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MERMITHID NEMATODE PARASITES IN MOSQUITOES 1

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In the arctic and subarctic regions where mosquitoes are very abundant but the larvae are widely dispersed and not concentrated, chemical control methods have generally given results of limited value. In these areas natural control methods using parasites or predators would be most valuable if made practical.

Mermithid-type nematodes were discovered emerging from mosquito larvae at Churchill, Manitoba and Coral Harbor, Southhampton Island, Northwest Territories by the authors during the summer of 1950. Additional collections were made by West the following Churchill.

Nematode infested fourth-instar mosquito larvae appeared to have the thorax, or abdomen, or both, whitish and swollen. Most frequently the nematodes were observe to be coiled inside of the thorax just below the integument. They were

also coiled evenly in the first and second abdominal segments. Occasionally specimens were observed irregularly coiled in the thorax, irregularly coiled in the thorax and abdomen, or entirely within the abdomen, doubled back several times. Infested mosquito larvae were usually slower moving and frequently were covered with a dense mass of epizootic protozoans (Vorticella-type) and algae. Such growth was also commonly found on larvae uninfested by nematodes, but it occurred in smaller quantities.

Larvae of Aedes communis (DeGeer) were the most frequently infested. two pools at Churchill in 1950 up to 100 per cent of the late developing larvae were infested and died. No larvae of A. excrucians (Walker) in these same pools were observed to be infested. Parasitization was most severe during the latter part of the larval season in early July. Nematodes in A. communis larvae were collected on 18 June, and from 5 to 18 July, 1950, and from 19 June to 4 July, 1951. An infested larva of A. nearcticus Dyar was collected at Coral Harbor, Southhampton Island on 3 July, 1950. An infested larva of A. nigripes Zett. was collected at Churchill in June, 1951.

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The nematodes normally emerge from he larval thorax by breaking through he side or bottom of the integument, eaving a large cavity. Partial emergence of the worms through the anus and hrough the air tube were observed, but both the worms and larvae died. nfested mosquito larvae pupated.

While the nematodes are emerging he larvae writhe and exhibit spasmodic novements and jerks. After emergence of the nematode, the larvae continue jerkng motions and die almost immediately or within a few hours. One larva was observed to die after 2.5 hours.

There is usually one nematode per mosjuito larva, but multiple infestations are common. In 1951 a sample of 84 infested nosquito larvae (A. communis) was placed n a beaker and 34 died without emergence of the parasites. From the 50 emaining larvae, 167 worms emerged, veraging 3.3 per larva. Larvae have been ollected containing 1, 2, 3, or 4 nemaodes. In the mosquito larvae when only one worm occurred it usually developed nto a female, and when more than one occurred more males developed.

A series of nematodes, together with hosts, was preserved in alcohol for identi-Several hundred nematodes rom field collected larvae have been observed in the laboratory. Worms were out in rearing beakers containing water or water and sand or organic materials. A few nematodes were put in water with Daphnia. Other nematodes were allowed o emerge in containers with water and and and thus were not subjected to

handling.

After leaving the host or being placed in container, the nematode moved to the pottom and entwined around bits of debris or formed an entangled mass with other worms. When the substrate was sand the worm usually anchored itself at one end and slowly moved the anterior (presumably) end through the water in a whip-like motion.

After about a month at room temperature all worms died and decomposed. Where sand was furnished nearly all worms burrowed into the sand during a period of two to three weeks and rested in a coiled position. When containers with worms were held in a refrigerator at approximately 6° C., the same behavior was observed except that a few individuals remained active on the surface of the sand for three to four months and resting, coiled individuals did not decompose until after three to four months.

139

No eggs were discovered in any container. The contents of some containers were allowed to dry at room temperature and the water level was maintained in other containers in a refrigerator and at room temperature. At intervals after eight to twelve months from time of emergence from the host, water and A. aegypti (L.) larvae were added to dry sand and wet sand was placed in A. aegypti rearing pans. No pre-parasitic larvae were observed and no nematode infestation of mosquito larvae occurred.

Beginning with the 1952 season the study of this mosquito larval parasite was turned over to the Laboratory of Entomology, Science Service, Canada Department of Agriculture, Belleville, Ontario. Field observations and attempts at laboratory rearing are being continued.

The writers interpreted the emerging worms from mosquito larvae to be immature. This has been confirmed by Dr. G. Steiner, Division of Nematology, U. S. D. A., Beltsville, Md., to whom preserved material was submitted for identification. According to Dr. Steiner (1951) the parasite belongs to the nematode family Mermithidae and is a new species of the genus Hydromermis which he is describing. One of the specimens is a pre-adult female and exhibits a vulva opening. These worms have only one host. velopment to the adult stage must then be completed as a free-living form. In nature the adult worms may be found under stones and plant debris or in the mud.

Parasitic nematodes were observed by Freeman (1950) to be the chief enemy of mosquito larvae at Frobisher Bay, Baffin Island in 1948.

"The worms were found in any type of pool and infested up to sixty per cent of the larvae in some pools. The worms lie coiled under the thoracic cuticle and form four rings around the oesophagus. This causes the thorax of the larva to appear swollen and the larva appears superficially to have a white-ringed thorax. Most of the parasites had completed their development within the mosquito larvae by June They emerge from the larva at the side of the thorax and the larva dies immediately after the worm emerges. These worms appear to emerge simultaneously and congregate in little knots or groups at the bottom of the pools where they later bore into sand, gravel, etc."

In French Guiana, nematodes were found in pairs, one large and one small worm, in the body cavity of A. aegypti by Gendre (1909). The larvae developed normally but just before pupation the worms left the host at the posterior end of the body by perforating the membrane surrounding the anus. The larger worm emerged first followed by the smaller and both died several hours later. The mosquito larvae died as a result of injuries. The parasitized larvae were collected from water in cavities in trees. No parasitized adults were found.

The life cycle of the mermithids is known to be reversed from that of the usual life cycle of parasitic nematodes. In Mermis and other genera the larvae are parasitic in insects, while the sexual stages are free living in soil or water (Borradaile et al., 1935, p. 256.). According to Muspratt (1945) nematodes other than Filaria, parasitic in mosquitoes have been recorded from the United States, Canada, Europe, Russia, India, Ceylon, Sumatra, These mermithids were Limnomermis Daday, Paramermis Linst, Mermis Duj. and Agamomermis Stiles. The latter is an artificial collective group for larvae which cannot be identified.

In Pennsylvania, larvae of Aedes vexans Meigen and Culex salinarius Coq. were found by Stabler (1945) on 11 August to be parasitized by a mermithid nematode. Up to 100 per cent of some collec-

tions of A. vexans larvae were found to be parasitized and all infested larvae died after emergence of the worm. Infested C. salinarius Coq. larvae were found in the same collections (Stabler, 1951). Of 217 larvae 90 were infested. The measurements of the worms ranged from o mm. to 18 mm. averaging 14.7 mm. Sections of the thoraces of the infested larvae showed that the worms lay coiled and twisted in the haemocoele just under the exoskeleton and encircling the internal thoracic structure. No gross anatomical changes were correlated with the presence of the parasite internally. One larva contained three small worms, the others only one.

The life cycle of the mermithids that destroy mosquito larvae was studied in India by Iyengar (1927). He found that the minute pre-parasitic worms swim in the water and probably penetrate the cuticle of the mosquito larvae. The smallest nematodes seen in the mosquito larval haemocoele were 650 to 700µ in length and 115 to 165µ in width. The parasitized mosquito larvae were found to remain as 4th instar longer than normal larvae. The larvae died on emergence of the nematodes. Muspratt (1945) found that both sexes and one or more individuals may parasitize a single mosquito larvae. The nematodes enlarged after emerging.

About 100 nematode larvae which had just emerged from mosquito larvae were put in a jar containing sand and water (Muspratt, 1947). The larvae burrowed into the sand and became dormant after a few days. The water was decanted and the sand was kept moist for 11-12 months. Some sand was then transferred into a jar containing over 100 A. aegypti larvae. Within 24 hours some of these contained small nematodes in the haemocoele and pre-parasitic larvae less than 1 mm. in length were seen swimming in the water. After 70–80 per cent were infected, new mosquito larvae were exposed. Nematode eggs hatched during 2 to 3 weeks. nematodes emerged from the mosquito larvae after 10 to 14 days. Apparently the nematodes emerging from mosquito larvae mature and lay eggs in sand and require 11 or 12 months before hatching, although the similar mermithid studied by Iyengar (1927) had a much shorter cycle.

The methods of dispersal of these nematodes is at present unknown, but it is of interest to note their presence in water in tree holes as well as in temporary vernal ground pools throughout the world.

A number of observers have reported nematode parasitism of adult mosquitoes as well as in the immature stages.

At Leipzig, Germany, Agamomermis sp. were collected in the abdominal cavity of larvae, pupae, and adults of Culex nemoralis by Stiles (1903). Infestation was thought to have occurred in the water. The infested insects were very sluggish in movement and many of them died from the effects of the parasite. The

ovaries of parasitized females were underdeveloped and during the years when the nematodes were most common the mosquitoes were less numerous.

In New Jersey, Smith (1904) collected large numbers of female Aedes sollicitans (Walker) which were infested with Agamomermis culicis Stiles (1903) from 25 June to 25 September, 1903. Up to 50 per cent of the specimens collected contained worms. The peak of infection occurred in late July and into August. The ovaries of A. sollicitans did not develop when the female was parasitized and the nematode was considered to be a material check on this species. Parasitized adults were collected far inland from the coast. The presence of the parasite did not prevent the females from feeding.

The species of mosquitoes recorded to be infested with mermithids include:

Mosquito	Stage	Locality	Authority
Aedes aegypti (L.)	L	Africa	Gendre (1909), Muspratt (1945)
A. aldrichii D. and K.		3.7 .1 A	Hearle (reas)
(= A. sticticus (Meig))	A	North America	Hearle (1929)
A. calceatus Edw.	L	Africa	Muspratt (1945)
A. cantans Meig.		n :	Shakhov (1927)
(≡ 1. stimulans (Walker)?)	A	Russia	Jenkins and West
A. communis (DeG.)	L	North America	
1. dorsalis (Meig.)		Russia	Shakhov (1927)
A. flavescens (Müller)	A	North America	Hearle (1929)
A. fulgens Edw.	<u>L</u>	Africa	Muspratt (1945)
A. haworthi Edw.	L	Africa	Muspratt (1945)
A. marshalli Theo.	L	Africa	Muspratt (1945)
A. metallicus Edw.	L	Africa	Muspratt (1945)
A. nearcticus Dyar	L	North America	Jenkins and West
A. nemorosus Meig.		_	0.11 ()
(= A. communis (DeG.))	•	Europe	Stiles (1903)
A. nigripes Zett.	L	North America	West
A. sollicitans (Walker)	A	North America	Stiles (1903)
A. zethus DeMeill	Γ .	Africa	Muspratt (1945)
A. vexans Meig.	L	North America	Stabler (1945)
		North America	Steiner (1924)
Anopheles gambiae Giles	L	Africa	Muspratt (1945)
A. leucosphyrus Dön	L	Sumatra	Walandouw (1934)
A. rufipes Gough	L	Africa	Muspratt (1945)
A. spp.	L .	India	Iyengar (1927)
Culex nebulosus Theo.	L	Africa	Muspratt (1945)
C. nemoralis Stiles	L, P, A	Europe	Stiles (1903)
(probably = nemorosus)			
$\equiv A.$ communis (DeG.))			
C. pipiens L.	L	North America	Stabler (1951)
C. salinarius Coq.	L	North America	Stabler (1945)
C. fatigans Wied.		India	Ross (1898)

Hearle (1926) reported that as many as 80 per cent of Aedes vexans Meig. mosquitoes in the Fraser Valley, British Columbia in 1920, contained a parasitic nematode, Paramermis canadensis Steiner (1924), but only about 20 per cent in 1921. The effect of the parasite was retardation of the development of the ovaries, since no parasitized females contained well-developed eggs. The largest worm measured 1.25 in. in length, and there were from one to six present in a single mosquito. A single female Aedes aldrichii D. and K. was found parasitized but no effect on egg development was apparent. It was stated that there was little doubt that the parasite considerably reduced the numbers of A. vexans. Hearle (1929) found an adult female Aedes flavescens (Müller) in Canada to be parasitized by a single small nematode.

Bates (1949) refers to mosquitoes being hosts during part of the life cycle of numerous filarial worms and reports that the filaria often cause considerable injury to their mosquito hosts. He also states that many mermithid nematodes have been reported from mosquitoes and that little is known concerning their biology. A number of references on the occurrence of filaria and mermithids in mosquitoes have been compiled by Speer (1927).

Steiner (1924) states that "many observations and facts support the theory that the members of the nema family of Mermithidae (which includes a series of different genera with at least several hundred species), play a much more important role than might have been expected in the control of many kinds of insects."

SUMMARY

Parasitic mermithid nematodes were discovered infesting the larvae of three abundant species of arctic mosquitoes. All of the infested mosquito larvae died and up to 100 per cent infestation of Aedes communis was observed in one pool. Observations were made of the habits of the nematodes and rearing was attempted.

It appears that mermithid nematodes

offer an effective means of natural contro if they can be reared in large numbers. This subject deserves detailed investigation, particularly the development of suitable method of rearing and disseminating the parasite.

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143

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THE IDENTIFICATION OF THE LARVA OF *PSOROPHORA* (*GRABHAMIA*) *SIGNIPENNIS* (COQUILLETT)

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Introduction. The usual published description of the fourth instar larva of P. signipennis states that the upper and lower head hairs are single. The writer has observed that a rather large percentage of specimens of this species collected in the southwest exhibit a varying number of branches of these head hairs. To the knowledge of the writer all but two of the previously published descriptions of this species state that the upper and lower head hairs are single, and these characters are usually used as a major point of differentiation of the species found in the United States. (Howard, et al., 1912; Dyar, 1928; Carpenter, 1941; King, et al., 1944; Matheson, 1944; Randolph and O'Neil, 1944; Carpenter, et al., 1946; Rozeboom (1942) states Pratt, 1946). that the lower and upper head hairs are either single or double and uses other characters to distinguish the larva of this species. Yamaguti and LaCasse (1951) state that the lower head hairs are one to three forked and the upper head hairs are single.

THE PRESENT STUDY. The writer has examined over 400 specimens of this species from various localities in the southwest and 29.9 per cent of these specimens had at least one of the head hairs double or triple. This variation in number of branches of the head hairs occurs in both the upper and lower hairs with equal regularity. Some of the specimens examined had one hair double or triple, some two, others three, and less than one per cent of the total 29.9 per cent had all hairs not single. The following briefly summarizes the head hair variation observed.

It should be noted that many specimens examined varied from the usual published description in other characters, such as the number of comb scales and the number of pecten teeth. Larvae having as few as four and as many as eight comb scales and as few as two and as many as eight pecten

Locality	No. examined	No. with head Hair variation	Percent
Austin, Texas Palmetto State Park, Texas Sonora, Texas Fort Sill, Oklahoma Camp Bullis, Texas Fort Hood, Texas Total	144 10 16 50 160 50	22 4 2 24 25 24 101	15.3 40 12.5 48 15.6 48
10.00			cluded in the ab

(Collections examined which consisted of less than ten specimens were not included in the above.)