of stomach worms in all of them when they were slaughtered after being held indoors over the following winter.

At the end of the period of contamination the pasture was left unoccupied and unentered for 7 months (November 1, 1940, to June 2, 1941). The pasture was then tested, with negative results, for the presence of larvae by direct microscopic

examination and attempted Baermann isolation of larvae from samples of fecal pellets and soil, and also by grazing 2 worm-free lambs on the pasture for 2 weeks. When these lambs were slaughtered, after being held for 3 additional weeks in a cage under conditions precluding accidental infection, no stomach worms or lesions suggestive of parasitism were found. Two other lambs, used at the same time to test another pasture for the survival of nodular worm larvae (Sarles, Jour. Parasitol., in press), and to serve as controls on the freedom of the test animals from accidental stomach worm infection, also showed no evidence of stomach worms.

Fecal examinations made on these 4 lambs, and on 2 others which were kept in cages as additional controls but which were not slaughtered because the experiment gave a negative result, failed to show any evidence of stomach worm eggs in 3 different fecal specimens examined during the 38 days between the time the lambs were weaned indoors and the time they were put on pasture. Cultures of small amounts of feces from each of the 6 lambs were negative 26 days after the animals were weaned and also at the time they were put on pasture. Two additional fecal examinations, made at the time the lambs were removed from pasture and at the time of slaughter, likewise failed to reveal any evidence of stomach worm infection.

The overwinter loss of *H. contortus* larvae occurred on a closely cropped pasture, during a fairly normal winter, followed by a dry spring, and unusually wet conditions favorable to the acquisition of infection prevailed during the grazing period of the test lambs. More detailed information concerning the pasture and weather conditions of this experiment are given in the paper dealing with the companion experiment on nodular worm survival (Sarles, *loc. cit.*).

These results show that on a pasture contaminated during the summer with small numbers of stomach worm eggs, the free-living stages of the parasite died off overwinter rendering the pasture safe for clean stock the next spring. The facts presented give additional support to the growing mass of evidence which shows that pastures which have been rested overwinter are relatively, if not completely, free of infective stomach worm larvae. Such pastures are therefore relatively safe for lambs and for treated breeding stock.

Overwinter survival on pasture of preparasitic stages of some nematodes parasitic in sheep. KENNETH C. KATES, U. S. Bureau of Animal Industry.

INTRODUCTION

In general, investigators have found that natural exposure to winter conditions causes a reduction in the number of larvae of certain species of sheep nematodes on pastures, whereas in the case of other species virtual sterilization of infected pastures resulted. Shorb (1942) reported that small numbers of preparasitic stages of *Ostertagia*, *Trichostrongylus*, and possibly *Nematodirus* survived the winter months on pasture at Beltsville, Md., while those of *Haemonchus*, *Cooperia*, *Chabertia*, *Bunostomum*, and *Oesophagostomum* failed to survive. Similar observations were made by Griffiths (1937) and by Swales (1940, 1942) in Canada. Sarles (1943a, b) is reporting that at Beltsville, Md., a 0.7-acre pasture infected with *Oesophagostomum columbianum* and *Haemonchus contortus* during the summer and fall of 1940 and rested from November 1, 1940, to June 2, 1941, was free from infective larvae of these two species when tested by grazing with parasite-free lambs. Furthermore, Zavadovskiĭ and Vorob'eva (1934) stated that in the vicinity of Moscow larvae of the *Trichostrongylidae* have little invasive power after being exposed to overwinter conditions on pasture. In contrast, Dikmans and Andrews (1933) observed at Beltsville, Md., that larvae of *Haemonchus*, *Ostertagia*, and *Nematodirus* survived on pasture over winter, and Baker (1939) reported that in New York State various nematode larvae survived at least 21 months on pasture; no quantitative data were given in either of these reports.

During the past year the writer had an opportunity to obtain further information on this subject, and the purpose of this paper is to present the results of this investigation.

PROCEDURE

A ¼-acre pasture at the United States Department of Agriculture Beltsville Research Center, Beltsville, Maryland, was continuously grazed for several months during the spring and summer of 1941 by 10 goats that were very heavily infected with various nematodes and for several weeks in the fall of the same year by 18 similarly infected sheep. Supplemental feed was given the animals at all times to prevent complete exhaustion of the pasture. Periodic differential egg counts were made on all of the infected animals; several deaths due to gross parasitism occurred during the period the animals were on the pasture. All animals that died were necropsied and the data concerning them have been reported elsewhere (Kates, 1942). The data obtained from egg counts and necropsies showed that during the spring, summer, and fall of 1941, there had been a continuous deposition on the pasture of very large numbers of eggs of *Haemonchus contortus, Trichostrongylus* spp., *Ostertagia* spp. and *Oesophagostomum columbianum,* and relatively smaller numbers of eggs of *Cooperia curticei, Nematodirus* spp., *Bunostomum trigonocephalum* and *Trichuris ovis.*

On October 15, 1941, all the animals were removed and the pasture was then allowed to lie idle over the winter. Beginning on May 2, 1942, when a good growth of grass first appeared, 2 lambs, parasite-free except for coccidia and *Strongyloides papillosus,* were grazed for 2 weeks on a small fenced-off area (30 by 60 feet) of this pasture. During this period the 2 lambs grazed this plot very close to the soil surface, insuring the ingestion of the majority of infective larvae present on the grass. Thereafter, the test lambs were placed in a clean pen to prevent further infection. Four weeks after their removal from the pasture they were killed and all the nematodes present in the gastrointestinal tract were collected and counted.

Temperature range and precipitation during the period of the experiment were about normal for the Beltsville region, except that October and November, 1941, and April, 1942, were unusually dry. Weather data were collected within a few feet of the experimental pasture, and these are summarized in table 1.

TABLE 1. *Summary of air temperatures (in shade) and precipitation during the period of the experiment*[a]

Month	Temperatures (° F.)[b]			Precipitation[c]
	Mean maximum	Mean minimum	Mean	
				Inches
1941				
October 15–31	74.4	40.6	57.5	0.24
November	59.7	30.2	44.9	1.04
December	51.0	27.7	39.3	3.44
1942				
January	41.3	21.7	31.5	1.64
February	40.6	21.8	31.2	3.21
March	55.6	32.9	44.2	5.44
April	71.5	40.0	55.7	0.66
May 1–16	79.8	51.2	65.5	1.33

[a] The experiment began October 15, 1941, and the test lambs were placed on pasture from May 2 to 16, 1942, when the test period terminated.

[b] Highest temperature recorded during experimental period was 92° F. and the lowest – 4° F.

[c] Very little snow cover was present on this pasture during the winter of 1941–1942. The longest period of snow cover was about 4 days in March, 1942.

RESULTS AND CONCLUSIONS

The total numbers of nematodes obtained from the two test lambs necropsied on June 13, 1942, 4 weeks after removal from the pasture, were as follows: 3 *Trichostrongylus colubriformis,* 5 *Haemonchus contortus,* 18 *Trichuris ovis,* 386 *Nematodirus* spp. (mainly *N. spathiger*), 514 *Ostertagia* spp. (mainly *O. circumcincta*).

For the conditions existing at Beltsville, Maryland, over the fall, winter, and spring of 1941–1942, these results show there was no evident survival on pasture of the preparasitic stages of *Oesophagostomum columbianum, Cooperia curticei,* and *Bunostomum trigonocephalum;* a very low survival of *Haemonchus contortus* and *Trichostrongylus* spp.; and a relatively high survival of *Ostertagia* spp., *Nematodirus* spp., and *Trichuris ovis.*

These observations also show that over-winter exposure of preparasitic stages of sheep nematodes on idle pastures results in a variable reduction in the number of larvae of certain species, and complete destruction of the larvae of others. The general consensus, confirmed by this report, is that the larvae of *Oesophagostomum, Cooperia,* and *Bunostomum* do not survive overwintering on pasture; that those of *Haemonchus* and *Trichostronglyus* either do not survive or survive in very small numbers depending upon (a) intensity of pasture infections, (b) severity of the winter weather, and (c) length of exposure period; and that the preparasitic stages of *Ostertagia, Nematodirus,* and *Trichuris* are the most resistant to the effects of winter weather conditions.

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The occurrence of swine ascarid eggs in the intestinal contents and in the droppings of wild rats. HARRY E. ZIMMERMAN, JR., U. S. Bureau of Animal Industry.

It has been observed by a number of investigators that eggs of hookworms, ascarids, whipworms, and other helminth parasites when ingested by dogs, pigs, and other mammals and birds may pass unharmed through the alimentary tract. In this connection, eggs of the swine ascarid were found in the intestinal contents of two of a series of wild rats trapped (during August and September, 1942) in

the vicinity of hog pens at the Beltsville Research Center, Beltsville, Maryland. The intestinal contents of the rodents were washed and sedimented and 39 ascarid eggs recovered; some of the eggs were nonsegmented, whereas others were in the 2-celled stage of segmentation. When cultured in tap water the eggs became fully embryonated in approximately 49 days; at that time they were fed to a white mouse to test the vitality of the contained embryos.

The day after infection 14 eggs and 2 dead embryos were recovered from the droppings of the test mouse. Three days after infection the mouse was necropsied and 11 live ascarid larvae were recovered from the liver; this demonstrated that some of the eggs were viable. No larvae were recovered from the lungs and alimentary tract of the mouse.

Subsequent examinations of droppings of wild rats collected in the vicinity of the animal pens and in the barns revealed the presence of swine ascarid eggs in some of the pellets; the eggs were unembryonated although some were in the 2-celled stage; all were capable of further segmentation.

In the course of investigations on swine parasites carried out during the past 10 years at the Beltsville Research Center extraneous infections with ascarids have occurred in several pigs even though the animals were members of litters farrowed by ascarid-free sows and maintained under the conditions of rigid sanitation as described by Spindler (1942, Proc. Helminth. Soc. Wash. 9(1): 22–23). In light of the finding of viable ascarid eggs in the alimentary tract and in droppings of wild rats, it is considered that rodents may have been responsible for the extraneous infections just mentioned. Rats feeding on the feces of ascaris-infected swine may have ingested eggs and later voided them in droppings in locations where sufficient moisture was available for complete development and where the eggs somehow became accessible to the pigs.

In light of the findings presented herein, the possibility that wild rats may occasionally be responsible for dissemination of ascarids and other parasites of livestock under farm conditions should be kept in mind.

Inoculations of *Trichomonas foetus* (Protozoa) in guinea pigs.[1] BANNER BILL MORGAN, Department of Veterinary Science, University of Wisconsin.

From available literature it appears that the guinea pig is not a suitable laboratory animal for continued cultivation of *Trichomonas foetus* through various methods of inoculations. The purpose of this paper is to present research on inoculations with *T. foetus* in 125 guinea pigs. From this data one may draw certain conclusions as to the feasibility of the guinea pig as a laboratory animal for inoculations with *T. foetus*. In a previous paper the writer (1942) reported on injections of *T. foetus* in white rats and mice. Byrne (1942) showed that rabbits could be utilized with success as laboratory animals for vaginal inoculations with *T. foetus*.

Intraperitoneal injections.—Riedmuller (1928, 1930) demonstrated that initial inoculations of *T. foetus* resulted in approximately 16 per cent of the animals positive. However, trichomonad exudate taken from a susceptible guinea pig and inoculated serially into other guinea pigs increased the virulence and 96 per cent of the animals became positive. Abelein (1932) obtained pure cultures of *T. foetus* by inoculation of contaminated trichomonad exudate into guinea pigs. This has been confirmed by Riedmuller (1932) and Nelson (1938). Witte (1933) by injection of trichomonad pyometra material or pure cultures of *T. foetus* could infect 41 per cent of all guinea pigs inoculated. The early German work usually associ-

[1] Published with the approval of the Director of the Wisconsin Agricultural Experiment Station. Project No. 622–V, Trichomoniasis and other reproductive diseases of cattle.

ated abortions in guinea pigs as positive for *T. foetus* regardless of negative findings of the causative organism. Consequently, some of this work cannot be evaluated accurately.

Walsh, McNutt, and Murray (1934) first demonstrated that *T. foetus* was not always recovered from abortions which occurred after injections of trichomonads. Thirteen pregnant guinea pigs were inoculated, 6 aborted, but trichomonads were recovered from only one. Futamura (1935) inoculated vaginal discharges of trichomonad-infected cows into 7 pregnant guinea pigs. Six animals aborted from 1 to 20 days after inoculation. Trichomonads could be recovered in a majority of cases. Jensen (1938) inoculated 10 pregnant guinea pigs with trichomonad pyometra material, and only one animal aborted. Zeetti (1940) produced death in female guinea pigs with inoculations of trichomonad cultures or exudate. Trussell and McNutt (1941) inoculated 9 guinea pigs with pure cultures of *T. foetus* without success.

Vaginal inoculations.—Riedmuller (1928) produced vaginitis in 2 guinea pigs by inoculations with trichomonad exudate from an infected cow. Witte (1933) conducted similar experiments with 8 guinea pigs. One animal aborted and trichomonads were recovered, while another animal showed a persistent uterine infection. Zeetti (1940) produced abortions in guinea pigs by vaginal inoculations of *T. foetus*. Trussell and McNutt (1940) failed to obtain infections with the inoculation of 2 guinea pigs with pure cultures of *T. foetus*.

Other methods of inoculation.—Riedmuller (1928) failed to infect guinea pigs by intravenous and subcutaneous injections. Witte (1933) injected pregnant guinea pigs intramuscularly, subcutaneously, intracardially, conjunctivally, and orally with pure cultures of *T. foetus*. One animal inoculated subcutaneously and one intramuscularly died from trichomoniasis. The remainder showed little or no symptoms. Trussell and McNutt (1941) failed to infect 3 guinea pigs by subcutaneous inoculations or 10 animals intracranially with *T. foetus* cultures. Lwoff and Nicolau (1935) produced fatal encephalomyelitis in guinea pigs by subdural inoculations.

One hundred and twenty-five guinea pigs were used in the following experiments. Injected materials consisted of the following solutions: (1) living *T. foetus* centrifuged and washed 3 times and suspended in 0.7 per cent saline in a concentration of 10 million per cc, (2) liquid portion of 72-hour *T. foetus* cultures composed of buffered saline citrate solution with 5 per cent bovine serum. The concentration was approximately 3 million living organisms per cc, (3) bacteria-free trichomonad pyometra fluid from an infected cow with a count of 1½ million living organisms per cc, (4) liquid portion of unused sterile culture material. Solution 4 was used for controls.

Experiment 1. To demonstrate the effects of intraperitoneal inoculations of T. foetus in guinea pigs.—Twenty-four guinea pigs in 2 groups of 12 were injected with 5 and 10 cc respectively of solution 1. The animals showed no ill effects and the average maximum time that motile trichomonads could be recovered from the peritoneal cavity was 12 hours. Animals were examined daily for 1 month by introducing a sterile needle into the body cavity and withdrawing some abdominal fluid. After 30 days the animals were killed and examined but no trichomonads were recovered. Five controls were negative.

Twenty guinea pigs in 2 groups of 10 were injected with 5 and 10 cc, respectively, of solution 2. Two animals, both inoculated with 10 cc of material, appeared sluggish and were positive for motile trichomonads for 10 and 14 days, respectively. One of the animals died on the 11th day and *T. foetus* was recovered from the peritoneal cavity. The second animal was sacrificed on the 15th day and motile trichomonads were also recovered in pure culture from the abdominal cavity.

Approximately 10 cc of purulent exudate was removed from this animal. The remainder of the animals were negative for 25 days when they were sacrificed. No motile trichomonads were recovered. Five controls were negative. Attempts to infect 18 guinea pigs with solution 3 yielded one animal positive for 5 days while the remainder were negative. This animal also showed a vaginal discharge which was positive for *T. foetus*. Thus, it appears that in the majority of cases guinea pigs are refractory to intraperitoneal injections of *T. foetus*. An occasional guinea pig may become infected. The factors involved for positive cases are not clear.

Experiment 2. To demonstrate the effects of subcutaneous inoculations of T. foetus in guinea pigs.—Eight guinea pigs were inoculated subcutaneously with 8 cc of solution 3. Three weeks later all 8 guinea pigs had developed abscesses which contained from 3 to 12 cc of purulent fluid containing millions of motile trichomonads (2 million per cc). Five more guinea pigs were inoculated subcutaneously with 5 cc each of the purulent fluid collected aseptically. Two abscesses were positive after 21 days and 3 were positive at 30, 38, and 42 days, respectively. Six controls were negative. Attempts to produce subcutaneous abscesses containing motile trichomonads in 20 guinea pigs with solutions 1 and 2 failed. Thus, subcutaneous injections in guinea pigs with sterile trichomonad pyometra material produced trichomonad abscesses in all attempts. Serial transfers subcutaneously under aseptic conditions were also successful. Subcutaneous injections of washed *T. foetus* or in culture material failed.

Experiment 3. To demonstrate the effects of vaginal inoculations of T. foetus in guinea pigs.—Thirty non-pregnant guinea pigs were divided into 3 groups of 10 each and inoculated with 2 cc of solutions 1, 2, and 3, respectively. Each animal was examined 10 times during a 30-day period. Only 2 guinea pigs were positive for *T. foetus* for 5 days after inoculation with solution 3. Eight controls were negative. Thus, vaginal inoculations of *T. foetus* failed in the majority of cases to produce trichomonad infections in guinea pigs as could be detected by direct smears or cultures.

SUMMARY

Sixty-two guinea pigs divided into 3 groups were inoculated intraperitoneally with washed *Trichomonas foetus* suspended in saline with a concentration of 10 million living organisms per cc, liquid porton of 72-hour *T. foetus* cultures composed of buffered saline-citrate solution with 5 per cent bovine serum; the concentration was approximately 3 million living organisms per cc, and sterile trichomonad pyometra material from an infected cow with a count of 1½ million living organisms per cc. Only 3 animals became infected. Twenty guinea pigs were refractory to subcutaneous injections of *T. foetus* in pure culture while 12 guinea pigs were positive to subcutaneous injections of sterile trichomonad pyometra material from an infected cow. Abscesses remained positive for *T. foetus* up to 42 days. Thirty guinea pigs inoculated vaginally with bacteria-free trichomonad pyometra material, only 2 animals were positive for 5 days.

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Physiological observations upon a larval *Eustrongylides*. V. The behavior in abnormal warmblooded hosts. THEODOR VON BRAND and RICHARD P. CULLINAN,

Department of Biology, The Catholic University of America.

Various species of *Fundulus* quite regularly are infected with a larval *Eustrongylides* in the vicinity of Baltimore and Washington (Chapin 1926, von Brand 1938). Although positive specific determination of these worms is unfeasible it appears likely that they are the larvae of *Eustrongylides ignotus* Jägerskiöld (B. G. Chitwood, personal communication). A similar, or perhaps the same parasite was found by Hunter (1937, 1942) in various fishes of the lower Hudson area and in Connecticut, where the infection is sometimes so heavy that the fish become unmarketable. In so far as the region of Baltimore and Washington is concerned, identical worms were found by Dr. J. B. Parker (personal communication) in the eel, yellow perch, white perch, pickerel, cat fish, large mouth black bass, crappy and rockfish. In *Fundulus*, the worms occur predominantly in cysts located in the mesenteries, relatively rarely in cysts of the liver, the mesenteric fat, the peritoneum or the ovarian capsule (Cullinan, unpublished) and still more rarely in the muscles (v. Brand 1938). Dr. Parker, on the other hand, found the worms in the above mentioned fish predominantly in the muscles, an observation corresponding to those of Hunter (1937). The worm seems then, as far as the coldblooded intermediate hosts are concerned, fairly unspecific. The adults have been found by Jägerskiöld (1909) in *Botaurus* sp. and *Ardea* sp., by Chapin (1926) in *Ardea herodias* and by Cram (1933) in *Nycticorax nycticorax hoactli*. Hunter (1937) found immature adults in *Nycticorax naevius naevius* and *Butorides virescens*. In view of this relative unspecificity of hosts and in view of the ease with which these nematodes can be kept *in vitro* in a variety of media (von Brand and Simpson, 1942) it appeared worthwhile to find out how long the worms would remain alive in abnormal hosts.

The worm's cysts were isolated from infected *Fundulus heteroclitus*, and either the total cysts or the worms extracted from the cysts were transferred to experimental animals. Young ducks, chickens, medium sized rats, and a rabbit served as experimental animals. The worms were introduced either orally, subcutaneously, or intraperitoneally as discussed in turn below. The hosts were autopsied after death occurred or were sacrificed for autopsy after varying periods.

Infections per os.—For these experiments 5 8-day-old ducks, 6 12-day-old chickens, and 1 rabbit were used. Attempts to feed these worms to rats failed be-

cause the worms proved too large for swallowing. The oral feeding of parasites yielded only disappointing results. From the rabbit, autopsied after 3 days, no worm was recovered. Similarly 4 ducks, to each of which 2 worms had been fed, and 3 chickens which had received from 3 to 12 worms each proved to be negative when sacrificed 3½ hours to 6 days after infection. This is due to the fact that the worms pass rather rapidly into the gizzard where they are quickly ground to pieces. Small parts of worms were recovered after 20 minutes from the gizzard of a duck which had been fed 2 worms. A chicken fed 9 worms and sacrificed after 30 minutes yielded only 3 live worms from the crop, the others having perished in the gizzard; in another chicken a living worm was found after 15 minutes in the oesophagus. We have only one case in which orally fed worms stayed alive for any length of time in a chicken. This bird had received 16 worms and had died 18 hours later. Upon autopsy 4 living worms were found in the body cavity. They had apparently escaped the grinding action of the gizzard by boring through the wall of the fore stomach where perforations were plainly seen. The immediate cause of death of the host was severe internal bleeding brought about by the wanderings of the worms. It should be remembered that the normal hosts do not have the grinding gizzard that is characteristic of ducks and chicken; they possess rather a stomach that is soft-walled in all compartments and hence chances of parasite survival are here obviously better.

Intraperitoneal infections.—Six rats were used, each receiving 3 worms through an abdominal incision. Two rats died after 24 hours, 1 after 48 hours, death being due in all cases to hemorrhages in the body cavity brought about by at least one worm in each rat invading the liver. All parasites were recovered alive, those not in the liver were lying free in the body cavity. The 3 remaining rats were killed 7, 30 and 53 hours after infection. Here too the worms were alive, all those of the 7-hour and 2 of the 30-hour rat lying free in the body cavity. One parasite of the 30-hour rat and all of the 53-hour rat, however, had found their way into the mesenteries, where they were apparently still crawling around. They were not yet curled up and no sign of beginning cyst formation could be found.

Subcutaneous infection.—The subcutaneous infection of parasites yielded the best results. A small incision was made to permit the formation of a subcutaneous pocket large enough to hold several worms. These had been extracted from the fish aseptically using the technique described by von Brand and Simpson (1942). The incision was then closed by means of Michel clips.

Six ducks were used, 2 of them receiving 1 worm each, three 2 worms each and the last one 5 worms. These were inserted into the neck region in each case. The hosts showed no ill effect and were killed after periods varying from 24 hours to 14 days. Autopsy revealed that the worms had died quickly. Only 2 living worms were recovered, one in a duck killed after 24 hours, the other in one sacrificed 6 days after infection. The worm in the latter case was quite apparently near its death. All the other worms (3 days to 14 days after infection) were dead and in most cases partially disintegrated. All the worms were found at or near the subcutaneous pocket, in no case had they undertaken extensive migrations. The worms when found were usually not curled up but stretched out. Moreover, a marked host reaction had surrounded them with a thick walled cyst of strong connective tissue. The parasites were several inches long and the cyst enclosed them so completely that it could easily be removed from the surrounding loose subcutaneous tissue as a semi-rigid tube.

The reaction of chickens to subcutaneously introduced parasites was quite different from that of ducks. Three chickens (46 days old) were used receiving 2, 3 and 4 worms respectively in the neck region. All these chickens were found paralyzed and near death after 24 hours. Autopsy showed that the worms were alive and had undertaken considerable migrations, a few staying in the subcutaneous

tissues, others boring through the muscles. In each chicken one worm had found its way to the medulla oblongata, and in one case an additional worm had penetrated the spinal cord. A histological examination showed that the worms had actually penetrated the central nervous system, destroying entire nerve tracts and accounting thus for the clinical symptoms observed. Figure 1 shows the way in which the worms gained access to the central nervous system, they bored through the connective tissue connecting the vertebrae. Definite signs that the worms feed during their migration were found. The intestine of the worm shown in figure 1 was more or less filled with dark staining debris, whereas the intestines of worms taken from the fish cysts are usually empty or contain at most some lightly staining material.

In addition to the birds, 18 rats received 3 worms each, inserted subcutaneously at various parts of the body: near the shoulder, on the back or in the vicinity of the hip. Six of these rats died between 7 hours and 17 days after infection, 3 were killed (1 after 48 hours and 2 after 3 days) because the symptoms indicated clearly

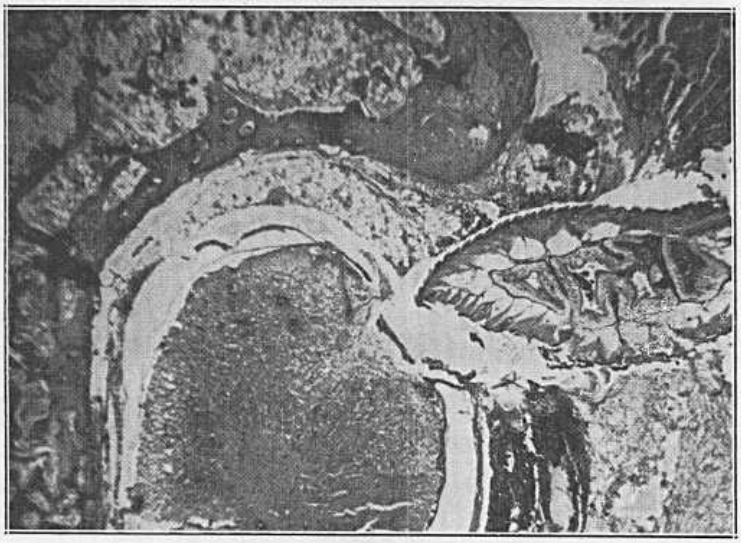

FIG. 1. Oblique section through neck region of chicken showing a *Eustrongylides* finding its way to the central nervous system. Intestine of worm contains dark staining masses indicating that it had fed during its migration. Infection 24 hours old.

that death was not far off. Penetration of one or more worms either into the liver or the chest cavity accompanied in both cases by severe internal bleedings was evidently the cause of death. It is of interest to note that in rats likewise some worms succeeded in entering the central nervous system. The worm shown in figure 2 had entered the spinal cord and was lying stretched out through a considerable part of the spinal cord. In this and another similar case the rats were completely paralyzed, whereas in 2 other cases in which the damage to the central nervous system was less extensive only parts of the body were paralyzed.

The remaining rats showed no clinical symptoms. They were killed after periods varying from 6 to 27 days after infection. It was found that the worms performed extensive migrations. One worm, for example, introduced into the subcutaneous tissue of the shoulder was recovered under the skin of the thigh. Some worms stayed under the skin, and were surrounded by a cystwall. Contrary to the findings on ducks, the worms were curled up and the cystwall was usually thin.

Other worms were recovered from the muscles, where very little tissue reaction was seen, others worked their way to the body cavity where they usually were found curled up encysted in the mesenteries. All worms recovered up to 10 days after infection were alive, later on dead ones were encountered and in one rat autopsied after 27 days all 3 worms had died and were partially disintegrated. The worms showed, with 2 exceptions, no sign of development. The exceptional cases occurred in a rat, killed after 20 days, in which 2 living worms engaged in the process of molting were

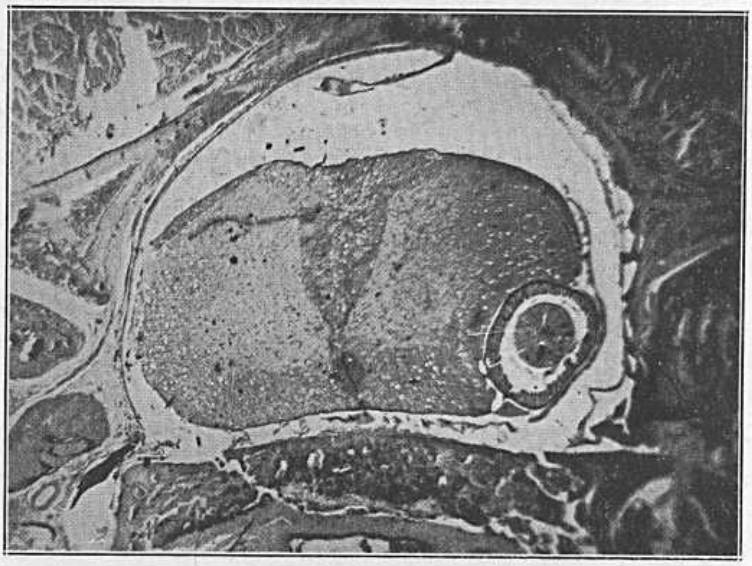

FIG. 2. Cross section through spinal cord of a rat. The worm (*Eustrongylides* sp.) was lying stretched out in spinal canal having destroyed large parts of the spinal cord. Infection 3 days old.

found in subcutaneous cysts. They had retracted from the old skin and were actively moving inside this sheath.

SUMMARY

Larval *Eustrongylides* from *Fundulus* were transplanted orally, intraperitoneally or subcutaneously into varied abnormal warm-blooded hosts. The best results were achieved with subcutaneous implantation. Ducks proved to be very resistant, the parasites soon died and were surrounded by heavy cystwalls. In young chickens the worms performed extensive migrations, some of them entering the central nervous system. Many of the infected rats were killed by damage to internal organs caused by the migrating worms. The parasites stayed alive in rats up to 20 days and few worms showed definite signs of development.

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ALBERT HASSALL
1862–1942

Dr. Albert Hassall, bibliographer, former assistant chief of the Zoological Division of the Bureau of Animal Industry, United States Department of Agriculture, and life member of the Helminthological Society of Washington, died September 18, 1942.

Dr. Hassall was born in Woolwich, Kent, February 11, 1862. He received his early education in private schools and later attended the Royal College of Veterinary Surgeons in London where he was graduated as a Member of the Royal College of Veterinary Surgeons in 1886. While in London Dr. Hassall came under the influence of the late Dr. T. Spencer Cobbold, who at that time was the most distinguished of all English parasitologists, and became one of his disciples and ardent admirers. As a student, Dr. Hassall amassed a sizable collection of helminths and this collection now forms a part of the Helminthological Collection of the United States National Museum.

Shortly after his graduation, Dr. Hassall came to the United States and in 1887 entered the government service as an employee of the then young Bureau of Animal Industry. Except for a few months in 1890–91, he served continuously until his retirement in 1932. During the greater part of his career, Dr. Hassall was a member of what is now the Zoological Division, serving first as an assistant to Dr. Cooper Curtice, and then in various capacities under Drs. Charles Wardell Stiles and B. H. Ransom, and finally as assistant chief of the Zoological Division under Dr. M. C. Hall. After his retirement in 1932 he was appointed Collaborator; this gave him official status in the Bureau and enabled him to continue his work on the Index Catalogue of Medical and Veterinary Zoology which was his brain child and chief interest. This work, published for the most part under the authorship of Stiles and Hassall, is recognized the world over as of inestimable value and is perhaps the greatest contribution to parasitological literature. In recognition of this important contribution to parasitology and veterinary medicine, Dr. Hassall was awarded the Steel Memorial Medal by his *alma mater* in 1922.

Although Dr. Hassall took little interest in societies, he was one of the early members of the Helminthological Society of Washington and served as its president in 1920–21; he was elected to life membership in 1931.

Dr. Hassall was a sincere friend and his helpfulness to the younger parasitologists who were privileged to know him will not be forgotten. His death marks the passing of another of the few early American parasitologists and the last of the early veterinary parasitologists. In his death the world has lost a distinguished scientist and the Society one of its outstanding members.

The Society extends to his family its sincerest expression of sympathy.

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