Not for publication

THE INCIDENCE OF NEMATODES IN LARVAE AND ADULTS OF THREE GENERA OF SCARABAEID BEETLES IN SOUTHEASTERN MANITOBA, WITH OBSERVATIONS ON OTHER PARASITES

by
D. M. Rosenberg

FOREST RESEARCH LABORATORY
WINNIPEG, MANITOBA
INTERNAL REPORT MS-10

DEPARTMENT OF FORESTRY
OCTOBER, 1965

INTRODUCTION

"An important question for entomologists is the role and importance of nematodes in regulating natural populations of insects" (Welch 1962). Further, "Nematodes generally play an insignificant role in the natural regulation of insects but with certain groups of insects their regulation is significant". The purpose of this project was to determine the incidence of nematode parasites in populations of May beetles and their larvae, white grubs, in southeast Manitoba. Observations on the incidence of other parasites were made in the course of the search for nematodes. This work was done while the author was employed as a graduate student.

LITERATURE REVIEW

Ritcher (1949) noted that the scarabaeid subfamily Melolonthinae has worldwide distribution and that many of the most injurious members of the family Scarabaeidae belong to this group. Adults of most of the genera in the subfamily feed on the foliage of trees and shrubs. Adult *Phyllophaga* are commonly called May or June beetles (or June "bugs"). These are names loosely applied to related genera as well.

In many parts of the world the larvae of Melolonthinae cause much damage to the roots of grasses, legumes, small fruit plants, shrubs and trees. Of particular economic importance are members of the tribes Melolonthini and Sericini (Ritcher 1958) which include such common genera as *Phyllophaga* and *Serica*, respectively. The larvae of species belonging to the subfamily Melolonthinae are commonly called white grubs.

The morphology, taxonomy and biology of larval Scarabaeoidea were studied by Hayes (1929). Most of the information for the taxonomy of white grubs in this project was obtained from a study of larval Melolonthinae done by Ritcher (1949). The larval morphology and taxonomy of some Indian Scarabaeoidea and West Indian Melolonthinae related to the larvae of this study were described by Gardner (1935) and Böving (1942a) respectively.

Adult taxonomy was treated by Sim (1928), Luginbill and Painter (1953) and Dillon and Dillon (1961). Taxonomic work on both larvae and adults of the genus *Phyllophaga* was done by Böving (1942b). Other papers include a morphological work by Hayes (1922) and revisionary works by Horn (1887) and Glasgow (1916).

Literature on the biology of economically important Scarabaeidae is voluminous, as the Scarabaeidae have worldwide distribution and are pests of economically important crops and of tree plantations. Forbes (1907) discussed life history; food (of the larvae and adults), including common preferences; reproductive habits; relation to weather and seasonal change; hibernation; movements, migrations and dispersals, natural enemies and diseases; and methods of control for the genus Lachnosterna (=Phyllophaga). Biological aspects were also considered by Davis (1913, 1916), Forbes (1916a), Webster (1916), Hayes (1919, 1920, 1925, 1929), Pettit (1930), Chamberlin and Callenbach (1943), Chamberlin, Fluke and Callenbach (1943), Hammond (1948), Shenefelt and Simkover (1951), and Shenefelt (1956a). Nairn and Ives (1965) studied flight patterns of Serica spp. and three species of Phyllophaga in southeastern Manitoba. Much of the literature on the biology of Scarabaeidae was reviewed by Ritcher (1958).

Damage caused by May beetles and white grubs was reported in the north eastern United States, (Davis 1913), in Iowa (Webster 1916; Ball and Walter 1918), in Ontario (Hammond 1936), and in forest tree nurseries in Wisconsin (Shenefelt and Simkover 1950). The white grub problems in plantations in southeastern Manitoba were discussed in more recent papers (Warren 1962; Warren and Hildahl 1963). Criddle (1918), Gibson (1918), Hammond (1936), Shenefelt (1956b), Shenefelt and Simkover (1950, 1951) and Shenefelt et al (1954) discussed physical and chemical control measures. Polivka (1953) reviewed the status of insecticides used to control white grubs in turf. Nairn and Ives (1965) reported preliminary results of using chemical insecticides in machine planting to protect newly planted trees. Works by Hammond (1940, 1946, 1948, 1960) and Chamberlin and Fluke (1947) included control of adults as well as larvae. Chamberlin and Fluke (1947) noted that control of adults was impractical. Shenefelt and Simkover (1951) agreed. Factors such as polyphagous habits, lengths of flight periods and distances of flight make adults harder to control than grubs.

Of special interest to this study is the literature dealing with the natural enemies of May beetles and white grubs, especially nemic and dipterous parasites and mites. Portions of the papers by Forbes (1907), Davis (1913, 1916) and Criddle (1918) were devoted to the natural enemies of white grubs and May beetles. Criddle (1918) gave a brief account of the natural enemies of white grubs in Manitoba. He reported an 8% mortality caused by mermithids over the years of his study. Tachinids and dexiids were the common dipterous parasites; and mites caused many deaths. Van Dine (1912, 1913) presented an account of efforts to introduce beneficial parasites into Puerto Rico. Davis (1919) discussed parasites (insect and nematode), insect predators, mites, and "diseases" of the larvae; insect parasites and spider enemies of the adults and the vertebrate predators of Phyllophaga, and credited nematodes with being effective in their attack on white grubs. He reported a "nematode disease" that struck in the vicinity of Lancaster, Wisconsin, during the later summer and fall of 1915. By early October, 90% mortality in the white grub populations had resulted. Identifications revealed two species to be present: Diplogaster aerivora Cobb and immature Cephalobus (?) sp. Mermithids were found in only four of the several thousand grubs reared. He also reported many tachinid parasites infecting larvae and adults, and mites infesting grubs in the field and in breeding cages. These mites were occasionally found in sufficient numbers to weaken or even kill hosts in the field. Adults were also sometimes infested but these mites were apparently nymphal migratory stages. Hayes (1919) noted that natural enemies of Lachnosterna lanceolata Say were not abundant in Kansas. In a later paper (Hayes 1920), he enumerated the enemies of some Kansas Lachnosterna and reported that mermithids were frequently reared from grubs and that nematodes (probably Diplogaster aerivora) were responsible for mortality of grubs in rearing cages and were abundant in grub collections of L. lanceolata. Dipterous parasites were active enemies of the beetles in his study region and mites were also pests of rearing cages. Parasitism of white grubs by the wasp Tiphia was reported in a later paper (Hayes 1928). Hammond (1940) mentioned dipterous parasites in his study of white grubs in Eastern Canada. Partial control of white grubs by nematodes in Wisconsin was reported by Chamberlin and Fluke (1947). In his review of the biology of Scarabaeidae, Ritcher (1958) stated that pleurostict scarabaeids are attacked by many parasites. The list included nematodes, Diptera and mites. Tachinids are numerous in melolonthine, ruteline and dynastine larvae and are the main parasites of adult scarabaeids. Several species of pyrgotids and a few species of sarcophagids also attack the adult beetles. Welch (1962) noted that "14 families of nematodes are known to be associates of 16 insect "orders" and that "there is a greater frequency of nematode occurrence in those insect orders that are associated with the soil". He indicated which families of nematodes are of most interest to those concerned with insect control.

The following is a review of literature concerned specifically with nematodes as parasites of various scarabaeids. Four new oxyurid parasites of scarabaeid larvae were described by Christie (1931). A new genus (Scarabanema) was erected for the species of nematode which came from the posterior end of the intestine of an unidentified larva which belonged to the Rutelinae or Melolonthinae; two new oxyurids from the larvae of a species of Osmoderma were placed in the genus Thelastoma Leidy and one in the genus Aurorus Leidy. Chamberlin (1944) reported a species of Diplogaster present in the cephalic region of white grubs Phyllophaga spp. Polozhentsev (1952) reported finding mermithids in soil samples that were taken to determine the degree of infestation of scarabaeid beetles. These mermithids were found to play a positive role as parasites of the cockchafers, and were found in sandy soil, sand loam, and the chernozem-like soil of the Buzulukscogo Woods. Basir (1956) listed five thelastomatid nematodes (of five different genera) from various positions in the alimentary tracts of five scarabaeid larvae (three of which were Osmoderma, one Melolontha and one melolonthine or ruteline). Neoaplectana melolonthae was described by Weiser (1958) from grubs of a May beetle from the region of Senica, Slovakia, and later reported (Weiser 1960) in Melolontha melolontha L. larvae taken from the region of the lower Morava River in Czechoslovakia.

Glaser, McCoy and Girth (1940) stated that the infective stages of Neoaplectana glaseri Steiner live in soil and are capable of sustained existence for at least one and a half years in the absence of host insects. Attack is not significant below 18°C. This nematode does not confine its attack to Japanese beetle (Popillia japonica Newm.) alone. The larvae of several species of Phyllophaga, among others, are also infected. It is apparent that N. glaseri is a general parasite of insects and that there is justification for field investigations of its potentialities in the control of a number of insects in addition to the Japanese beetle. The authors suggested that the adult beetles could act as vectors in the dissemination of the parasite. Chamberlin (1944) reported an attempt to infect Phyllophaga spp. with the nematodes N. glaseri and Neoaplectana sp. Neither of these species were recovered but other nematode species were found in the grubs examined. The most abundant of these nematodes seemed to be a species of Diplogaster. Dumbleton (1945) showed that N. glaseri is a potential control agent of the soil-inhabiting pasture pests Odontria and Oxycanus. Weiser (1958) managed to infect Melolontha melolontha and Melolontha hippocastani with Neoaplectana melolonthae and Couturier (1963) discussed the infection of M. melolontha by two mermithid parasites Pseudodermis hagmeiri and Tunicamermis melolonthae. Unlike the thelastomatids, mermithids are usually found in the hemoccele. Experimental contamination was discussed and photographs of nematode-infected M. melolontha were provided. Schmiege (1963) studied a species of Neoaplectana to determine its value as a parasite of forest insect pests. Negative factors in the potential value of this nematode as an insect parasite include moisture requirements, dispersal ability, searching ability and difficulty in handling the nematode. Positive factors include the wide range of parasitization possible, tolerance to broad temperature ranges, ease of propogation and storage, resistance to chemicals, ease of application, and the lack of evidence of host resistance. Therefore, the possibility of using nematodes to control the scarabaeids of this study seems bright. The most recent review of entomophilic nematology is by Welch (1965).

METHODS

Collection of Material

Larvae were collected from eight locations in the southeast part of Manitoba. Grubs from Stead, Milner Ridge and the Pineland Nursery near Hadashville were obtained by digging; those from Badger were collected by following a tree planting machine; those from Whitemouth were collected by following a tractor-drawn plough which was preparing the area for hand planting.

Adults were collected from six areas (Table 1). Those from Vassar and Piney and some from Wampum were light trap collections. Those from the Pineland Nursery and Whitemouth and the remainder from Wampum were hand collections.

Dissections

As many larvae as possible were dissected live. After being collected the living grubs were, where possible, refrigerated in the field until they could be brought to the laboratory. If the grubs could not be dissected immediately, they were keptin a cold room (34°F) in the laboratory. Each grub was placed in a 7 cm. Petri plate filled with distilled water before dissection and identified to genus. Most of those from Milner Ridge were examined carefully for mites; others received cursory examinations. The larvae were examined externally for nematodes and, in some cases, a close examination of the mouth parts was made. Each larva was cut down one side and across the dorsum, anteriorly and posteriorly. The gut was opened and the head capsule broken (Merrill and Ford 1916; Chamberlin 1944). Gross dissections were performed because of the large amount of material and the short time available to examine it. After dissection, preserved grubs were returned to a vial containing the same preservative. Live dissections were left overnight at room temperature so that any nematodes would emerge (Chamberlin 1944). This procedure ensured that large nematodes would be intact and allowed small nematodes which might otherwise be missed to emerge. The following morning microscopic examinations were made on the water samples from specimens randomly chosen and on all specimens that were suspected of harboring nematodes. Larva dissected live were put into 70% ethanol for storage. Nematodes were removed from the dissected larvae and placed in T.A.F.-F.A.A. fixative.[1]

Adults were collected from ultraviolet light traps or by hand. They were stored in refrigerators in the field and then in the cold room (34°F) in the laboratory until they could be dissected. Some of the adults were put in rearing cages with foliage and kept in the Forest Insect Survey rearing room. Adults could not be kept alive too long in the cold room or in the rearing room. As many as possible were dissected live; the remainder, including live beetles which could not be dissected immediately and dead beetles, were pierced and placed in T.A.F.-F.A.A. until they could be dissected. Sex and species determinations were made for all beetles and external examinations for nematodes were made. The elytra and wings were removed and examined for mites; the abdominal tergites were removed and the interior of the abdomen examined. The thorax was split open and, in the live dissections, the head was split open. All dissected beetles were put into T.A.F.-F.A.A. fixative.

Parasite Identifications

Dipterous larvae were separated into different taxa on the basis of spiracular structure. In only one case could a tentative identification be made. Nematodes were identified to family level by Dr. H. E. Welch. Time did not permit further identification, (i.e. genus and species) to be made.

RESULTS AND CONCLUSIONS

The numbers of each genus and species of larval and adult beetles collected in each area are shown in Table 2. One group of adults from Piney

---

[1] 4 parts T.A.F. (7 ml. formalin; 2 ml. triethanolamine; 91 ml. distilled water): 1 part F.A.A. (10 ml. formalin; 10 ml. glacial acetic acid; 80 ml. distilled water). - recommended by Dr. H. E. Welch (personal communication).

(i.e. May 14) has been separated from the other Piney material because they were all Phyllophaga anxia males which represents a biased sample.

Table 3 lists the dipterous and nematode parasites found in the white grubs examined, the genera of the white grubs infected, the specimen numbers of the infected white grubs and the areas from which the infected grubs were collected. Table 4 lists similar information for the adults, except that notes on dipterous eggs found on the beetles are included. Badger had the highest percentage infestation of dipterous larvae in white grubs. The small average number of dipterous larvae occurring per Dichelonyx grub from Whitemouth may be due to the small size of the grub. Dipterous parasites may be exercising some control over the populations of May beetles at Wampum. Frequent multiple infestations by the dipterous larvae may be due to the large size of the host beetle. The dipterous larva recovered from white grub number 358 and labelled Species D is likely Ptilodexia (Dexiidae), and the one from May beetle number 468 and labelled Species G may be a sarcophagid. The dipterous larvae from the remainder of the white grubs and May beetles (i.e. the dipterous larvae labelled Species, A, B, C, E and F) may be tachinids. Seven different taxa of dipterous larvae were recognized. It is peculiar that no Phyllophaga spp. larvae were infested with mermithids. The mature mermithid found on the P. drakii adult had probably emerged before the insect was preserved.

The adult material represented biased collection because: (1) much of it was obtained from light traps which yield a high percentage of males (Nairn and Ives 1965); (2) collections were not made regularly throughout the season; and (3) most of the material was "expendible stock" from other studies. Hand collections give a more equitable sex distribution and are treated separately.

The following table is a breakdown of species, sex, and numbers of adults collected by hand at Wampum and the Pineland Nursery:

Area	Date	P. drakei ♂	P. drakei ♀	P. anxia ♂	P. anxia ♀	Serica spp. ♂	Serica spp. ♀
Wampum	June 2	17	31	-	-	-	-
Pineland	June 3 - 4	3	9	31	20	1	1
Pineland	ca. June 8	12	48	57	16	-	1
Pineland	June 8 - 9	6	8	3	5	-	-
Pineland	ca. June 8*	-	-	50	11	-	-

* Part of a larger group of adults

There were no nematodes in any of these collections and only the beetles collected ca. June 8 were infested with dipterous larvae (4.2% parasitism). The average number of dipterous larvae per infested beetle was 2.9, and only P. anxia were infested. Five infected beetles were from the collection containing only P. anxia but seven came from collections containing P. drakei. In addition, more males than females were infested. Individual light trap collections from the period June 24-July 1 showed a higher amount of parasitism which may indicate that as the season progresses the incidence of parasitism rises.

A summary of the mite infestations on all the adult beetles (collected by light trap and by hand) appears in Table 5. Summaries were compiled for collections exceeding 25 beetles. An average of 65% of adult beetles were found to be infested with mites (includes "light" and "heavy" categories) but 95% of P. drakei from Vassar were infested. It seems likely that a heavy infestation of mites would have some harmful effect on the adult beetle; and if this type of infestation occurred in a sufficient proportion of the population, some control over that population would be exercised. The high percentage of P. drakei infested (80) may indicate some sort of control over that species.

The areas of collection, degree of infestation of the beetles by mites (terms used to describe the infestation - i.e. "none", "light", and "heavy" denote the same as before), sex of the beetles, and numbers within each degree of infestation for adult P. drakei and P. anxia collected by hand is shown in Table 6.

Table 7 is a summary of this data grouped by sex and species. P. drakei had a higher average percentage infestation by mites than P. anxia and male and female beetles had an approximately equal infestation percentage.

No nematodes were found in the microscopic examinations of the live dissections. However, what can be described as "strings of beads" and "short rods" were observed from some of the specimens. In an attempt to identify these, Steinhaus (1963) and Welch (personal communication) were consulted. Positive identifications were not made and the identifications ventured are speculative. The "strings of beads" may have been gregarines (Protozoa). Weiser (in Steinhaus 1963) recorded gregarines in Melolontha melolontha from France. The rods may have been bacteria, possibly of the genus Clostridium.

Miscellaneous observations

1. Some of the grubs from Stead were found to be diseased. The fat in these resembled descriptions given by Surany (1960) for certain diseased Oryctes.

2. Larva #408 contained a dipterous larva that was parasitized by what appeared to be an hymenopteran. This was the only case of hyperparasitism found.

3. It was interesting to find corixids under the elytra of three adults from two different locations (Vassar and Wampum). They were identified[1] as Callicorixa audeni Hung. It is possible that they were attracted to the light traps and flew onto the May beetles inside the trap. Or, this may be a commensal relationship: the corixids using the May beetles for transportation.

4. Some of the dipterous larvae found in the white grub larvae and some found in the adult beetles had a peculiar brown cuticular cone on the posterior end of the body, covering the spiracular end of the larva. This "covering" extended around the body and became lighter in color anteriorly. This could have been the start of pupation or it could represent some sort of host reaction . . . encapsulation of the parasite. Couturier (1963) recorded an encapsulation reaction to the nematode Tunicamermis by Melolontha melolontha.

5. The abdominal contents (except for genitalia) and thoracic musculature of some adult beetles parasitized by certain dipterous larvae were completely eaten out. These undoubtedly were advanced cases of parasitism. If the damage was done in the male following mating and in the female following egg laying then these Diptera would be of little use in the biological control of the May beetles. However, if this damage in the male and female prevented mating and egg laying respectively (either through destruction of the organs of reproduction or the flight muscles) then these Diptera would be of decided value to the biological control of May beetles.

DISCUSSION

Factors Influencing the Incidence of Nematodes

The low incidence of nematodes infecting the adult and larval beetles is difficult to explain. Welch (1962) listed three important factors for the function of neoaplectanid nematodes as biological control agents: moisture, moderate temperature, and host density.

Moisture requirements: - Nematodes are entirely dependent on water for their activities; some species can survive dry conditions but they cannot move, feed, lay eggs, etc., until moisture is restored to the soil and they become active again (Wallace 1963). Winslow (in Sasser & Jenkins 1960) also noted the importance of water to nematode survival. "[Nematodes] cannot lead an active existence in the absence of free water in the soil". Schmiege (1963) in his study of the feasibility of using a neoaplectanid nematode to control some forest insect pests noted that moisture requirements of the nematode are a negative factor in the nematode's potential

---

1 by J. C. E. Melvin. For. Ins. Survey. Can. Dept. For.

value as an insect parasite. Dry conditions may depress nematode activity but extreme moisture conditions in a soil also do not favor nematodes (Wallace 1963). The 1965 summer season in southeastern Manitoba was a particularly rainy one. Perhaps this was a factor in reducing numbers of nematodes. However, nematode activity is not influenced by soil moisture alone and many other factors vary simultaneously with moisture and this is when the problem becomes difficult (Wallace 1963).

Temperature: - When considering whether or not the season's temperatures adversely affected the numbers of nematodes available for infecting white grubs and May beetles the following factors would have to be considered (Wallace 1963):

1. Different nematode activities (hatching, reproduction, movement, etc.) may have different temperature requirements.
2. Age and nutritional state may affect temperature responses.
3. Different populations of the same species may have different temperature characteristics. Attempts to explain a low occurrence of nematodes on the basis of temperature would, therefore, be difficult if not impossible.

Host density: - Only the Milner Ridge area was considered to be heavily populated by white grubs: the Whitemouth field was second. It is interesting to note that all the white grubs infected by mermithids came from the Whitemouth location and the cadaver containing the neoaplectanids came from Milner Ridge. Welch (1962) told of discontinuous parasite distribution within and between populations of the same host. It is possible that localized concentrations of nematodes were missed.

Possibilities of Establishing a Program of Biological Control Using Nematodes in Southeastern Manitoba

A strong case can be presented against the immediate establishment of such a program.

1. There does not seem to be sufficient white grub and May beetle damage in this area to warrant its institution.
2. The low incidence of nematodes during the 1965 season is not encouraging.
3. The lack of extensive field trials of entomophilic nematodes is a factor to be considered.

Whether or not sufficient economic damage is caused by white grubs and May beetles to warrant initiation of a possibly very expensive biological control program is a key issue. If extensive surveys carried out over a period of years showed that white grubs and May beetles were causing marked damage then it might be an idea to compare the costs of chemical control over the next several years with the cost of initiating a biological control program. However, biological control is less controversial, less dangerous to man and is sometimes longer lasting than control achieved by chemicals.

Because nematodes did not seem to have a significant role in the regulation of white grubs and May beetles in the 1965 season does not mean that they cannot be. Perhaps in other years, when conditions are more favorable, nematodes will be important in control.

The lack of intensive field trials of entomolophilic nematodes may be the greatest drawback to initiation of a program of biological control. It is known that "the Neoaplectanidae, Tylenchoidea (particularly the Allantonematidae), Aphelenchoidea and Mermithidae ... are of most potential to economic entomology" (Welch 1962). Neoaplectanidae have particular value because they can be manipulated successfully as biological control agents against soil insects. Neoaplectana glaseri has survived for long periods at low host densities which may be of importance in this area. Work has been done on the Steinernematid DD-136. There have been no field trials of other entomophilic nematodes but mermithids offer the greatest promise of these (Welch 1965). Therefore, it would seem necessary for some basic work to be done before a program of biological control could be initiated against white grubs and May beetles.

BIBLIOGRAPHY

Ball, E. D. and E. V. Walter. 1918? Injury from white grubs in Iowa. Iowa Agr. Expt. Stn. Circular No. 60.

Basir, M. A. 1956. Oxyuroid Parasites of Arthropoda. Zoologica, 38. 79 pp.

Böving, A. G. 1942a. Descriptions of the larvae of some West Indian Melolonthine beetles and a key to the known larvae of the tribe. Proc. U. S. N. M. 92 (3146): 167-176.

Böving, A. G. 1942b. A classification of larvae and adults of the Genus Phyllophaga (Coleoptera: Scarabaeidae). Memoirs ent. Soc. Wash. No. 2. pp. 1-96.

Chamberlin, T. R. 1944. Observations on nematodes associated with white grubs. J. Econ. Ent. 37 (2): 313-314.

Chamberlin, T. R. and J. A. Callenbach. 1943. Oviposition of June beetles and the survival of their offspring in grasses and legumes. J. Econ. Ent. 36 (5): 681-688.

Chamberlin, T. R. and C. L. Fluke. 1947. White grubs in cereal and forage crops and their control. Univ. of Wisc. Res. Bull. 159. pp. 1-15.

Chamberlin, T. R., C. L. Fluke and J. A. Callenbach. 1943. Species, distribution, flight, and host preferences of June beetles in Wisconsin. J. Econ. Ent. 36 (5): 674-680.

Christie, J. R. 1931. Some nemic parasites (Oxyuridae) of Coleopterous larvae. J. Agr. Res. 42 (8): 463-482.

Couturier, A. 1963. Recherches sur des Mermithidae, nématodes parasites du hanneton commun (Melolontha melolontha L. Coleopt. Scarab.) Ann. Épiphyties, 14 (3): 203-267. (In French; English summary.)

Criddle, N. 1918. The habits and control of white grubs in Manitoba. Agr. Gazette of Can. 5 (5): 449-454.

Davis, J. J. 1913. Common white grubs. U. S. D. A. Farmer's Bull. 543. pp. 1-20. (Revised, 1918; Farmer's Bull. 940, pp. 1-28).

Davis, J. J. 1916. A progress report on white grub investigations. J. Econ. Ent. 9 (2): 261-281.

Davis, J. J. 1919. Contributions to a knowledge of the natural enemies of Phyllophaga. State of Ill. Nat. Hist. Survey Bull. Vol. XIII, 1-138.

Dillon, E. S. and L. S. Dillon. 1961. A Manual of Common Beetles of Eastern North America. Row, Peterson and Company; Evanston, Ill.

Dumbleton, L. J. 1945. Bacterial and nematode parasites of soil insects. N. Z. J. Sci. & Technol. 27: 76-80.

Forbes, S. A. 1907. On the life history, habits and economic relations of the white-grubs and May-beetles. Univ. of Ill. Agr. Expt. Stn. Bull. No. 116: 447-480.

Forbes, S. A. 1916a. A general survey of the May beetles (Phyllophaga) of Illinois. Univ. of Ill. Agr. Expt. Stn. Bull. No. 186. pp 215-257.

Forbes, S. A. 1916b. The influence of trees and crops on injury by white grubs. Univ. of Ill. Agr. Expt. Stn. Bull. No. 187. pp 261-265.

Gardner, J. C. M. 1935. Immature stages of Indian Coleoptera (Scarabaeoidea). Indian Forest Records (new series), 1 (1): 1-176.

Gibson, A. G. 1918. Prevent white grub injury. Can. Dept. Agr. Crop Protection. Leaflet No. 5.

Glaser, R. W., E. W. McCoy and H. B. Girth. 1940. The biology and economic importance of a nematode parasitic in insects. J. Parasitol. 26: 479-495.

Glasgow, R. D. 1916. Phyllophaga Harris (Lachnosterna Hope): A revision of the synonymy and one new name. Bull. Ill. State Lab. of Nat. Hist. Vol. XI. 365-379.

Hammond, G. H. 1936. White grub control in Ontario. Can. Dept. Agr. D.F.C.G.I. No. 148.

Hammond, G. H. 1940. White grubs and their control in Eastern Canada. Can. Dept. Agr. Publication 668; Farmer's Bull. 86. pp.1-18.

Hammond, G. H. 1946. How to foretell and control outbreaks of white grubs. Can. Dept. Agr., Div. of Entomology: Processed Publication No. 42. pp. 1-5.

Hammond, G. H. 1948. White grubs. Can. Dept. Agr. Processed Publication No. 88. pp. 1-4 (Revised, 1953; pp. 1-6).

Hammond, G. H. 1960. Control of white grubs in Eastern Canada. Can. Dept. Agr. Publication No. 1069. pp. 1-8.

Hayes, W. P. 1919. The life-cycle of Lachnosterna lanceolata Say. J. Econ. Ent. 12 (1): 109-117.

Hayes, W. P. 1920. The life histories of some Kansas Lachnosterna. J. Econ. Ent. 13 (3): 303-318.

Hayes, W. P. 1922. The external morphology of Lachnosterna crassissima Blanch. (Scarabaeidae: Coleop.). Trans. Amer. Micros. Soc. 41 (1): 1-28.

Hayes, W. P. 1925. A comparative study of the history of certain phytophagous scarabaeid beetles. Ag. Expt. Stn. Kansas State Agr. Coll. Kansas Tech. Bull. 16. pp. 1-146.

Hayes, W. P. 1929. Morphology, taxonomy and biology of larval Scarabaeoidea. Ill. Biol. Monog. 12 (2): 1-119.

Horn, G. H. 1887. Revision of the species of Lachnosterna of America north of Mexico. Trans. Amer. ent. Soc. 14 (27): 209-296.

Luginbill, P. Sr. and H. R. Painter. 1953. May beetles of the United States and Canada. U. S. D. A. Tech. Bull. No. 1060. pp. 1-102.

Merrill, J. H. and A. L. Ford. 1916. Life history and habits of two new nematodes parasitic in insects. J. Agr. Res. 6 (3): 115-127.

Nairn, L. D. and W. G. H. Ives. 1965. White grub studies - 1964. Can. Dept. For. Interim Res. Rept., For. Ent. Lab., Winnipeg, Manitoba.

Pettit, R. H. 1930. June beetles or white grubs in Michigan. Agr. Expt. Stn., Mich. State College. Circular Bull. No. 132. pp. 1-10.

Polivka, J. B. 1953. Present status of insecticides for control of grubs in turf. Ohio Farm and Home Res. 42 (306).

Polozhentsev, P. A. 1952. (New Mermithidae from the sandy soil of pine woods.) (In Russian.) Trudi Gelmintologicheskoi Laboratorii Akademii Nauk. S. S. S. R. 6: 376-382.

Ritcher, P. O. 1949. Larvae of Melolonthinae. Ky. Agr. Expt. Stn., Univ. of Ky. Bull. 537. pp. 1-36.

Ritcher, P. O. 1958. Biology of Scarabaeidae. Ann. Rev. Entomol. 3: 311-334.

Schmiege, D. C. 1963. The feasibility of using a neoaplectanid nematode for control of some forest insect pests. J. Econ. Ent. 56 (4): 427-431.

Shenefelt, R. D. 1956a. Seasonal abundance of white grubs. Univ. Wisc. Forest Res. Notes No. 25.

Shenefelt, R. D. 1956b. A further note on protecting machine transplanted trees from white grubs. Univ. Wisc. Forestry Res. Notes No. 26.

Shenefelt, R. D. and H. G. Simkover. 1950. White grubs in Wisconsin forest tree nurseries. J. Forestry 48 (9): 429-434.

Shenefelt, R. D. and H. G. Simkover. 1951. Insecticides for control of white grubs. J. Econ. Ent. 44 (3): 359-362.

Shenefelt, R. D., H. R. Liebig and R. C. Dosen. 1954. Protecting machine transplanted trees from white grubs. Univ. Wisc. Forestry Res. Notes. No. 17.

Sim, R. J. 1928. Phyllophaga (Scarabaeidae) of the United States and Canada. State of N. J. Dept. of Agr., Bureau of Statistics and Inspection. Circular No. 145, pp. 1-60.

Steinhaus, E. A. 1963. Insect Pathology. An advanced treatise. Vol. 2. Academic Press, N. Y.

Surany, P. 1960. Diseases and biological control in rhinoceros beetles Oryctes spp. (Scarabaeidae: Coleoptera) South Pacific Commission Technical Paper No. 128.

Van Dine, D. L. 1912. Progress report on introductions of beneficial parasites into Porto Rico. First Report of the Board of Commissioners of Agriculture of Porto Rico. (for the period July 1, 1911 to Jan. 1, 1912). pp. 31-47.

Van Dine, D. L. 1913. The introduction of parasites of May beetles into Porto Rico. Second Report of the Board of Commissioners of Porto Rico. (for the period Jan. 1, 1912 to Jan. 1, 1913). pp. 36-48.

Wallace, H. R. 1963. The Biology of Plant Parasitic Nematodes. Edward Arnold (Publishers) Ltd., London. 280 pp.

Warren, G. L. 1962. White grub studies. Can. Dept. For. Interim Research Report; Forest Entomology Laboratory, Winnipeg, Manitoba

Warren, G. L. and V. Hildahl. 1963. Report of white grub damage in a red pine population in southeastern Manitoba. Can. Dept. For. Information Report; Forest Entomology Laboratory, Winnipeg, Man.

Webster, R. L. 1916. White grubs in Iowa. Iowa Agr. Expt. Stn. Circular No. 29, pp. 1-4.

Weiser, J. 1958. Ein neuer Nematode als Parasit der Engerlinge des Maikäfers, Melolontha melolontha in der Tschechoslowakei. (A new nematode as a parasite of the grubs of Melolontha melolontha in Czechoslovakia). Internat'l Conference of Ins. Pathol. and Biol. Contr. (1st), Prague, Transactions, pp. 331-336. (In German).

Weiser, J. 1960. Nemoci ponrav chroustů ČSR. (Infections of Melolontha larvae in Czechoslovakia). Vestn. Česk. Spolecnosti Zool., 24: 71-74. (English summary).

Welch, H. E. 1962. Nematodes as agents for biological control. Proc. Entomol. Soc. Ont. 92: 11-19.

Welch, H. E. 1965. Entomophilic nematodes. Ann. Rev. Entomol. 10: 275-302.

Winslow, R. D. 1960. Nematode ecology. In: 'Nematology Fundamentals and Recent Advances with Emphasis on Plant Parasitic and Soil Forms'. Edited by J. N. Sasser and W. R. Jenkins. Chapter 36. Chapel Hill: Univ. N. Carolina Press.

Table 1

Description of the area from which white grubs and May beetles were collected

Area	Insect stage collected	Map location	Brief description
Agassiz	larvae	18-13-10 E. P. M.	Red pine plantation planted 1963
Badger	larvae	LS 10 & 15-29-3-12 E. P. M.	Old burn; fairly heavy sod cover
Milner Ridge	larvae	36-13-9 E. P. M.	40 foot wide strip beneath a power line; clumps of shrubs; thin sod layer
Pineland Nursery	larvae adults	36-7-11 E. P. M.	Larvae: most came from a plantation of red pine and jack pine cultivated and seeded in the fall of 1964; Adults: 2 light traps located in open pasture
Piney	adults	LS 10 & 15-4-2-11 E. P. M.	Abandoned, open pasture
Stead	larvae	LS 13-15-17-8 E. P. M.	1961 burn; low shrubs; light sod cover
Vassar	adults	LS 1-19-2-13 E. P. M.	Pasture - still under grazing, last cultivation: 1963
Wampum	larvae adults	Larvae - LS 1 & 2-31-1-13 E. P. M.	Larvae- open pasture; medium sod cover; sand clay loam
		Adults - LS 15-8-1-13 E. P. M. LS 2-17-1-13 E. P. M.	Adults- old burn; regeneration sparse; thin sod layer (Nairn and Ives, 1965)
Whitemouth	larvae adults	14-7-11 E. P. M.	Area previously cleared of timber; fairly heavy sod cover
Whiteshell	larvae adults	8-12-15 E. P. M.	Cultivated land

Table 2

The areas from which larval and adult beetles were collected and the numbers of each genus and species collected at a particular area

Area where collected	Larvae: Phyllophaga spp.	Larvae: Dichelonyx spp.	Larvae: Serica spp.	Adults: P. drakei	Adults: P. anxia	Adults: P. nitida	Adults: Serica sp.	Adults: Diplotaxis sp.
Agassiz	1	-	-					
Badger	68	-	7					
Milner Ridge	73	-	13					
Pineland Nursery	8	-	-	86	194	-	3	-
Piney				16	11	6	-	1
Piney (May 14)				-	99	-	-	-
Stead	2	-	39					
Vassar				19	7	1	-	-
Wampum	1	-	-	94	3	1	-	-
Whitemouth	194	122	32	3	1	2	2	-
Whiteshell	8	-	-	-	2	-	-	-
Totals	355	122	91	218	317	10	5	1
Percentage males	-	-	-	53	82	50	40	0

Table 3

Dipterous and nematode parasites found in the white grub larvae examined

Location	Specimen No.	Genus	Parasite
Bader	8	Phyllophaga	2 Diptera larvae - Species A
	24	"	1 Diptera larva - "
	36	"	2 Diptera larvae - "
	47	"	1 Diptera larva - "
	51	"	2 Diptera larvae - "
	60	"	1 Diptera larva - "
	73	"	3 Diptera larvae - "
Whitemouth	90	Dichelonyx	1 Diptera larva - Species B
	151	"	1 nematode - 1 immature mermithid
	154	Serica	1 nematode - 1 immature mermithid
	155	Dichelonyx	1 Diptera larva - Species B
	163	"	1 nematode - 1 immature mermithid
	171	"	1 Diptera larva - Species C
	175	"	1 nematode - 1 immature mermithid
	195	"	1 nematode - 1 immature mermithid
	196	"	1 Diptera larva - Species C
	337	"	1 nematode - 1 immature mermithid
	358	"	1 Diptera larva - Species D
Milner Ridge	480	Phyllophaga	Juvenile nematodes (Neoaplectanidae ?)
Stead			4 - free-living nematodes --- 3 prepared for examination --- 2 mature and 1 immature mermithids --- 1 not prepared - most likely mature; a mermithid

Table 4

Dipterous and nematode parasite found in May beetles examined; notes on dipterous eggs found on the beetles

Location	Specimen No.	Species and sex	Parasites, etc.
Pineland Nursery	91	*P. anxia* ♀	9 Diptera larvae - Species E
	136	*P. drakei* ♀	4 Diptera eggs
	164	*P. anxia* ♂	1 Diptera egg
	207	"	2 Diptera larvae - Species E
	214	"	3 Diptera larvae - Species E
	226	"	2 Diptera larvae - Species E
	233	"	2 Diptera larvae - Species E
	263	"	3 Diptera larvae - Species E
	268	"	2 Diptera larvae - Species E
	497	"	4 Diptera larvae - Species E
	498	"	2 Diptera larvae - Species E and 4 Diptera eggs
	507	"	? Diptera eggs
	512	"	3 Diptera larvae - Species E and 2 Diptera eggs
	515	"	2 Diptera larvae - Species E and 3 Diptera eggs
	528	"	1 Diptera larva - Species E
Wampum	373	*P. drakei* ♂	2 Diptera larvae - Species E
	376	"	3 Diptera larvae - Species E
	377	"	2 Diptera larvae - Species E
	380	"	4 Diptera larvae - Species E
	382	"	1 Diptera larva - Species E
	383	"	2 Diptera larvae - Species E
	384	"	5 Diptera larvae - Species E
	388	"	1 Diptera larva - Species E
	389	"	3 Diptera larvae - Species E
	441	"	3 Diptera larvae - Species E
	442	"	3 Diptera larvae - Species E and 5 Diptera eggs
	443	"	1 Diptera larva - Species E
	445	*P. drakei* ♀	3 Diptera larvae - Species E
	447	*P. drakei* ♂	3 Diptera larvae - Species E and 5 Diptera eggs
	448	*P. anxia* ♂	1 Diptera larva - Species E
	449	*P. drakei* ♂	1 Diptera larva - Species E
	451	"	4 Diptera larvae - Species E
	452	"	1 Diptera larva - Species E
	453	"	3 Diptera larvae - Species E
	454	"	2 Diptera larvae - Species E
	457	"	2 Diptera eggs
	458	"	5 Diptera larvae - Species E
	461	"	5 Diptera larvae - Species E and 8 Diptera eggs

Table 4 (Concl.)

Location	Specimen No.	Species and sex	Parasites, etc.
Wampum (cont'd)	463	P. drakei ♂	2 Diptera larvae - Species E
	464	"	1 Diptera larva - Species E
	465	"	2 Diptera larvae - Species E
	466	"	1 Diptera larva - Species E
	467	"	2 Diptera larvae - Species E
	468	"	3 Diptera larvae - Species E
			1 Diptera larva - Species G
	470	"	2 Diptera larvae - Species E
	471	"	3 Diptera larvae - Species E
	472	"	4 Diptera larvae - Species E
	473	"	2 Diptera larvae - Species E
Vassar	401	"	6 Diptera eggs
	404	"	1 Diptera larva - Species F
	408	"	1 nematode - mature ♂ mermithid
			1 Diptera larva - Species E (?)
Piney	417	P. anxia ♂	2 Diptera larvae - Species E
	418	"	1 Diptera egg
	422	"	2 Diptera larvae - Species F
	424	P. drakei ♂	3 Diptera eggs
	437	"	3 Diptera larvae - Species E
	440	P. anxia ♂	4 Diptera larvae - Species E

Table 5

A summary of mite infestations on the adult P. drakei and P. anxia collected by light trap and by hand

Area of Collection	P. drakei No. adults collected	P. drakei Percentage infested Light	Heavy	Total	P. anxia No. adults collected	P. anxia Percentage infested Light	Heavy	Total	Total No. adults collected	Total Percentage infested* Light	Heavy	Total
Pineland Nursery	86	72	3	76	194	53	1	53	283	58	1	59
Vassar	19	68	26	95	7	42	28	71	27	59	26	89
Piney	16	75	6	81	11	45	0	46	34	50	3	68
Piney (May 14)	-	-	-	-	99	58	2	60	99	58	2	60
Wampum	94	70	11	81	-	-	-	-	98	69	10	81
Total	215	71	9	80	311	59	2	55	541	60	4	65

* Infestations were rated as light if approximately 1-50 mites were present and as heavy if the number of mites exceeded 50.

Table 6

Numbers of hand-collected adult P. drakei and P. anxia infested with mites in different areas (infestation ratings as before)

Collection	None		Light		Heavy	
	♂	♀	♂	♀	♂	♀
			P. drakei			
Wampum	5	7	12	20	0	4
Pineland (3-4)	2	4	1	4	0	1
Pineland (ca. 8)	2	9	9	39	1	0
Pineland (8-9)	2	2	4	5	0	1
			P. anxia			
Pineland (3-4)	17	12	14	8	0	0
Pineland (ca. 8)	27	6	29	10	1	0
Pineland (8-9)	2	2	1	3	0	0
Pineland (ca. 8)*	17	7	33	4	0	0

* Part of a larger collection.

Table 7

Percentages of mite infestations on hand-collected P. drakei and P. anxia adults grouped by species and sex for each area (number of beetles in parentheses)

	Grouped by species disregarding sex		Grouped by sex disregarding species		
	P. drakei	P. anxia	Males	Females	Total
Wampum	75 (48)	-	71 (17)	77 (31)	75 (48)
Pineland (3-4)	50 (12)	43 (51)	44 (34)	45 (29)	44 (63)
Pineland (ca. 8)	80 (60)	55 (73)	58 (69)	61 (64)	67 (133)
Pineland P. anxia	-	61 (61)	66 (50)	36 (11)	61 (61)
Pineland (8-9)	71 (14)	50 (8)	56 (9)	69 (13)	64 (22)
Total	75 (134)	53 (193)	59 (179)	60 (148)	62 (327)