

# THE INFLUENCE OF HOST STAGE AND SEX UPON THE SIZE AND COMPOSITION OF THE THELASTOMATIDS PARASITIC IN THE HINDGUT OF *PERIPLANETA AMERICANA* L. AND *NEOSTYLOPYGA RHOMBIFOLIA* (STOLL)

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## INTRODUCTION

THE COCKROACHES are an ancient and highly successful form of insect life. They have been in existence since Pennsylvanian times and the fossil record indicates that in general body form they have remained a very stable group (Moore *et al.*, in Cochran *et al.*, 1975). About 3500 named species of living cockroaches have been catalogued (Princis, in Cochran *et al.*, 1975; Rehn in Roth and Willis, 1960) but additional species are being found and named continually, so that the final number probably will far exceed the number of 4000 suggested by James and Harwood (in Cochran *et al.*, 1975).

Rehn (1945) has pointed out that the majority of cockroach species are not domiciliary pests. In fact, the pest species constitute less than 1% of all cockroaches.

The medical importance of cockroaches is much greater than generally realised as they have been shown to harbour pathogenic bacteria, serve as intermediate hosts for pathogenic helminthes and to carry helminth eggs, viruses, protozoa and fungi affecting man and other vertebrate animals (Roth and Willis, 1957; Roth and Willis, 1960); Tarshis, Cornwell, James and Harwood, Rueger and Olson, Pulner and Sarchenko, in Cochran *et al.*, 1975). Sixteen species of cockroaches are considered vectors of pathogenic organisms affecting man (Roth and Willis, 1975a). It has been identified that cockroaches harbour about 80 species of Aschelminthes, 5 species of Acanthocephala (Guthrie and Tindall, 1968). Cockroaches have been shown to be natural intermediate hosts for 12 species of

Helminthes and intermediate hosts experimentally for 11 other species (Cornwell, 1968). Acholomu and Finn (in Cochran, 1975), Khairul Anuar (1976) and Schaefer (1970) have also shown that *Periplaneta americana* is a natural intermediate host of *Moniliformis moniliformis* in Puerto Rico, Penang and Honolulu respectively.

Parasites of cockroaches have been reported from throughout the world (Chitwood, 1932; Macadow, Semans, Armen, Mac Kinnon, Hoyte, Jenkins in Yuan and Kevin Cahill, 1970 and Guthrie and Tindall, 1968). There have been no studies of the parasites, particularly nematodes of the cockroach, *Neostylopyga rhombifolia* (Stoll) but there have been a few studies on the common american cockroach *Periplaneta americana* L. in Penang (Khairul Anuar, A. and Paran, T. P., 1976, 1977). Thus, the study is of importance especially in Asia and other neighbouring countries where people work, eat and sleep with cockroaches. Their habit of feeding on both human faeces and human food is an example of their potential health hazard to man. The importance of this point becomes clearer when it is realised that cockroaches move freely from building to building or from sewer or privy to human habitations. The present study is devoted mainly to find the frequency occurrence of the parasites belonging to the family Thelastomatidae, super family Oxyuroidea (Chitwood, 1932) and also the worm load of those parasites in *Periplaneta americana* and *Neostylopyga rhombifolia*. These nematodes, the oxyurids, are parasites in the hindgut of cockroaches (Welch and Jarry in Poinar, 1975; Hominick and Davey 1972; Basir 1956;

Chitwood, 1932). The influence of host stage and sex upon the size and composition of the worms are also discussed.

Parasites encountered in the study are *Hammer-schmidtella diesingi* (Hammerschmidt, 1838), *Thelastoma malaysiense* (Khairul Anuar, 1977), *Leidynema appendiculata* (Leidy, 1850) and *Severianoia severianoii* (Schwenke, 1926; Travassos, 1929). Under present study *Severianoia severianoii* was only found in *Neostylopyga rhombifolia*. *Thelastoma malaysiense* is a new species (Khairul Anuar, 1977) and was shown to be harboured by the common american cockroach, *Periplaneta americana*, in Penang. *Hammer-schmidtella diesingi* and *Leidynema appendiculata* were also shown to be harboured by *Periplaneta americana*. They were previously shown by Khairul Anuar (1977), Hominick and Davey (1972).

### Relationship between Oxyurids, Cockroaches and Vertebrates

Parasites of the family Thelastomatidae live in the hind gut of cockroaches, the lining of which is lost at ecdysis. They not only survive the hazard but take advantage of the fact that the exuviae can be contaminated with eggs and may be eaten (Crofton, 1966). Nematodes are capable of developing forms which can survive the hazard imposed by repeated moultings. Peregrine (1974), had indicated that the parasite *Thelastoma attenuatum* can respond to unfavourable conditions within the host by producing its resistant stage, in this case the egg. This would suggest a fairly high degree of host-parasite specificity.

Galeb (in Cochran *et al.*, 1975) had shown experimentally that oxyurids eat the same food as the host insect and that if one starves them by withholding food from the host, the oxyurids die and disappear, these worms are not parasites but commensals. Experiments also showed that *Thelastoma* appears to feed indiscriminately on host hindgut contents and it is also capable of digesting its own food, hence not relying on the host for predigested material (Lee, 1958). Poinar (1975) suggested that the parasites of this family are found in those insects having a slow rate of food movement and a well developed microbial flora. It has not been determined if their nourishment consists of partially digested food or microbial agents in the host's gut or both. None have been cultured as artificial media and their association with the host appears to be an obligate one. Although generally considered as parasites their feeding habits also qualify them as commensals. Dobrovolny and Ackert (1934) stated that "all observations seemed to indicate that the health fertility and activity of the heavily infected cockroaches were comparable with those of non-

parasitised specimens". Thus, they are innocuous to their hosts and only Rostom and Taylor (in Poinar, 1975) described instances where pinworms apparently damaged the peritrophic membrane and gut wall of their hosts.

Nutritional studies show that entomogenous nematodes have several methods of absorbing nutrients in the hosts body cavity. These include normal oral uptake, absorption through the cuticle which may or may not be aided by microvillarlike processes and uptake by phagocytosis.

There is no indisputable report of an obligate insect parasitic nematodes obtaining nourishment from vertebrates or plants. There are reports of mermithid nematodes occurring in man and six such cases have been reviewed by Foster (in Poinar, 1975). Entomogenous nematodes could enter vertebrates in drinking water or in food containing parasitised insects, however, in such cases they probably would be killed and expelled through the rectum. Up to present study, entomogenous nematodes present no serious threat to plants or animals.

### MATERIALS AND METHODS

The host, *Periplaneta americana* L. was caught in large numbers at night using a special trap (PAT.P. Japan, Type DS. 12) (Khairul Anuar and Paran, 1976, 1977). The other host, *Neostylopyga rhombifolia* (Stoll), was caught by hand since it was rarely found in large numbers. The cockroaches, adults and nymphs collected from all parts of campus were identified into stages and sexes. The numbers of cockroaches caught are shown below:-

Sex and stage	<i>Periplaneta americana</i> L.	<i>Neostylopyga rhombifolia</i> (Stoll)
Female adult	50	20
Female nymph	50	20
Male adult	50	20
Male nymph	50	20

The host were anaesthetized with ether and dissected immediately for parasites in saline (6.9%) to avoid rupturing the nematodes. The gut was removed and transferred into a cavity block of saline. The gut was then teased open in saline with fine needles and was examined under a dissecting microscope with transmitted light. The female nematodes were picked with fine pipettes and they were classi-

ficd to different species with aid of the compound microscope. The number belonging to each species was also noted. Measurements were also made. Drawings were made with the use of slide projection.

The number of adult females of nematodes was used as an index of population of the three species in a particular hindgut. This was because of the extreme variation in numbers of juvenile nematodes (Hominick and Davey, 1972).

## RESULT

### *Periplaneta americana* L.

The overall infection rate of nematode infection among male and female adults and nymphs is given in Table I. The percentage of adult female hosts infected by nematodes was the highest (96%). This was followed by adult male hosts, 94% and 86% in the case of both male and female nymph hosts. The adults were more highly infected compared to the nymphs and the overall infection was found to be 90.5%.

Three species of nematodes were found to be associated with 100 *Periplaneta americana* dissected. They were *Hammerschmidtella diesingi*, *Leidynema appendiculata*, and *Thelastoma malaysiense*. The infection by *Hammerschmidtella diesingi* was found to be highest in adult females (60%). When stages and sexes of roaches were ignored, the percentage parasitism by *Hammerschmidtella diesingi* was 56%. There was no significant difference in the degree of infection among male and female and among adult and nymph roaches. The degree of parasitism by *Leidynema appendiculata* was found to be highest in immature roaches (32%) but there was no significant difference in the degree of infection among adults and nymphs of *Periplaneta americana*. Adult female and immature roaches were highly infected by *Hammerschmidtella diesingi* and *Thelastoma malaysiense* and least infected by *Leidynema appendiculata*. This is shown in Table II.

However, the host carried more than one species of nematodes. In the case of female adult roaches, the percentage of infection by only *Hammerschmidtella diesingi*, *Leidynema appendiculata* and *Thelastoma malaysiense* were found to be 16%, 12%, 22% respectively; in male adults, 14%, 10%, 28% respectively; and in female and male nymphs 10%, 8% respectively. Single infection by *Hammerschmidtella diesingi* was found to be highest in female adults (16%) and the single infection by *Thelastoma malaysiense* was found to be highest in male adults (28%). There was almost the same degree of infection by *Leidynema appendiculata* in all hosts. All these are tabulated in Table III. There was no difference in the degree of single infection by the 3 worms in all nymphs of both sexes but there was some small difference in both adult and nymph hosts.

Double infection by nematodes was quite common. The infection by both *Hammerschmidtella diesingi* with *Thelastoma malaysiense* was found to be the highest and that by *Hammerschmidtella diesingi* with *Leidynema appendiculata* showed little variation. Double infection was found to be much higher in the nymphs.

Triple infection was shown to occur only in adult hosts, 8% in female adults and 6% in male adults. This forms a low infection.

Table IV gives the frequency distribution of number of species of nematodes. For adults of both sexes, 1 species per host was the commonest whereas in nymphs of both sexes, 2 species per host was the commonest. Three species per host did occur in the adults but never in the nymphs.

### *Neostylopyga rhombifolia* (Stoll)

Table V gives an overall infection rate of nematode infection among adults and nymphs of *Neostylopyga rhombifolia*. It is clearly shown that the adults were more highly infected compared to the nymphs.

Table I

The overall infection rate of nematode infection among male adults, female adults and nymphs of *Periplaneta americana* L.

	Examined			Positive			% Infected		
	Male	Female	Total	Male	Female	Total	Male	Female	Average
Adult	50	50	100	47	48	95	94	96	95
Nymph	50	50	100	43	43	86	86	86	86

The infection rate in adult female and adult male and immature roaches did not differ significantly. ( $P \leq 0.050$  by Chi-square test)

**Table II**  
**Nematodes in 200 *Periplaneta americana* L.**

	Adult male roaches			Adult female roaches			Immature roaches			Total	
	Total no. Examined	No. with Parasite	% with Parasite	Total no. Examined	No. with Parasite	% with Parasite	Total no. Examined	No. with Parasite	% with Parasite	No. with Parasite	% with Parasite
<i>H. diesingi</i> *	50	26	52	50	30	60	100	56	56	112	56
<i>T. malaysiense</i> *	50	33	66	50	30	60	100	58	58	121	60.5
<i>L. appendiculata</i> **	50	12	24	50	15	30	100	32	32	59	29.5

\* The infection rate in adult male and female, and immature roaches did not differ significantly. ( $P \leq 0.05$ , by Chi-square test).

\*\* The infection rate in adult male and female and immature roaches differed significantly. ( $P \leq 0.95$ , by Chi-square test).

Table III

Percentage *Periplaneta americana* L. with single infection, double infection and triple infection of adult female nematodes

	No. of Host Examined	only H	only L	only T	H + L	H + T	T + L	H + L + T
Female adult	50	16	12	22	8	28	2	8
Male Adult	50	14	10	28	4	28	4	6
Female nymph	50	10	8	8	10	36	14	0
Male nymph	50	10	8	8	10	36	14	0
Adult	100	30	22	50	12	56	6	14
Nymph	100	20	16	16	20	72	28	0

Table IV

Frequency distribution of number of species of nematodes

No. of species per host	Female		Male		Adult	Nymph
	Adult	Nymph	Adult	Nymph		
0	2	7	3	7	5	14
1	25	13	26	13	51	26
2	19	30	18	30	37	60
3	4	0	3	0	7	0

Table V

The overall infection rate of nematode infection among male adults, female adults and nymphs of *Noestylomyia rhombifolia* (Stoll).

	Examined			Positive			% Infected i		
	Male	Female	Total	Male	Female	Total	Male	Female	Average
Adult	20	20	40	18	19	37	90	95	92.5
Nymph	20	20	40	6	8	14	30	40	35.0

The infection rate in adult females and adult males did not differ significantly but differed significantly in nymph females and nymph males. ( $P < 0.005$ , by Chi-square test).

The infection rate in adults and nymphs differed significantly ( $P < 0.025$ , by Chi-square test).

*Leidynema appendiculata* was the most predominant nematode in all stages and sexes of the hosts. The percentage of parasitism in both male and female adults was found to be 60% and 20% in the case of immature ones. The second degree of infection was that by *Thelastoma malaysiense* in adults of both sexes and *Hammerschmidtella diesingi* in immature roaches. *Severianoia severianoii* was uncommon in the adults of both sexes but it was found to parasitise immature roaches (12.5%). The overall infection was highest by *Leidynema appendiculata* (40%).

The infection by *Thelastoma malaysiense* and *Leidynema appendiculata* were found to be high in adults and low in nymphs but that by *Severianoia severianoii* was found to be the reverse.

Single infection was found to be highest by *Leidynema appendiculata* in all stages and sexes of the roaches. For double infection, adults were more highly infected by *Hammerschmidtella diesingi* with *Leidynema appendiculata* and *Thelastoma malaysiense* with *Leidynema appendiculata*, whereas, for

nymphs by *Hammerschmidtella diesingi* with *Leidy-nema appendiculata*.

Table VIII gives the frequency distribution of the number of species of nematodes present. Most adults had a species per host whilst most nymphs had 0 species per host. Triple infection only occurred in adult females and 4 species per host was uncommon in any of the hosts dissected.

### DISCUSSION

The results point out clearly that the parasites that were harboured by the two hosts were the same, the exception being in the case of *Severianoia severiano* which was only found in *Neostylopyga rhombifolia*. Thus, we can say that the worms form the common parasites of local (Penang) cockroaches.

Males and females are not equally susceptible to parasites (Hominick and Davey, 1972). It is clearly shown that females were more highly infected

than males. Dobrovolsky and Ackert (1934), Bozeman (1942) and Gordon (1970) working with various Thelatomatids and cockroaches also reported that the level of infestation was highest in adult female hosts. This is different from many known facts that males are more highly infected with parasites. Berry (1962) studied trematodes *Cercaria ubiquitousoides* of marine snails, *Littorina saxatilis*. There seemed to be greater numbers of infected female than infected male hosts. The reason given was that higher mortality occurred in the male and more males die young while more females survive longer. This explains greater accumulation of trematodes in females. The same argument can be applied to that of cockroaches under study. But, the longevity of both male and female of cockroaches is not studied, presumably female longer.

Hominick and Davey (1972) suggested that host nutrition may play an important part in the differences in parasitism of males and females. Though all stages of the hosts may take food from the same source, quantitative and even qualitative differences

Table VII

Percentage *Neostylopyga rhombifolia* (Stoll) with single infection, double infection and triple infection of adult female nematodes.

	No. of host Examined	% infected with								
		only H	only L	only T	only S	H + L	H + T	T + L	L + S	H + L + T
Female adult	20	10	30	20	0	10	5	15	0	5
Male adult	20	15	20	15	0	15	0	25	0	0
Female nymph	20	5	10	5	5	10	0	0	5	0
Male nymph	20	0	10	0	15	5	0	0	0	0
Adult	40	25	50	35	0	25	5	40	0	5
Nymph	40	5	20	5	20	15	0	0	5	0

Table VIII

Frequency distribution of number of species of nematodes

No. of species per host	Female		Adult		Adult	
	Adult	Nymph	Adult	Nymph	Adult	Nymph
0	1	12	2	14	3	26
1	12	5	10	5	22	10
2	6	3	8	1	14	4
3	1	0	0	0	1	0
4	0	0	0	0	0	0

Note: Although the percentage of parasitism often gives a rough estimate of the incidence of parasitism, it can be erroneous



in food intake may exist. Wharten *et al.* (1965) suggested that adult female cockroaches have a greater preference for cellulose than do nymphs or adult males. This availability of cellulose may play a crucial role in determining the numbers of worms.

There was comparatively higher infection rate in adults than in nymphs. This is probably due to the fact that most of the worms that had infected the hosts can withstand the molting events in cockroaches except *Leidynema appendiculata* in *Periplaneta americana*. Besides that, it is a well known fact that molt leaves at least a part, if not all, of the parasite burden intact (Leibersperger, in Jarry, 1964). Jarry (1964) used this evidence to explain the cumulative infestation that allows the worms to attain high numbers in adult cockroaches. Also, it is probably that most of the worms are able to survive osmotic stress that may occur in the hindgut of molting cockroaches.

Some nematodes have been shown to produce their own neurosecretory factor(s), others are able to utilise an exogenous source of insect hormones in controlling their developmental processes. Nadakal and Nayar (1968) suggested that the corpus allatum hormone of *Periplaneta americana* may affect the fecundity of three species of oxyurid nematodes but their evidence is inconclusive. *Hammerschmidtella diesingi* depends upon a supply of neurosecretory material from the median neurosecretory cells (M.N.C.) of *Blatta orientalis* for maximum development (Gordon, 1968). Gordon (1969) had also suggested that it is the host's M.N.C. - corpora cardiaca complex which is essential to the nematode for successful parasitism. Thus, we can conclude that the lack of host neurosecretory material ('activation hormone') directly or indirectly caused either a partial destruction and/or elimination of adult nematodes from the insect or prevented the parasite larvae from developing into adult nematodes. This explains the lower number of worms found in nymphs.

In *Neostylopyga rhombifolia*, there was a significant difference in the degree of infection of *Leidynema appendiculata* among the adults and nymphs.

In *Neostylopyga rhombifolia*, there was a significant difference in the degree of infection of *Leidynema appendiculata* among the adults and nymphs. More occurred in adult roaches. This is probably because most juveniles of this Thelastomatid do not survive the complex series of events involved in molting of the host. Lee (1960) and Khairul Anuar (1977), Hominick and Davey (1972) also observed that *Leidynema appendiculata* survived the molt in *Blatta orientalis* and *Periplaneta americana*

respectively. Comparing between the adult and nymphal stages of *Periplaneta americana*, *Leidynema appendiculata* was more common in the immature ones but there was no significant difference. This contradicts with the well known fact that the number of *Leidynema appendiculata* are low in nymphs (Hominick and Davey, 1972). This is probably due to some worms which take advantage of the fact that the exuviae can be contaminated with eggs and may be eaten during early instars, when they do not go far to search for food. This could also be due to accidental result of small samples. It is a well known fact that many nematodes belonging to the family Thelastomatidae lay their eggs in strings or in clumps of two or three (Basir, 1956). As a result of this cockroach ingests more than one egg at one time, but neither the laid eggs, nor eggs removed from the uterus of *Hammerschmidtella diesingi* and *Leidynema appendiculata* possess a sticky coat (Lee, 1960). In the rectum of cockroach, the eggs of *Leidynema* are usually at the multi-celled stage. Hominick (1974), observed that under some circumstances, the eggs of *Leidynema appendiculata* may undergo precocious development to the infective stage in the uterus of the mother. Hence, there is possibility of autoinfection. If this infection takes place during the host's nymphal stage, more worms will be present. The potential significance of the above fact is difficult to explain because there is no information on how frequently cockroaches with infective nematode eggs in their hindguts occur in the population under study and how frequently it occurs during the nymphal stage of the cockroaches.

Single infection by *Leidynema appendiculata* was found to be higher in adults than in the nymphs of both species of hosts and single infection formed the highest percentage compared to its double and triple infections. Besides the fact that it cannot withstand moulting, it can only survive well when it does not occur with any other species of worms, that is, when there is competition for food, space, etc. In *Periplaneta americana* slight variation was found in its nymphal stage. Hominick and Davey (1972) had also pointed out that *Leidynema appendiculata* is not able to withstand competition. The number of *Leidynema appendiculata* is low in nymphs and high in adults because in its nymphal stage, it is faced with competition with *Hammerschmidtella diesingi* which is predominant.

In *Neostylopyga rhombifolia*, *Thelastoma malay-siense* had the same fate as that of *Leidynema appendiculata*. The infection rate in adult males and adult females did not differ significantly, but it differed significantly in adults and nymphs. In *Periplaneta americana*, there was no significant difference in the degree of infection among adult



males and adult females and immature roaches. This is probably due to the fact that *Thelastoma malaysiense* is unable to survive the complex series of events that are occurring during moulting.

There was no significant difference in the degree of infection by *Hammerschmidtella diesingi* among adults and nymphs of both hosts. The results show that it can withstand competition with *Leidynema appendiculata* which is predominant in the adults. This is in contrast to a fact given by Hominick and Davey (1972) that *Hammerschmidtella diesingi* is unable to withstand competition with *Leidynema appendiculata*. Hominick and Davey (1972) had shown that *Hammerschmidtella diesingi* infested nymphs cumulatively and its numbers decrease in adults, possibly, because the juveniles and even adults are not able to withstand competition. Galeb (in Hominick and Davey, 1972) stated that *Hammerschmidtella diesingi* by its shape and enormous fecundity annihilates *Leidynema appendiculata*, so that the former increases in number and the latter becomes more and more rare.

Results also show that adult females were more highly infected by *Hammerschmidtella diesingi* than the adult males. This is probably due to reasons given earlier as why females were more highly infected than males. There was no significant difference in the degree of infection among male and female roaches.

*Severianoia severiano* is a rare species and was found to harbour by only *Neostylopyga rhombifolia*. It was never occurred in the adults and the degree of parasitism was only 12.5%. This is probably due to the fact that it has a very low fecundity, that it reaches its sexual maturity very early such that it is never survived until hosts' adult stage. The other possible reason is that the worms might have a short life-cycle, too short to compare with hosts' life-cycle or longevity. As stated earlier the nutrients taken by adults and nymphs might be slightly different. If these worms depend on certain food taken by the nymphs but not eaten by the adults, the worms will not survive in the latter. Other than that, a very low tolerance to osmotic changes that occur during moultings can be used to explain this fact. The observation that it was never found in *Periplaneta americana* is interesting. This forms the difference in the suitability of hosts for *Severianoia severiano*.

#### SUMMARY

*Periplaneta americana* L. was shown to harbour a maximum of three species of Thelastomatidae, namely, *Thelastoma malaysiense*, *Leidynema appendi-*

*culata* and *Hammerschmidtella diesingi*. Besides these three worms, *Neostylopyga rhombifolia* (Stoll) was found to harbour *Severianoia severiano*. The infection by *Leidynema appendiculata* was generally more prevalent in the adult hosts (especially *Neostylopyga rhombifolia*) whereas the infection by *Hammerschmidtella diesingi* was more or less of the same degree either in adult or in nymph hosts. This is probably due to the inability of *Leidynema appendiculata* juveniles to survive the moulting of the hosts while *Hammerschmidtella diesingi* can successfully do so. The infection by *Thelastoma malaysiense* was found to be high in the adult hosts of both species. This shows that its juveniles are also unable to survive the moulting of the hosts. *Severianoia severiano* was found to occur only in *Neostylopyga rhombifolia* (Stoll) and it only infected the nymphal stage of the host. There was no significant difference in the degree of infection among the adult male and adult female hosts (both species). The infection rate in adults and nymphs differed significantly in *Neostylopyga rhombifolia*.

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