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HOST AND HABITAT USE BY PARASITOIDS (HYMENOPTERA: PTEROMALIDAE) OF HOUSE FLY AND STABLE FLY (DIPTERA: MUSCIDAE) PUPAE

D. L. Olbrich1 and B. H. King1

ABSTRACT

House fly and stable fly pupae were collected during the summer from a dairy farm in northern Illinois. *Spalangia nigroaenea* accounted for most of the parasitoids recovered from house flies. *Spalangia nigra*, *S. endius*, *Muscidifurax* spp., and *S. nigroaenea* accounted for most of the parasitoids from stable flies. The majority of flies were house flies late in the summer and stable flies early in the summer. Higher percentages of house flies tended to be in samples containing lower substrate moisture and higher substrate temperature. Parasitism of stable flies started earlier and peaked weeks before that of house flies, with overall parasitism highest from mid- to late-summer. Parasitism of house flies, but not stable flies, differed significantly among habitats, being greater in calf hutch samples. Hymenopterous parasitoids from house flies tended to include a greater percentage of *S. nigroaenea* (and a lower percentage of *Muscidifurax* spp.) in calf hutches versus drainage or edge habitats and in substrates consisting of mostly wood shavings versus mostly manure. Within samples, differential parasitism of fly species was not detected for *S. nigroaenea*, *S. endius*, or *Muscidifurax* spp.; but *S. nigra* preferentially parasitized stable flies.

Controlling filth flies such as stable flies, *Stomoxys calcitrans* (L.) (Diptera: Muscidae), and house flies, *Musca domestica* L. (Diptera: Muscidae), on farms is important. Stable fly adults bite livestock and humans and decrease feed efficiency in beef cattle and the production of milk in dairy cattle (e.g., Bruce and Decker 1958; Campbell et al. 1987). House flies do not bite but can spread human and animal pathogens (Kettle 1984; Burgess 1990). When abundant, filth flies might lead to nuisance lawsuits against producers (Seymour and Campbell 1993).

Filth flies commonly are controlled, at least in part, with insecticides. However, both house flies and stable flies have demonstrated resistance to several insecticides (e.g., Cilek and Greene 1994; Keiding 1999), some insecticides may decrease populations of natural enemies (Geden et al. 1992), and there is public concern about pesticide residues in food.

Although biological control of filth fly populations with hymenopterous parasitoids has many advantages over insecticides, currently the direct costs (i.e., excluding health and environmental costs) are less with insecticides (Andress and Campbell 1994). Better understanding of the ecology and behavior of both filth flies and their potential biological control agents may improve the efficacy of biological control and may help maximize naturally occurring control (Smith and Rutz 1991a, Jones and Weinzierl 1997). For example, by knowing habitat and host preferences of parasitoids, farmers could choose parasitoid species in relation to habitats and fly species that are particularly troublesome (Smith and Rutz 1991a).

Within the United States, the relative occurrence of house flies versus stable flies on different parts of dairy farms and in different substrates has

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been examined in Nebraska (Meyer and Petersen 1983). Microhabitat effects on parasitoids of these flies have been studied on dairies in Florida (Greene et al. 1989), California (Meyer et al. 1991), New York (Smith and Rutz 1991a; Smith and Rutz 1991b), Nebraska (Seymour and Campbell 1993), and Denmark (Skovgård and Jespersen 1999). Variation among farms and years has been documented (e.g., Meyer et al. 1991; Smith and Rutz 1991a; Jones and Weinzierl 1997). Our focus in the present study was on variation among habitats within a single farm in a single season. Specifically, we examined house flies, stable flies and their hymenopterous parasitoids in the summer on a dairy farm in northern Illinois in relation to three different habitats, calf hutches, drainage areas, and edges (e.g., fencelines). Habitat differences in moisture, temperature, and substrate type were characterized. We then examined relationships of these variables and week with 1) the percentage of filth fly populations that were house flies versus stable flies, 2) parasitism rate of house flies and stable flies and 3) hymenopterous parasitoid species composition.

MATERIALS AND METHODS

Fly pupae collection. The study was conducted on a dairy farm located in Harvard, Illinois, in the northeastern part of the state. The farm was about 200 continuous acres, but the area sampled was about 20 acres. Three hundred cattle were maintained on a pasture-rotation and confinement schedule (Olbrich 2002). Pesticides were not applied during this study or in the nine months prior to this study.

Sampling was conducted in 2000 from the second week in June to the first week in September. Fly pupae were collected weekly from two sites within each of three habitats (calf hutches, drainage areas, and edges) for a total of six samples each week. (During the last week, one rather than two samples was collected from drainage and edge habitats; however, excluding this last week from analyses had negligible effects.) The calf hutches were blue plastic dome-like structures (168 cm × 122 cm × 117 cm) that housed one calf (0-3 months of age), each with a large open entryway in front, two side windows (one on each side), and two rear windows. The drainage areas were low-lying areas that collected water when rain was sufficient. The edges were areas along fences and feed bunks, where manure, soil, and/or feed accumulated.

At each site, samples were collected for approximately 1 h or until about 80 pupae were obtained. Substrate was collected with a trowel within a 20 cm radius of where fly pupae were seen and up to 10 cm deep (Jones and Weinzierl 1997, Smith and Rutz 1991b).

To increase statistical power, substrates were lumped into two frequent categories, mostly manure versus mostly shavings (8 samples that contained only spoiled feed were excluded from these analyses). Beginning in July, percentage moisture was measured by placing a Kelway soil tester (Kel Instruments Co., Wyckoff, NJ) into the center of each sample. Temperature was measured by inserting a general purpose mercury thermometer into the substrate prior to digging and leaving it there for approximately 1 min. Such simple measurement techniques were chosen to determine whether they might be useful to farmers in deciding which parasitoid species to release.

House fly and stable fly pupae were individually extracted from the substrate. Light red pupae were excluded because such young pupae would have had minimal to no exposure to parasitism. Each pupa was placed in a closed test tube to prevent the earlier emerging hymenopterous parasitoids from parasitizing any as yet unemerged hymenopterous parasitoids (King 1997). Tubes were held at room temperature, and fly pupae and emergent parasitoids were identified (Skidmore 1985, Rueda and Axtell 1985, Gibson 2000). *Muscidifurax raptor* Girault and Sanders and *M. zaraptor* Kogan and Legner were combined as...
Muscidifurax spp. (following Jones and Weinzierl 1997, Smith et al. 1987). Vouchers were deposited at the Illinois Natural History Survey Center for Biodiversity, catalog numbers “Insect Collection 12,272 through 12,309.” Only intact fly pupae were included, i.e., those from which a parasitoid, fly or nothing emerged after collection (following Jones and Weinzierl 1997). Dented fly pupae were included because flies and parasitoids sometimes emerged from them.

Analyses. Statistical analyses were performed using SPSS 10.0 for Windows, and all tests used a significance level of 0.05. Means are presented with standard errors; when values are not presented with standard errors, they were not computed per sample, per habitat or per week but rather by summing across all fly pupae collected. We tested for 1) relationships among environmental variables, 2) relationships between environmental variables and the percentage of house fly versus stable fly pupae, and 3) effects of environmental variables on parasitism rate of each fly species. Parasitism rate was calculated as the number of fly pupae from which parasitoids emerged after collection divided by the number of all intact fly pupae collected. Data were analyzed primarily by two-way ANOVA (analyses of variance) and correlations (see Results and Discussion). However, effects of week and habitat on percentage parasitism were analyzed by nonparametric tests because both log and arcsine transformations were unsuccessful at normalizing all cells. First, we tested for an interaction using an extension of the Kruskal-Wallis for a two factor analysis (Zar 1996). Because this analysis requires equal cell sizes, weeks without 2 samples per habitat were excluded (e.g., weeks for which a sample lacked that particular fly species). When no significant interactions were found, main effects were then tested using the full data set with one factor Kruskal-Wallis.

Whether parasitoids were recovered more often from one host species than from the other was examined from two perspectives, a collection-wide perspective and a within-sample perspective. The collection-wide perspective pooled all samples into one large sample. Then a chi-square test was used to compare two ratios to each other. One ratio was the number of parasitoids from house flies to the number from stable flies; the other ratio was the number of intact house fly pupae to the number of intact stable fly pupae. In contrast, the within-sample perspective used a paired t-test to compare the percentage of parasitoids in a sample that emerged from house flies to the percentage of intact fly pupae in that same sample that were house flies. The within-sample perspective reduces the number of environmental effects influencing host usage, e.g., week and habitat, though it does not eliminate microhabitat effects, e.g., differences in depth that the host species were found.

In addition to analyzing our own data, we also analyzed data on collections of intact house fly and stable fly pupae from two published studies (Seymour and Campbell 1993, Greene et al. 1989) in order to examine host species usage in those studies. Chi-square tests were used to test whether the number of parasitoids recovered from house flies relative to the number recovered from stable flies was different from that expected if recovery was proportional to the relative abundance of the fly species among all intact house fly and stable fly pupae collected.

RESULTS AND DISCUSSION

Habitat differences in moisture, temperature, and substrate. A two-way ANOVA on percentage moisture revealed no significant interaction between week and habitat ($F = 0.99$, df = 19, 27, $P = 0.49$). Percentage moisture differed significantly among the three habitats ($F = 8.99$, df = 2, 19, $P = 0.002$). Specifically, the drainage samples were usually the wettest and were never the driest, averaging 62% ± 5% (6 – 95%, N = 19), versus 46% ± 5% (15 – 88%, N = 18) for the edge and 38% ± 4% (10 – 71%, N = 20) for the calf hutch. Percentage moisture also differed among weeks ($F = 2.57$, df = 9, 18, $P = 0.042$), tending to be lowest midsummer.
A two-way ANOVA on temperature revealed a significant interaction between week and habitat ($F = 3.29$, $df = 22, 36, P = 0.001$). Temperature of the substrate differed among habitats ($F = 12.18$, $df = 2, 22, P < 0.001$) and among weeks ($F = 2.25$, $df = 11, 22, P = 0.051$). Generally the calf hutch substrate was the hottest. During the study, temperature initially increased, then leveled off and was just beginning to decrease at the end of the study. Across all samples, moisture and temperature were negatively correlated ($r = -0.29, N = 56, P = 0.032$).

Substrate type differed significantly among the three habitats ($\chi^2 = 53.75$, $df = 2, P < 0.001$), particularly for calf hutch versus drainage and edge habitats ($\chi^2 = 53.17$, $df = 1, P < 0.001$). Most samples from calf hutch consisted primarily of wood shavings, whereas most samples from drainage and edge habitats were primarily manure. Manure substrates had a significantly higher moisture content than wood shavings substrates ($56 \pm 4.1\%$ versus $38 \pm 3.4\%$; $t = 3.21$, $df = 49, P = 0.002$) and significantly lower temperature ($25 \pm 4^{\circ}C$ versus $28 \pm 4^{\circ}C$; $t = 3.85$, $df = 66, P = 0.001$).

**Percentage house flies versus stable flies.** 66% of the 4370 intact house fly and stable fly pupae that were collected were house flies. This is slightly less than reported for a Nebraska dairy (Meyer and Petersen 1983), Manitoba dairies (McKay and Galloway 1999), and a poultry house in Indiana (King 1990).

A two-way ANOVA on percentage house flies revealed no significant interaction between habitat and week ($F = 0.72$, $df = 24, 37, P = 0.80$). There was a significant week effect ($F = 3.79$, $df = 12, 24, P = 0.003$): stable flies made up a greater percentage of the fly pupae earlier in the summer and house flies a greater percentage later in the summer (Fig. 1). This temporal pattern is similar to results from a Nebraska dairy (Meyer and Petersen 1983), from cattle confinement in Nebraska (Seymour and Campbell 1993), and for adult flies from California dairies (Meyer et al. 1990).

No statistically significant difference was detected in the percentage of house flies versus stable flies among the three different habitats ($F = 3.18$, $df = 2, 24, P = 0.059$). However, the trend was that the percentage of house flies was greatest from the calf hutch and least from edges, perhaps due to moisture.

Figure 1. Mean ± SE percentage of house fly pupae versus stable fly pupae collected among all weeks.
differences among habitats. Likewise, the percentage of house flies decreased with increasing percentage moisture of the substrate \( (r^2 = 0.14; F = 8.58, df = 1, 55, P = 0.005; y = -0.58x + 97.51) \) and increased with increasing temperature \( (r^2 = 0.12; F = 9.44, df = 1, 73, P = 0.003; y = 3.12x - 19.16). \) In a Nebraska dairy, stable flies were more common than house flies in drainage ditches, and house flies were more common than stable flies in oat silage and spilled feed, perhaps related to moisture (Meyer and Petersen 1983). In our survey, percentage of house flies did not differ significantly between wood shaving substrate versus manure substrate \( (x = 6.9 \pm 55 \pm 5.7; t = 1.84, df = 66, P = 0.071). \)

**Percentage parasitism.** Adult flies emerged from 48% of house fly pupae and 45% of stable fly pupae. Nothing emerged from 35% of house fly pupae and 43% of stable fly pupae.

Summing across all samples, a significantly greater percentage of house flies than stable flies were parasitized \( (17\% \text{ versus } 12\%; \chi^2 = 18.01, df = 1, P < 0.001). \) However, when we compared parasitism of house flies versus stable flies within samples (i.e., using sample versus pupa as the statistical sampling unit), there was no statistically significant difference \( (11 \pm 2.1\% \text{ versus } 13 \pm 2.7\%; \text{paired } t\text{-test: } t = 0.69, df = 60, P = 0.49). \) The observation that house fly parasitism was greater than stable fly parasitism across all samples was due at least in part from low parasitism early in the season when stable flies were more prevalent than house flies. Other studies in which naturally occurring fly pupae were collected have found similar parasitism rates (e.g., Skovgård and Jespersen 1999, McKay and Galloway 1999; Smith et al. 1987). An overall trend of greater parasitism of house flies than stable flies was found in a 3 yr study of cattle feedlots in Illinois (Jones and Weinzierl 1997) and in two studies of confined livestock in Nebraska (Petersen and Meyer 1983a; Seymour and Campbell 1993). However, there was no significant difference in parasitism rates for house flies versus stable flies on California dairies studied by Meyer et al. (1990).

Parasitism varied significantly among weeks for both house flies \( (H = 44.86, df = 12, P < 0.001) \) and stable flies \( (H = 25.41, df = 12, P = 0.012). \) Parasitism rates were highest mid to late summer \( (\text{Figs. 2-3}) \), consistent with earlier studies (e.g., Petersen and Meyer 1983a; Seymour and Campbell 1993). We found no statistically significant interaction between habitat and week for percentage parasitism of house flies \( (H = 6.91, df = 16, P > 0.95) \) or stable flies \( (H = 3.00, df = 8, P > 0.90). \)

Parasitized stable flies were collected before parasitized house flies, and peak parasitism of stable flies occurred weeks before the peak for house flies \( (\text{Fig. 2-3}) \), similar to results from cattle confinements in Nebraska in two different years (Seymour and Campbell 1993). The reason could be the greater proportion of stable flies earlier in the summer than later in the summer (Seymour and Campbell 1993; this study).

Parasitism of house flies varied significantly among habitats \( (\text{Table 1}) \), being greater in calf hutches than in edge samples. Parasitism of stable flies did not vary significantly among habitats \( (\text{Table 1}) \). Parasitism did not differ significantly between wood shaving substrate versus manure substrate for either house flies \( (19 \pm 4.1 \text{ versus } 10 \pm 2.4; U = 408.5, N = 66, P = 0.12) \) or stable flies \( (12 \pm 5.6 \text{ versus } 14 \pm 3.2; U = 315.5, N = 58, P = 0.17) \). We found no significant relationship between house fly or stable fly parasitism and percentage of moisture of substrate \( (F = 1.85, df = 1, 54, P = 0.18; F = 1.18, df = 1, 45, P = 0.28) \). Percentage of parasitism increased significantly with temperature for house flies \( (r^2 = 0.32; F = 16.01, df = 2, 68, P < 0.001; y = 74 - 7.35x + 0.19x^2) \) but not for stable flies \( (F = 0.15, df = 1, 63, P = 0.70). \)

Habitat-substrate effects on parasitism have also been reported for Florida dairies (Greene et al. 1989), but not for California dairies (Meyer et al. 1991); and for Nebraska cattle confinements there were substrate effects on parasitism of house flies, but not of stable flies (Seymour and Campbell 1993). Smith and
Figure 2. Weekly mean of the percentage parasitism of house fly pupae by different hymenopterous parasitoid species.

Figure 3. Weekly mean of the percentage parasitism of stable fly pupae by different hymenopterous parasitoid species.
Rutz (1991a, b) found that the incidence of parasitism in New York dairies varied with substrate, exposure, and habitat. However, differences in methods make comparison with our study difficult.

Parasitoid species composition. House flies were predominantly parasitized by *S. nigroaenea* (72%) and *Muscidifurax* spp. (14%) (Table 2). *S. nigroaenea* accounted for a greater percentage of the parasitism of house flies in samples from calf hutches than in samples from drainage or edge habitats, whereas the reverse was true for *Muscidifurax* spp. (Fig. 4). Similarly, more of the parasitism of house flies was by *S. nigroaenea* in wood shaving substrates than in manure substrates (71 ± 7.4% versus 31 ± 7.7%; *t* = 3.70, df = 40, *P* = 0.001), whereas for *Muscidifurax* spp. it was the reverse (33 ± 8.5%, *N* = 23 versus 8 ± 4.0%, *N* = 19; *U* = 144.5, *P* = 0.035). Our finding of *Muscidifurax* spp. in different substrates than *S. nigroaenea* is consistent with results from California dairies (Meyer et al. 1991) and New York dairy farms (Smith and Rutz 1991a). As in our study, on the New York dairy farms *S. nigroaenea* was the predominant parasitoid species from calf hutches.

In contrast to house flies, most stable fly parasitism was fairly evenly divided among four species, *S. nigroaenea*, *Muscidifurax* spp., *S. nigra* and *S. endius* Walker (Table 2). The predominant species of parasitoid varies among surveys of naturally occurring fly pupae on dairies (Greene et al. 1989; Meyer et al. 1990, 1991, Meyer and Petersen 1982; Petersen and Meyer 1983a; Smith et al. 1987, McKay and Galloway 1999). The fact that *S. nigroaenea* did not dominate from stable flies contrasts with a study of Illinois cattle feedlots (Jones and Weinzierl 1997).

Across both fly species, *S. nigroaenea* and *Muscidifurax* spp. tended to be found throughout the summer, whereas *S. nigra* were only found in early- to mid-summer (Figs. 2-3). *S. cameroni* Perkins was relatively uncommon and *Nasonia vitripennis* (Walker) was absent from both fly species, similar to Jones and Weinzierl’s (1997) study of Illinois cattle feedlots. *N. vitripennis* was also uncommon on dairies in Maryland and New York (Smith and Rutz 1991a, Geden et al. 1992). One factor contributing to the low incidence of *N. vitripennis* in these studies may be its preference for much drier substrates (Smith and Rutz 1991c).

One small percentage of the *S. nigroaenea* appear to have been in some type of delayed development (see also Merchant et al. 1987, Petersen and Meyer 1983b). Live individuals were found in the test tubes 4 mo. after their hosts had been collected, yet at 26°C, development duration and maximum longevity each average less than 1 mo. (Ramsdell 1985).

Table 1. Percentage of Parasitism of House Fly Pupae and Stable Fly Pupae from Three Habitats from a Dairy Farm in Northern Illinois in 2000.

<table>
<thead>
<tr>
<th></th>
<th>House fly</th>
<th>Stable fly</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean ± SE</td>
<td>N</td>
</tr>
<tr>
<td>Calf hutch</td>
<td>20 ± 4.5a</td>
<td>24</td>
</tr>
<tr>
<td>Drainage</td>
<td>11 ± 2.9ab</td>
<td>24</td>
</tr>
<tr>
<td>Edge</td>
<td>7 ± 2.8b</td>
<td>24</td>
</tr>
</tbody>
</table>

*H* = 6.39, *df* = 2, *P* = 0.041

Means within a column followed by different letters are significantly different by follow up Mann-Whitney *U*, *P* < 0.05

Rutz (1991a, b) found that the incidence of parasitism in New York dairies varied with substrate, exposure, and habitat. However, differences in methods make comparison with our study difficult.
Host species usage. Parasitoids emerging from one host species more than from another can logically result from females preferring to oviposit in one host species, from differences in parasitoid survival between host species, or from differences among host species in their habitat preferences (e.g., in depth, moisture, temperature, or type of substrate) and female parasitoids preferring the habitat of one host species over the other.

Collection-wide, *S. nigroaenea* were recovered from house flies significantly more often than expected. This may be because *S. nigroaenea* was absent early in the summer when stable flies make up a greater percentage of the fly pupae. In contrast, no differential parasitism of house flies was observed when we controlled for host species availability on a sample by sample basis (Table 3). In confined cattle operations in Nebraska, collection-wide *S. nigroaenea* was also preferentially from house flies in both years studied (new analysis of data from Seymour and Campbell 1993: \( \chi^2 = 220.86, df = 1, P < 0.001 \); \( \chi^2 = 220.86, df = 1, P < 0.001 \)). Laboratory experiments suggest that female *S. nigroaenea* preferentially oviposit in stable flies (Ramsdell 1995).

*S. nigra* were recovered more frequently from stable flies than from house flies within samples and collection-wide (Table 3), similar to a study of confined livestock in Nebraska (Petersen and Meyer 1983a). This suggests for biological control that *S. nigra* may be more suitable to control stable flies than house flies and should be released earlier in the season when stable flies are more abundant.

Within samples *S. cameroni* were recovered from house flies significantly, but only very slightly, more frequently than expected (Table 3). Collection-wide, the pattern was the same, but it was not statistically significant (Table 3).
Figure 4. Mean percentage of parasitism of house fly pupae that was by different hymenopterous parasitoid species, among habitats.

Figure 5. Mean percentage of parasitism of stable fly pupae that was by different hymenopterous parasitoid species, among habitats.
Collection-wide there also was no statistically significant host preference on Florida dairies (new analysis of data from Greene et al. 1989: \( \chi^2 = 0.012, df = 1, P = 0.91 \)). On Nebraska cattle confinements there was no statistically significant host preference in either year studied but collection-wide the trend was for a greater proportion to come from stable flies (new analysis of data from Seymour and Campbell 1993: \( \chi^2 = 0.54, df = 1, P = 0.46; \chi^2 = 2.86, df = 1, P = 0.091 \)). Among fly pupae from an Indiana poultry house, *S. endius* and *S. cameroni* both showed no preference for house flies or for stable flies (King 1990).

*Muscidifurax* spp. showed no significant difference in host species usage either within samples or collection-wide (Table 3). In contrast, *Muscidifurax* spp. was recovered disproportionately from house flies collection-wide on Florida dairies (new analysis of data from Greene et al. 1989: \( \chi^2 = 63.03, df = 1, P < 0.001 \)) and on Nebraska cattle confinements in both years studied (new analysis of data from Seymour and Campbell 1993: 1983: \( \chi^2 = 327.17, df = 1, P < 0.001 \); 1984: \( \chi^2 = 458.63, df = 1, P < 0.001 \)). In an Indiana poultry house, *M. raptor* was from house flies more frequently than expected by chance and stable flies less frequently (King 1990).

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LITERATURE CITED


