Feeding Activity of Carabid Beetles and Spiders on Gypsy Moth Larvae (Lepidoptera: Lymantriidae) at High-density Prey Populations[1, 2]

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ABSTRACT Pitfall traps and tree bands were used to collect arthropods at two sites in southeastern New Hampshire during the 1982-83 outbreak of gypsy moth. Pitfall traps caught more individuals (74% of total) than tree bands. Carabid beetles (59%) and spiders (22%) were the dominant groups. Twenty-two genera and 48 species of carabid beetles and 31 genera and 43 species of spiders were collected. Spiders had more species and a larger proportion (38%) collected under tree bands than did carabid beetles (12%).

Guts of all individuals were tested for presence of gypsy moth proteins using ELISA. Fourteen genera and 26 species of carabid beetles tested positive with the highest percent (50%) recorded for *Calosoma frigidum* Kirby. Twenty-eight genera and 31 species of spiders tested positive with the highest percent (57%) recorded for *Haplodrassus bicornis* (Emerton). Positive test results were generally higher in tree band collections for species in either group. Positive tests may have been overestimated in the carabid beetles through carrion feeding, and in the spiders because of the extended period required to digest meals.

KEY WORDS Carabidae, carabid beetles, spiders, gypsy moth, ELISA, biological control, serology, *Lymantria dispar.*

The gypsy moth, *Lymantria dispar* (L.), (Lepidoptera: Lymantriidae) has become a well-established, serious defoliator of forests in northeastern United States and much effort has been expended on its biology and control (Doane and McManus 1981). Because the gypsy moth is an exotic insect in North America, part of this effort has centered on classical biological control with the importation of many parasitic Hymenoptera and Diptera (Reardon 1981). Few arthropod predators have been introduced; the most notable is the caterpillar hunter *Calosoma sycophanta* L. This insect is currently being studied by Weseloh in Connecticut (Weseloh 1985a, 1985b, 1988). Native insect and spider predators have received little attention as evidenced by the brief list presented by Smith and Lautenschlager (1978, 1981).

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Many of the native arthropod predators that might feed on gypsy moth are nocturnal feeders and remain hidden and inactive during daylight making direct observations very difficult. The feeding mechanisms involved are varied and direct gut content analyses by classical methods are not always applicable. Even within the carabid beetles food may contain undigested fragments of prey organisms or be completely extraintestinal (Thiele 1977, Evans and Forsyth 1985). Removing predators from their habitat as in laboratory feeding studies or introducing prey labelled with radioactive materials may have a disruptive effect on natural predator-prey interactions. The least disruptive technique has been serology because it can analyze activity that has occurred hours or sometimes days before while the prey and predator remain undisturbed in their natural habitat.

Serology has limitations in application; it cannot answer such questions as the number of prey eaten, when the prey was eaten or if the prey was alive or dead when eaten (active predation versus carrion feeding). For practical reasons most serological predator-prey studies have involved single prey species (i.e. Douglas-fir tussock moth: Fichter and Stephen 1981) with polyphagous predators (i.e. carabid beetles: Hance and Reiner 1987, Hagley and Allen 1988, or spiders: McIver 1981).

Serology has not been used in previous studies of arthropod gypsy moth, predator-prey relationships. The purpose of this study was to enumerate which carabid beetle and spider species were present and active on the lower trunks and ground surface when late instar gypsy moths were present and to determine which of these species were more likely to feed on gypsy moth larvae using the enzyme-linked immunosorbent assay (ELISA).

Materials and Methods

Field Studies. Collections of potential predators of gypsy moth larvae were made at two forested sites in southeastern New Hampshire, one a dry ridge with shallow soils and exposed granite bedrock and the other with clay soils and bordering on a swamp. Red oak (*Quercus rubra* L.) was the dominant tree species on both sites with lesser amounts of white birch (*Betula papyrifica* Marshall), red maple (*Acer rubrum* L.), American Beech (*Fagus grandifolia* Ehrhart), white pine (*Pinus strobus* L.) and white ash (*Fraxinus americana* L.). Gypsy moths were very abundant and caused nearly complete defoliation during the two years of this study. A nucleopolyhedral virus drastically reduced caterpiller survival in the second year.

Tree bands and pitfall traps were the two methods chosen for collecting arthropods. They are appropriate for organisms found on tree trunks and the ground surface but not for foliage inhabitants. Thus, this study concentrated on predators of late larval instars which descend to protected areas on tree trunks and in the ground litter during daytime for resting and ascend at night to feed on foliage. Each pitfall unit was composed of two 12.06 cm diameter plastic cups separated by a 91.44 cm long plastic barrier (Reeves 1980). Ten pitfall units were used at the dry ridge site in 1982 while five were used at each site in 1983. Traps were spaced at least 25 meters apart. Organisms collected from the pitfall traps each day were combined into a single sample from each site. Two trees, preferably oak, were banded near each pitfall trap location. Bands were made of a dark green canvas material wrapped around trees and cut into 15 - 20 cm wide flaps and

collections made with a modified insect net (Dunn and Reeves 1980). All organisms collected each day from tree bands were combined into a single sample from each site.

Collections began when third-instar larvae were first observed on the lower trunks of trees, and continued through pupation. All collections were made as soon after sunrise as possible and frozen (-20°C) to prevent degradation of the stomach contents. Pitfall and tree band samples were taken seven days per week in 1982 (27 May - 30 July) but only four days per week in 1983 (7 June - 30 July). In 1983 organisms in the pitfall traps and tree bands were removed and released on Monday to clear the traps of weekend accumulation and samples were taken on the four days following.

All adult carabid beetles (Carabidae) and spiders (Aranea) were identified to species. Other arthropods were identified to class, order or family, depending on the difficulty and time required in identification.

Laboratory Studies. The anti-gypsy moth antiserum was produced in rabbits against starved, macerated third instar larvae, from which the cuticle had been removed, by ImmunoSystems Inc., Biddeford, ME. These larvae were obtained from the Gypsy Moth Methods Development Laboratory at Otis Air Force Base, MA. The antibody, at an initial concentration of 13.2 mg antibody/ml glycerol, was stored at -20°C. The anti-gypsy moth-horseradish peroxidase conjugate (Ab-HRPO) was also produced by ImmunoSystems Inc. using a modification of the methods described by Wilson and Nakane (1978).

A double antibody sandwich ELISA was used as described by Voller *et al.* (1979). Briefly, the anti-gypsy moth antiserum was diluted to 1 μg/ml in PBS/T buffer (0.1 M sodium phosphate buffer, pH 7.4, containing 0.15 M NaCl and 0.5 g/l Tween 20) and 0.2 ml added to each well of a 96-well Immulon-2 microtiter plate (Dynatech Laboratories, Alexandria, VA). The plate was incubated overnight at room temperature and washed three times with PBS/T. The samples to be tested were prepared as described below, 0.2 ml added to each well and incubated for 2 h at room temperature. After washing with PBS/T, 0.2 ml of the Ab-HRPO diluted to 1 μg/ml in PBS/T was added to the wells and incubated for 1 h at room temperature. Finally, the plate was washed and 0.2 ml of OPD substrate solution (0.1 M sodium phosphate, adjusted to pH 5 with 0.1 M critic acid, containing 0.4 g/l o-phenylenediamine and 0.01% H_2O_2) was added to each well. The results were read visually and compared to positive and negative controls. Positive controls were dilutions of macerated gypsy moth larvae (from 1:200 to 1:516,000) in PBS/T; negative controls contained either no sample or an unrelated caterpillar species. Positive reactions were an orange color whereas negative samples were clear or a faint yellow color.

Tests for cross-reactions were done with all lepidopterous larvae other than gypsy moth found at the collection sites. These were frozen upon collection and prepared for testing by removing the cuticle and any hairs and macerating the remaining tissue in PBS/T. These were tested by ELISA as previously described.

Feeding tests were carried out with *Calosoma frigidum* Kirby, *Pterostichus mutus* Say, *P. pensylvanicus* LeConte and *Lycosa* spp. to determine how long gypsy moth proteins could be detected in the gut. Field collected adult beetles and spiders were starved for two days, placed in individual containers with a gypsy moth larva and observed until feeding was complete (usually about 30 minutes from first attack for beetles). Test individuals were kept at approximately 25°C

(room temperature) throughout the starvation and post feeding period. These were frozen at intervals up to 24 hours and tested as in field-collected organisms.

Analyses. The gypsy moth larval stages present as shown in Figures 1 and 2 were determined from specimens collected under tree bands. Instars were identified by measuring the length of the epistomal suture which is used as an indication of the entire head capsule width (Sorge 1979). The dates when each life stage was present and the approximate time span when each stage was predominant were combined for both sites and years and these latter time periods were used for determining feeding percentages on each stage in Figures 1 and 2. A Chi-square test for fitness was done for all genera of carabid beetles and spiders that had at least 20 individuals with a positivie reaction and for the five most abundant species of carabids, comparing percents positive between pitfall traps and tree bands.

Results

A positive reaction with the anti-gypsy moth antiserum was obtained in the control wells indicating that the ELISA could be used to determine presence of gypsy moth or gypsy moth-like antigens. A range in color intensity from dark orange at high antigen concentrations to light yellow at the most dilute antigen concentrations showed that a quantification of the amount of antigen present was possible, although not done in this study.

Of the Lepidoptera tested for cross-reactions, only the larvae of one genus of underwing moth (Lepidoptera: Noctuidae: *Cotacala*) produced a positive reaction. Other larvae tested included *Malacosoma americana* (F.), *Agrotis* sp., two unidentified species of Noctuidae and one each in Geometridae and Arctiidae. Because only two *Cotacala* larvae were collected under the tree bands during the 1982-83 seasons, the possibility that a positive test resulting from feeding on one of these larvae seems negligible.

In the laboratory feeding tests the three carabid beetle species readily fed on late instar gypsy moth larvae while none of the *Lycosa* spp. fed. The analyses of the beetles showed that gypsy moth protein could be detected in the guts of ground beetles for up to 24 hours while starved individuals all produced negative results. Forest carabid beetles are primarily nocturnal hunters (Thiele 1977) and, because nighttime field temperatures were nearly always at or below laboratory room temperature, the chance of an individual carabid completely digesting a gypsy moth meal and testing negative within a 24 hour period was considered remote. It was thus appropriate for field collections of carabid beetles to be made at 24 hour intervals and as soon after full daylight as possible. Digestion rates for other organisms tested were not determined. Spiders are known to have an extended digestive period (McIver 1981) and percentages of positive test results for these organisms on a 24 hour collection interval or undoubtedly inflated.

Five classes of arthropods were collected and tested (Table 1) with predatory habits most likely to be expected in the Insecta, Chilopoda and Arachnida. The positive test results found in the Crustacea and Diplopoda were considered the result of carrion feeding and their importance may only be in the further dissemination of the nucleopolyhedrosis virus disease organisms.

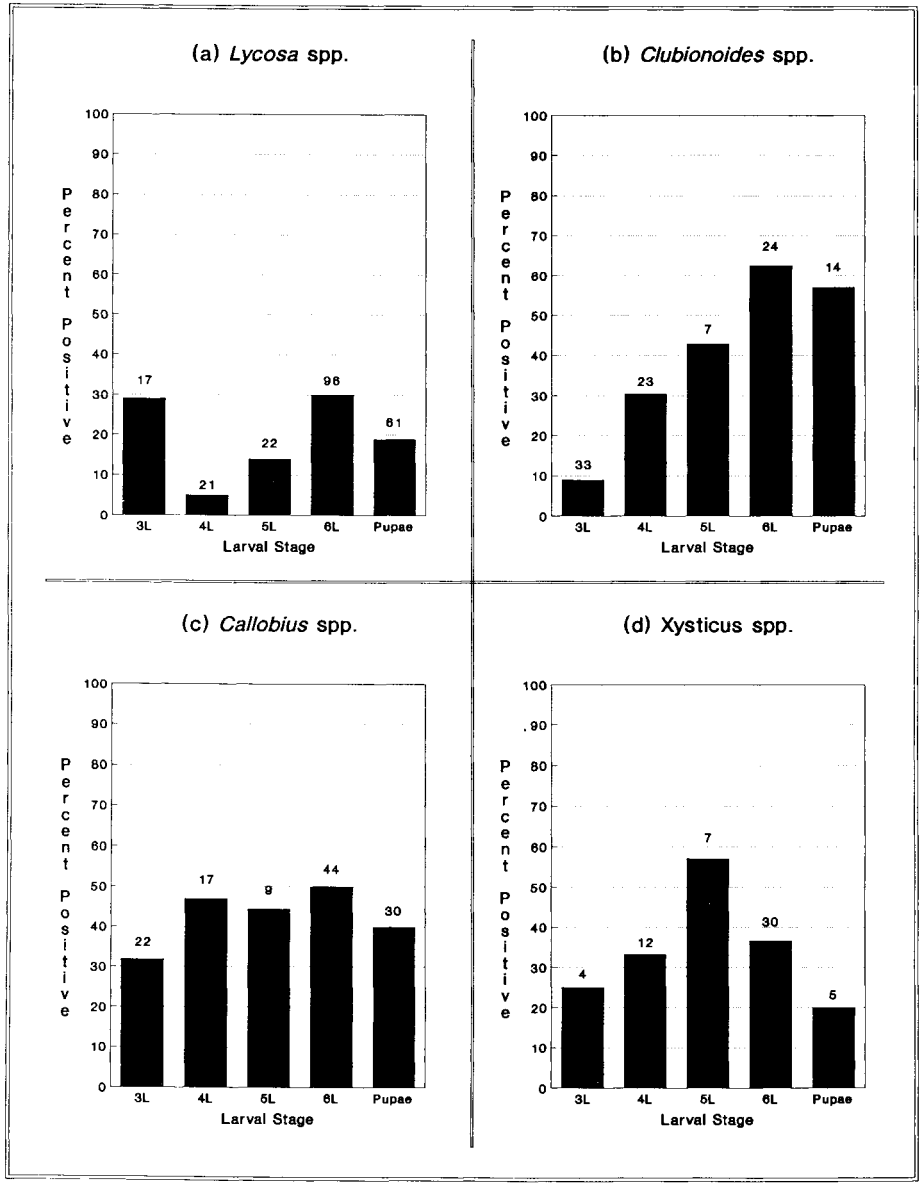

Fig. 1. Percent positive to ELISA for spider genera a) *Lycosa* spp. b) *Clubionoides* spp., c) *Callobius* spp. and d) *Xysticus* spp. by gypsy moth stage from third instar (3L) through pupa. Number tested given above each bar.

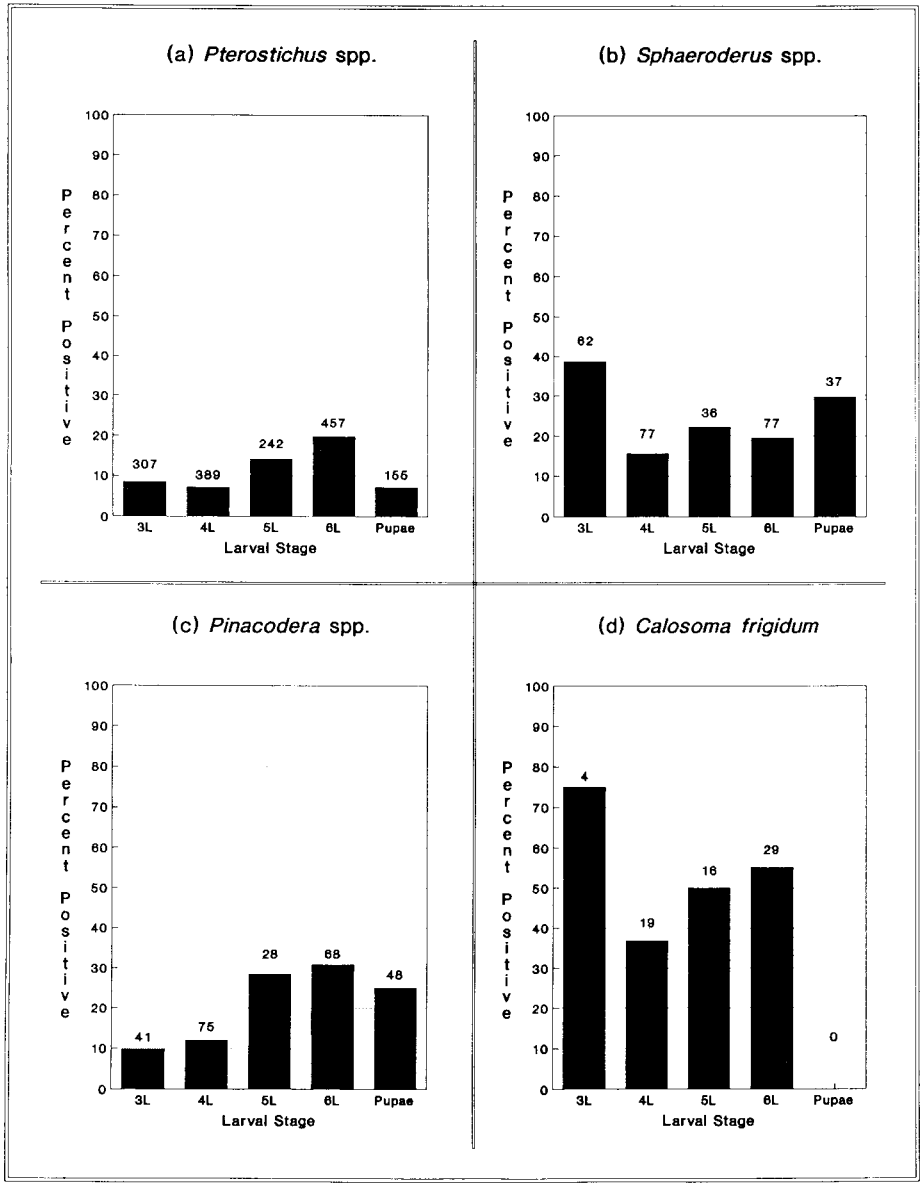

Fig. 2. Percent positive to ELISA for carabid beetle genera a) *Pterostichus* spp. b) *Sphaeroderus* spp. and c) *Pinacodera* spp. and d) *Calosoma frigidum* Kirby by gypsy moth stage from third instar (3L) through pupa. Number tested given above each bar.

Table 1. Number tested (No.) and percent positive (%+) by ELISA for common categories of arthropods compared by trap method. Less common categories listed at end of table.

CLASS Order Family	All traps combined No.	%+	Pitfall traps No.	%+	Tree bands No.	%+
CRUSTACEA						
Isopoda	115	31	58	24	57	39
CHILOPODA	59	17	56	18	3	0
DIPLOPODA	35	49	35	49	0	0
ARACHNIDA						
Aranea	1029	28	641	22	388	39
Phalangida	19	68	3	33	16	75
INSECTA						
Coleoptera						
Cantharidae	10	20	0	0	10	20
Carabidae	2778	14	2445	13	333	28
Elateridae	143	48	10	60	133	47
Lampyridae	70	63	3	67	67	63
Staphylinidae	95	1	91	1	4	0
Tenebrionidae	23	26	8	12	15	33
Dermaptera	115	21	0	0	115	21
Dictuoptera	18	17	6	17	12	17
Grylloptera						
Rhaphidiophoridae	114	51	105	51	9	44
Hemiptera						
Pentatomidae	17	12	15	13	2	0
Hymenoptera						
Formicidae	42	10	11	0	31	13

Additional families (number caught): Berytidae (1), Chrysomelidae (1), Cicindellidae (1), Histeridae (6), Melandryidae (2), Scarabaeidae (9), Silphidae (2) and Gryllidae (1). Positive results were present in the Berytidae, Chrysomelidae and six of the Scarabaeidae.

The Chilopoda is the only class considered to be completely predaceous. Contact with prey organisms is considered necessary before attack and prey is subdued using poison injected into the prey by the modified first pair of legs. Carrion feeding has not been documented in centipedes and the 17% positive results is probably an accurate indication of their gypsy moth larval feeding activity.

In the Arachnida both the Aranea and Phalangida have been documented as feeding on gypsy moth (Smith and Lautenschlager 1978). The high percent positive (68%) for the Phalangida was undoubtedly influenced by carrion feeding. This order is known to feed on a wide variety of food materials including live and dead insects, fungi, bird droppings and various plant materials (Edgar 1971).

The Aranea rarely attack prey that are not active (i.e. spruce budworm eggs, Jennings and Houseweart 1978) and, like centipedes, subdue their prey using poison fangs. The 28% positive results for this group undoubtedly represent true predation. However, the longevity by which food items are digested (up to seven days for *Pardosa sternalis* Thorell, McIver 1981) is sure to inflate this figure. Even so the percents positive for the genera and species encountered do provide a measure of the relative value of spiders as feeders of late instar gypsy moth larvae.

Aranea accounted for 22% of total arthropods caught with a majority (62%) from pitfall traps. Thirteen families (Table 2), 31 genera and 43 species (Appendix I) of Aranea were caught and tested. Positive test results were seen in 10 families, 28 genera and 31 species. Wandering or hunting spiders were more likely to be caught using the trap methods employed; sedentary web-spinning spiders (Araneidae, Theridiidae, Linyphiidae, etc.) were poorly represented or absent. The Clubionidae, Philodromidae and Salticidae were more commonly collected under tree bands and their percents positive were invariably higher than typical litter dwelling families such as Gnaposidae and Lycosidae. Even for spider families and species which were more abundantly caught in pitfall traps, the percents positive were sometimes more than twice as high under tree bands (i.e. Agelenidae, Thomisidae, *Zelotes fratris* Chamberlin and *Xysticus fraternus* Banks in Tables 2 and 3). However, the highest percent positive was 57% for *Haplodrassus bicornis* (Emerton), a gnaphosid collected primarily by pitfall traps.

Of the more abundant families, the highest percent positive (44%) was the cribellate Amaurobiidae which was represented in this study by a single species, *Callobius bennetti* (Blackwall). Nearly equal numbers were collected under tree bands and in pitfall traps (Table 3).

The proportion positive for the four most commonly collected genera of spiders from tree bands and pitfall traps during different larval instars of gypsy moth are shown in Figure 1. Of these four genera *Lycosa, Clubionoides* and *Xysticus* are active hunters while *Callobius* builds webs. *Callobius* had a high overall and relatively uniform percent positive (Fig. 1c); there was no significant difference in the comparisons between larval stages (Chi-square = 3.15, $P > 0.05$). Of the other genera, *Clubionoides* were all collected from treebands while nearly all *Lycosa* came from pitfall traps. Significant differences were found for *Lycosa* (Chi-square = 9.92, $P \leq 0.05$) and *Clubionoides* (Chi-square = 12.65, $P \leq 0.025$) but not for *Xysticus* (Chi-square = 1.22, $P > 0.05$).

Six orders and 19 families of insects were collected and tested (Table 1). The separation of carrion feeding from active predation in these categories was based primarily on known feeding behavior or preferences. We considered Dermaptera (21%) to be carrion feeders like the Diplopoda and Isopoda. The Blattellidae, or wood roaches, with 17% positive and Rhaphidiophoridae, or camel crickets, with 51% positive are of uncertain value as predators.

In the Coleoptera the two families with the highest percents positive (Elateridae 48% and Lampyridae 63%) have unknown gypsy moth larval predatory capabilities and deserve further investigation. Our results indicate that Staphylinidae do not generally feed on either live or dead gypsy moth larvae. The potential of the remaining families as predators of gypsy moth larvae cannot be determined based on the numbers collected in this study.

Table 2. Number tested (No.) and percent positive (%+) by ELISA for common spider families compared by trap method.

Family	All traps combined		Pitfall traps		Tree bands	
	No.	%+	No.	%+	No.	%+
ATYPIDAE	2	50	2	50	0	0
AMAUROBIIDAE	122	44	56	32	66	54
THERIDIIDAE	4	0	0	0	4	0
LINYPHIIDAE	1	0	0	0	1	0
AGELENIDAE	51	33	34	24	17	53
HAHNIIDAE	6	0	6	0	0	0
PISAURIDAE	3	33	2	0	1	100
LYCOSIDAE	297	22	291	22	6	33
GNAPHOSIDAE	218	24	146	20	72	33
CLUBIONIDAE	205	31	57	16	148	37
THOMISIDAE	76	37	41	24	35	51
PHILODROMIDAE	14	29	5	20	9	33
SALTICIDAE	30	33	1	0	29	34

Table 3. Number tested (No.) and percent positive (%+) by ELISA for the more common spider species compared by trap method.

Species	Pitfall traps		Tree bands	
	No.	%+	No.	%+
Callobius bennetti (Backwall)	54	33	65	54
Wadotes hybridus (Emerton)	10	30	2	0
Lycosa frondicola Emerton	29	28	1	100
L. gulosa Walckenaer	69	16	0	0
Schizocosa saltatrix Hentz	17	18	0	0
Trochosa terricola Thorell	39	13	2	0
Drassyllus niger (Banks)	35	6	3	100
Gnaphosa muscorum (L. Koch)	15	13	0	0
Haplodrassus bicornis (Emerton)	28	57	2	50
Harpyllus ecclesiasticus Hentz	3	0	15	27
Zelotes fratris Chamberlin	39	8	12	33
Aqroeca ornata Banks	28	18	1	0
Clubionoides excepta (L. Koch)	0	0	99	36
Strotarchus piscatoria (Hentz)	2	50	8	25
Xysticus elegans Keyserling	15	33	8	62
X. fraternus Banks	18	22	10	50
Habrocestum pulex Hentz	1	0	20	50

The Carabidae was the most abundant family collected comprising 59% of all organisms and with 14% positive by ELISA. Most species are opportunistic predators with carrion feeding possible but the proportionate share unknown. Only four immature carabids were tested, all with negative results.

Positive test results were found in 14 of the 22 genera collected (Appendix II) and, except for *Chlaenius,* these were the most abundant genera. Of the 48 species collected 26 had positive test results (Appendix II) and again these were generally the more abundant. Most of the carabids were collected in pitfall traps (88%). Only four species were more commonly collected from tree bands than pitfall traps (Table 4). Pitfall trap/tree band comparisons show that four of the five common species collected by both methods had higher percents positive under tree bands, although these differences were not significant statistically (Chi-square = 7.52, $P > 0.05$).

Table 4. Number tested (No.) and percent positive (%+) by ELISA for the more abundant species of ground beetles compared by trap method.

Species	Pitfall traps		Tree bands	
	No.	%+	No.	%+
Sphaeroderus canadensis Chaudoir	85	36	0	0
S. lecontei Dejean	204	20	0	0
Carabus nemoralis Müller	29	38	0	0
Calosoma frigidum Kirby	18	33*	50	56*
Notiophilus aeneus Herbst)	57	3*	7	29*
Patrobus longicornis (Say)	13	8	0	0
Myas cyanescens Dejean	15	7	0	0
Pterostichus adoxus Say	56	11	0	0
P. lucublandus (Say)	62	24	0	0
P. pensylvanicus LeConte	595	18	1	0
P. mutus Say	782	7	1	0
P. melanarius Illiger	20	5	0	0
P. stygicus Say	30	13	0	0
Synuchus impunctatus (Say)	132	14	1	0
Platynus decentis Say	12	0*	29	34*
Harpalus rufipes DeGeer	35	6	0	0
H. viduus LeConte	19	21	0	0
Dicaelus dilatatus Say	62	5	0	0
D. politus Dejean	103	4	0	0
Pinacodera limbata Dejean	4	25*	64	16*
P. platicollis Say	17	6*	175	24*
Cymindis cribricollis Dejean	24	8	0	0

* Differences in percents positive between tree bands and pitfall trap collections were not significant using Chi-square at $P = 0.05$.

Calosoma frigidum had the highest percent positive (50%) of any carabid species. Figure 2d presents a comparison of the percents positive by ELISA for treebands and pitfall traps for *C. frigidum* during gypsy moth larval instars 3-6 and the pupal stage. No adult *C. frigidum* were collected during the pupal stage and no significant differences between larval instars (Chi-square = 0.99, $P > 0.05$) were

found. Three other genera were similarly compared (Fig. 2). No significant differences were found for *Sphaeroderus* (Chi-square = 7.42, $P > 0.05$) and, even though the percents positive were generally lower for *Pterostichus* and *Pinacodera,* significant differences were found (Chi-square = 25.35, $P \leq 0.01$ and Chi-square = 11.67, $P \leq 0.025$, respectively). Percentages were highest at instar six for these two genera.

Discussion

Carabid beetles and spiders were shown to be the arthropod groups most commonly trapped in areas where late instar gypsy moth larvae rest or select for pupation. The carabid beetles were dominant in edaphic habitats with relatively few species showing arboreal preferences. Campbell *et al.* (1977) has postulated that where gypsy moth larval resting places abound on trees, fewer larvae enter the litter layer for resting and pupation. In our study there were few resting or pupating locations on the tree boles and the numbers of gypsy moth present caused many larvae to move into the litter where Campbell *et al.* suggest much of the predation by mammals and arthropods occurs. The numbers of carabid beetles caught in the pitfall traps indicates they may be the most important group of arthropod predators in the litter, yet the range in the percents positive indicates only certain species may actually feed on gypsy moth. For gypsy moth larvae that remain on the trunks of trees, we believe that spiders may play a more important role than carabid beetles. The only carabid beetle or spider species present and with high percents positive by ELISA in both locations were *C. frigidum* and *C. bennetti.*

The prey catching and feeding habitats of spiders make serology especially valuable for determining gypsy moth feeding potential but the extended digestive process requires a more elaborate experimental design than used in this study. The relative importance of the species tested provides some insight into their gypsy moth predatory potential. Our data suggest that families and species found on tree trunks are more likely to encounter and feed on gypsy moth larvae while edaphic hunting species either have less availability or less preference for gypsy moth larvae as prey. During low population densities fewer gypsy moth larvae would be expected to enter the litter resulting in an even higher proportion of spiders feeding on gypsy moth larvae to be found on tree boles. The more important arboreal species include *Clubionoides excepta* (L. Koch) (most abundant species on tree trunks), *C. bennetti* (abundant on both tree trunks and in litter), *Habrocestum pulex* Hentz (50% tested positive on tree trunks) and *Xysticus elegans* Keyserling (44% tested positive). *Callobius bennetti* is the most sedentary of these four species, building irregular webs in crevices associated with bark or rocks (Kaston 1948), places in which gypsy moth larvae often select for resting and pupation. The only litter inhabiting species of importance seems to be *H. bicornis* (57% tested positive from pitfall traps). In general the Lycosidae do not appear to be important because of their relatively low percent positive and their reluctance to feed in the laboratory. Certain species, i.e. *Lycosa frondicola* Emerton at 30%, may be worth further investigation.

Approximately one half of the carabid beetle species collected in this study showed evidence of gypsy moth proteins in their guts and these were generally the more abundant species. Arboreal species appear more likely to feed on gypsy

moth than litter dwellers. A wide range in acceptability of gypsy moth larvae as food by various carabids is apparent even in species of similar size and abundance. *Calosoma frigidum* and *Dicaelus dilatatus* Say are both over 20 mm in body length and numbers caught are nearly equal (68 versus 62 respectively) yet the percents positive are widely divergent (50% versus 5% respectively). Size may also be a factor as the two species with the highest percents positive (*C. frigidum* and *Carabus nemoralis* Müller are both over 20 mm long. Two species, *Pterostichus mutus* and *P. pensylvanicus,* readily accepted gypsy moth larvae in the laboratory. Even though their percents positive were low (7 and 18% respectively) they were the most abundant carabid beetle species (50% of total carabid beetles) and appear to be reasonably good targets for further study. Carrion feeding has not been documented for *C. frigidum;* however, other species of carabid beetles may be scavengers and this factor must be taken into account when the percents positive are used.

Calosoma frigidum is possibly the best documentated carabid beetle predator of gypsy moth collected in this study (Burgess and Collins 1917, Smith and Lautenschlager 1978, 1981) and it is not surprising that it also had the highest percent positive of any carabid beetle species. Members of this genus are known to prey on caterpillars, and adults of *C. frigidum* are known to climb trees. Early hibernation of this species probably explains their absence during the gypsy moth pupation period in late July.

Sphaeroderus canadensis Chaudoir and *S. lecontei* Dejean have prolonged head and mouthparts, an adaptation for feeding on shell bearing snails, and represent 10% of all carabids collected. Percents positive were 36 and 20 respectively and, while no significant differences were found for this genus between the various stages of gypsy moth, of the four genera presented in Fig. 2, they had the highest percent positive during the pupal period, implying that pupae may be especially vulnerable to attack.

Many of the genera and species of carabid beetles and spiders were completely negative to ELISA. The reasons for these negative results are varied and may be the result of relative abundance, spatial or temporal distribution or prey preference of the carabid beetle or spider species. An even larger proportion of negative results might be expected during low gypsy moth population levels when predators may switch to more abundant alternative food sources. Thus species level observations of predator-prey interactions is important and generalizations based on family or higher taxonomic categories can be misleading when a single prey item such as gypsy moth is considered.

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Appendix I. Spider genera and species with number collected. Species with * with at least one individual testing positive by ELISA.

ATYPIDAE	*Sphodros niger* (Hentz)	2*
AMAUROBIIDAE	*Callobius bennetti* (Blackwall)	119*
	C. sp.	3*
THERIDIIDAE	*Dipoena nigra* (Emerton)	2
	Theridion sp.	2
LINYPHIIDAE	*Prolinyphia marginata* (C. L. Koch)	1
AGELENIDAE	*Agelenopsis* sp.	1*
	Coelotes sp.	2
	Coras medicinalis (Hentz)	6*
	C. sp.	8*
	Cryphoeca sp.	4*
	Wadotes hybridus (Emerton)	12*
	W. sp.	18*
HAHNIIDAE	*Neoantistea agilis* (Keyserling)	1
	N. magna (Keyserling)	5
PISAURIDAE	*Pisaurina mira* (Walckenaer)	3*
LYCOSIDAE	*Arctosa rubicunda* (Keyserling)	4
	Lycosa frondicola Emerton	30*
	L. gulosa Walckenaer	69*
	L. sp.	118*
	Pardosa moesta Banks	2
	Shizocosa avida (Walckenaer)	1*
	S. bilineata Emerton	1*
	S. saltatrix (Hentz)	17*
	S. sp.	5*
	Trochosa avara Keyserling	1
	T. terricola Thorell	41*
	T. sp.	8*
GNAPHOSIDAE	*Callilepis pluto* Banks	4*
	Drassodes sp.	3*
	Drassyllus niger (Banks)	38*
	Gnaphosa muscorum (L. Koch)	15*
	G. sp.	2
	Haplodrassus bicornis (Emerton)	30*
	H. hiemalis (Emerton)	1
	H. signifer (C. L. Koch)	1*
	Herpyllus ecclesiasticus Hentz	18*
	H. sp.	39*
	Poecilochroa copulata (Walckenaer)	5
	P. sp.	1
	Zelotes fratris Chamberlin	51*
	Z. sp.	10*
CLUBIONIDAE	*Agroeca ornata* Banks	29*
	A. sp.	2
	Castianeira cingulata (C. L. Koch)	2
	C. sp.	13*
	Clubiona mixta Emerton	3*
	C. obesa Hentz	6*
	C. sp.	9*
	Clubionoides excepta (L. Koch)	99*
	C. sp.	2
	Phrurotimpus alarius (Hentz)	16
	P. borealis Emerton	13*
	Strotarchus piscatoria (Hentz)	10*
	S. sp.	1
THOMISIDAE	*Coriarachne versicolor* Keyserling	1*
	C. versicolor-utahani	9*
	C. sp.	8*
	Xysticus elegans Keyserling	23*
	X ferox (Hentz)	6*
	X fraternus Banks	28*
	X sp.	1
PHILODROMIDAE	*Philodromus rufus vibrans* Dondale	2
	P. vulgaris (Hentz)	4*
	P. sp.	6*
	Thanatus sp.	2*
SALTICIDAE	*Habrocestum pulex* (Hentz)	21*
	Maevia inclemens (Walckenaer)	9

Appendix II Carabid beetle species with number collected. Species with * with at least one individual testing positive by ELISA.

Sphaeroderus canadensis (Chaudoir)	85*	*Harpalus rufipes* DeGeer	35*
S. lecontei Dejean	204*	*H. pensylvanicus* DeGeer	2
Carabus nemoralis Müller	29*	*H. erythropus* Dejean	4
Calosoma frigidum Kirby	68*	*H. lewisi* (LeConte)	2
Notiophilus aeneus Herbst	64*	*H. laticeps LeConte*	8
Patrobus longicornis Say	13*	*H. viduus* LeConte	19*
Myas cyanescens Dejean	15*	*H. spadiceus* Dejean	1
Pterostichus adoxus Say	56*	*H. indigens* Casey	3*
P. lucublandus (Say)	62*	*H. herbivagus* Say	4*
P. pensylvanicus LeConte	595*	*H. fallax* LeConte	4
P mutus Say	783*	*Amphasia interstitialis* (Say)	6
P. melanarius Illiger	20*	*Anisotarsus nitidipennis* (LeConte)	3
P. stygicus Say	30*	*Episcopellus autumnalis* Say	3
P. coracinus (Newman)	3*	*Diplocheila assimilis* LeConte	1
P. leconteianus Lutshnik	2	*Dicaelus elongatus* Bonelli	1
Synuchus impunctatus (Say)	133*	*D. dilatatus* Say	62*
Olistophus parmatus (Say)	3	*D. politus* Dejean	103*
Agonum retractum (LeConte)	7	*Chlaenius tricolor* Dejean	1*
A. cupripenne Say	1	*Pinacodera limbata* Dejean	68*
A. muelleri (Herbst)	3	*P. platicollis* Say	192*
A. fidele Casey	1	*Cymindis americana* Dejean	4
A. placidum Say	1	*C. cribricollis* Dejean	24*
Platynus decentis Say	41*	*C. neglecta* Haldeman	3
Amara cupreolata Putzeys	1	*Progaleritina janus* (F.)	1