Overwintering of *Stethorus punctum punctum* (Coleoptera: Coccinellidae) in Apple Orchard Ground Cover

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ABSTRACT Overwintering of adult *Stethorus punctum punctum* (LeConte) was investigated in apple orchards in south-central Pennsylvania. A separation technique was developed to remove adults from soil samples. Density of overwintering adults in the autumn was greatest in the area around the trunks of the apple trees, but because of the relatively small area of the ground cover in the trunk zone, more adults were located in the remainder of the herbicide strip and in the border between the herbicide strip and drive row. In the spring, however, >70% of the adults were located in the trunk zone. Adults were associated with fallen leaves and apple root suckers. Density of adults in orchard ground cover peaked in midautumn and declined to relatively low levels in the spring. Methods to conserve this predator while managing tufted apple bud moth, *Platynota idaeusalis* (Walker), are discussed.

KEY WORDS *Stethorus punctum punctum*, overwintering, apple, Coccinellidae, ground cover

LADYBIRD BEETLES IN the genus *Stethorus* are obligate predators of tetranychid mites contributing to mite management in tree fruit in various parts of the world (Kapur 1948). Conservation of *S. punctum punctum* (LeConte) for control of the European red mite, *Panonychus ulmi* (Koch), and the twospotted spider mite, *Tetranychus urticae* Koch, is the cornerstone of the apple-integrated pest-management program in Pennsylvania and surrounding areas of the mid-Atlantic region (Asquith and Hull 1979). This program currently involves selectively timed applications of low rates of organophosphate and carbamate insecticides, acaricides, and fungicides to control other major insect and disease pests (Anonymous 1996).

The major direct pest, tufted apple bud moth, *Platynota idaeusalis* (Walker), has developed resistance to the organophosphate insecticide azinphosmethyl with resulting control failures (Knight and Hull 1992). Because the carbamate/organophosphate insecticide mixtures used to control tufted apple bud moth threaten the successful IPM program, several alternative control tactics for tufted apple bud moth are being investigated. One tactic involves applying an insecticide to overwintering larvae of tufted apple bud moth in the orchard ground cover in the spring (Bode 1975, Knight and Hull 1988).

To be successful, management tactics directed to the ground cover must control tufted apple bud moth without causing excessive mortality of *S. p. punctum*, which also overwinters in the orchard ground cover. This predator belongs to a group of coccinellids that do not move to a new habitat for hibernation, but overwinter in the orchard and surrounding refugia singly or in small groups under tree bark, litter, or in the upper layers of the soil (Hodek 1973). However, little is known on the distribution of *S. p. punctum* in the various overwintering habitats in the orchard ground cover and surrounding refugia. Colburn and Asquith (1971) sampled overwintered *S. p. punctum* in apple orchards in May, but sampled only fallen leaves within 1.5-m apple trunks. They found the adult distribution skewed toward the trunks where leaves were present. The purpose of this experiment was to determine the spatial and temporal distribution of *S. p. punctum* adults in all available ground cover habitats in apple orchards and surrounding refugia so that management tactics developed for tufted apple bud moth larvae in the ground cover will not interfere with the mite predator.

Materials and Methods

Sampling Procedure. A technique was developed to separate overwintering *S. p. punctum* adults from soil samples. Samples of organic matter consisting of the O1 layer of loose leaves and undecomposed organic debris and the O2 layer of matted and decomposed organic material (Foth 1978) from an area of 0.1 m^2 within 1 m from the trunk of an apple tree were taken under each of 8 trees in February 1991. The organic matter was sifted through a 6-mm screen. The fine (<6 mm) and course fractions (>6 mm) were each placed in clear plastic containers (34 by 24 by 8 cm) (Freezette food saver, Max Klein, Baraboo, WI) with 2

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screened ventilation openings in the lid, and held in the laboratory with natural lighting. Adults were removed each morning for 13 d from resting places on the inside of the separator box lids. The samples were visually searched for remaining adults on day 13.

A further separation efficacy procedure was conducted in December 1992. *S. p. punctum* adults were separated from orchard soil samples as described above. Eighteen adults removed from samples after 1 or 2 d were buried in a sample of sifted soil with no *S. p. punctum*. Efficacy was rated as the number of adults collected again over the following 10 d.

Two samples of soil strata were taken 7 and 9 March 1991 from the herbicide strip of an apple orchard to determine the location of *S. p. punctum* overwintering sites. Samples were taken using a 0.1-m^2 frame within 1 m of the trunk at sites with known high adult densities. The 1st fraction removed was primarily leaf litter (O1 and O2). Next, a whisk broom was used to collect the remaining organic matter (O2) from the soil surface. Finally, 0–1-cm and 1–2-cm soil fractions (A_p, plow layer) were dug with a spade. Samples were brought to laboratory and sifted through 6-mm screen. The material >6 mm was discarded and the sifted fractions were placed in separator boxes. Adults were removed daily for 10 d.

Orchard Survey. Distribution of *S. p. punctum* adults was determined in the ground cover of four representative 'Yorking' apple orchards in Adams County, Pennsylvania, between autumn 1991 and summer 1993. The trees were 20–30 yr old and planted on standard or semidwarfing rootstocks at 190–303 trees per hectare. The understory of each orchard was stratified into 4 zones from which samples were taken (Felland et al. 1996). The trunk zone included a 0.6-m radius out from the trunk, the remainder of the herbicide strip extending from 1.5–2.0 m from the trunk, and the border zone included the 1.0- to 1.5-m strip outside the herbicide strip yet within the tree canopy. The drive row included all the area between the canopy edge of adjoining rows. The mean (±SEM) percent area in the 4 zones in the study orchards was 4.5 (0.6), 34.8 (8.8), 23.5 (7.2), and 37.2 (6.8), respectively.

Sampling in each of the 4 orchards was stratified into 4 compass quadrants. One tree was selected randomly, without replacement, from each quadrant on each sample date. The ground cover associated with the sample trees was evenly divided into 4, 10, 8, and 16 locations for the trunk, herbicide strip, border, and drive row zones, respectively, based on the different areas of the zones. Two samples were assigned randomly in each of the 4 zones per tree. Thus, a total of 128 ground cover samples was made per date. The orchards were sampled on 21–25 October, 5–8 November, and 6–10 December 1991; 15–17 April, 22 May–1 June, 30 July–3 August, 18–25 September, 26–30 October, and 2–9 December 1992; and 20–29 April, 24 May–2 June and 13–16 July 1993. Sampling was somewhat impaired by frozen soil during a portion of the December 1991 sampling period.

Ground cover refugia immediately surrounding the orchard sites were sampled on each date except July 1992. These sites included the trunk and herbicide strip zones of a peach orchard (1 site), brushy areas (2 sites), up to 2 m into hardwood stands (3 sites), and a soybean field (April–May at 1 site) in which fallen apple leaves had collected. Because the refugia did not completely surround the orchards, refugia sampling was not associated with each orchard quadrant. Rather, refugia were evenly partitioned and a total of 6–20 refugia samples were collected during each sample date.

The samples consisted of all the vegetation, soil organic matter including fallen leaves, and soil to a depth of 1 cm from a 0.05- to 0.1-m^2 area. Sample size was consistent on a given sample date. The vegetation, intact fallen leaves, and dropped apples were examined for *S. p. punctum* adults. The remaining organic matter and soil were sifted through 6 mm hardware cloth and the adults were removed with the above separation procedure. On most sampling dates the intact fallen leaves of the O1 layer were examined *in situ* to determine location of adults within the organic matter and the remaining O2 and Ap sample was placed in a plastic bag and sifted in the laboratory. In the December 1991 and April 1992 samples the entire sample was bagged and examined in the laboratory without the location of adults being differentiated as to soil layer. In the October samples both years the number of adults per infested leaf was recorded.

The percentage of the ground cover with fallen leaves, apple root suckers, broadleaf weeds, and grasses was visually estimated for each sample. The percentages were based on the amount of soil surface covered with the particular host. Host biomass was not measured, except in the case of fallen leaves, which were dried to a constant weight and weighed.

Density of *S. p. punctum* adults was calculated in three ways. First, the number per square meter was determined for each ground cover zone. Second, because the 4 zones were not uniform in size, the same data were converted to percentage per zone. This percentage was calculated by using the actual areas of each zone on an orchard-by-orchard basis. And 3rd, the number per square meter for the orchard as a whole was estimated. Density in the refugia was calculated only on a per-square-meter basis because sampling was not extensive enough to quantify the absolute density in the refugia relative to the orchard.

Analyses were conducted independently for the autumn, spring, and summer collections. The 3 autumn samples each year were pooled and only the April sample was included in the spring samples because no adults were recovered in May. The analyses were split plot with zone as whole plot,

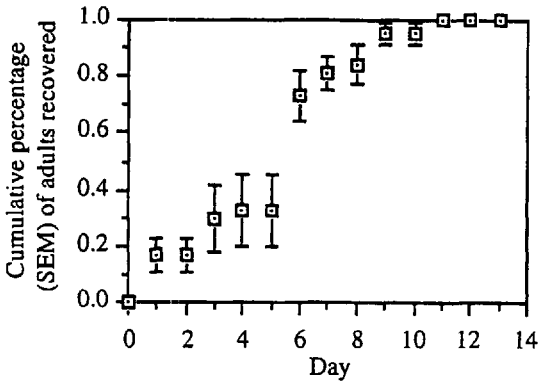

Fig. 1. Cumulative recovery in separator boxes of a total of 65 *S. p. punctum* adults from 8 samples of fallen apple leaves, February 1991, Biglerville, PA.

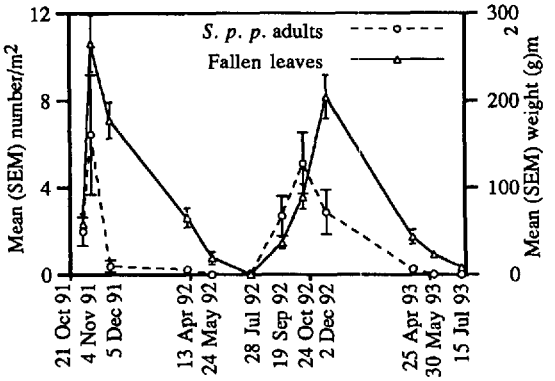

Fig. 2. Density of *S. p. punctum* adults and weight of fallen leaves in apple orchard ground cover, Adams County, Pennsylvania, 1991–1993.

orchard as replication, and orchard quadrant as split plot. The effect of orchard zone on density of adults and total adults was tested using analysis of variance (ANOVA) and means separated by the Fisher protected least significant difference (LSD) (Abacus Concepts 1988). Linear regression was used to determine the relationship of the variables fallen leaves, fallen leaves × percentage of apple root suckers, percentages of apple root suckers, broadleaf weeds, and grasses, and the density of the adults during the 3 seasons. The interaction between fallen leaves and percentage of apple root suckers was included to test whether the presence of fallen leaves among the root suckers was associated with increased overwintering populations of *S. p. punctum*.

Results and Discussion

Sampling Procedure. Ninety-seven percent of the live *S. p. punctum* adults were recovered in the fine fraction; 3% remained in the course fraction, suggesting that sifting was adequate to concentrate the adults in the fine sample from which they were removed. Recovery per day was ≈10% (Fig. 1). No live adults were recovered after 11 days. An additional 12% dead adults were recovered in a visual search on day 13. Efficacy of the sampling procedure was 94.4% in December 1992.

Thirty-five adults were recovered from the 2 samples taken in March 1991. Mean (±SEM) percent recovery was 88.3 (11.7), 8.3 (8.3), and 3.3 (3.3) from the sifted organic layer, sweepings from the soil surface, and the 1st cm of the Ap, respectively. No adults were found in the 1- to 2-cm sample of the Ap. Therefore, subsequent sampling was limited to the organic matter and the top 1 cm of soil.

Orchard Survey. In total, 441 adults were recovered in the samples taken during 1991–1993. In October and November 1991, 6.3 ± 2.7% (mean ± SEM) of the adults were recovered in the intact leaves of the O1 layer with the remainder recovered in the sifted organic matter. In September, October, and December 1992, 38.3 ± 1.9% were recovered in the O1 layer. One adult in September 1992 was recovered on a fallen apple, where it was probably feeding on overwintering European red mite eggs. In April 1993, 7.1% of the adults were recovered in the O1 layer. No adults were located during May. Only 2 adults were recovered in the July samples, both in a single fallen leaf. Adults were found in 30 leaves in the October samples both years. Of these, 8 had 2 adults each and 22 had a single adult each.

Density of *S. p. punctum* adults in the ground cover peaked in early November 1991 and in late October 1992 (Fig. 2). The sharp decline in density in December 1991 was not observed in December 1992 and may have resulted from reduced recovery from frozen leaf samples. Density was low in April and no adults were collected from ground cover samples in May. The majority of leaf-fall coincided with increasing densities of *S. p. punctum* in the ground cover in the autumn.

This study suggests that up to ⅓ of the population of adult *S. p. punctum* moved to overwintering sites as leaf-fall was beginning, and some possibly as early as July. Hodek (1973) noted that the coccinellids that did not change their habitat for hibernation are active well into the autumn as was observed in this study. Colburn and Asquith (1971) observed adults in orchards until late October. However, Putman (1955) found that females of *S. punctillum* began entering diapause from July onward. He did not determine when adults entered overwintering sites, but presumed that adults began seeking overwintering sites in October as leaf-fall progressed.

Adult density of *S. p. punctum* was highest around the trunks both in the pooled autumn samples and in April (Table 1). Adults were less common in the herbicide strip and border and were only infrequently located in the drive row at this time. Density in the trunk zone had less of a decline than the other zones between the autumn

Table 1. Mean ± SEM number of *S. p. punctum* adults per square meter in 4 zones of ground cover of apple orchards and in refugia during 3 seasons, Adams County, Pennsylvania, 1991–1993

Zone	Autumn	Spring	Summer
Trunk	28.27 ± 5.50a	3.64 ± 1.65a	0.00 ± 0.00a
Herbicide strip	4.13 ± 0.94b	0.08 ± 0.08b	0.00 ± 0.00a
Border	2.81 ± 0.84cd	0.16 ± 0.11b	0.06 ± 0.06a
Drive row	1.25 ± 0.48d	0.00 ± 0.00b	0.00 ± 0.00a
Refugia	1.00 ± 0.52	0.83 ± 0.47	0.00 ± 0.00

Means within columns followed by the same letter are not significantly different (Fisher protected LSD, $P > 0.05$).

and the spring samples. Adult density in refugia was not as high as the trunk zone, but tended to remain constant between the autumn and spring samples.

The distribution of adults among the 4 ground cover zones changed between the autumn and spring samples (Table 2). The highest percentage of adults in the autumn were found in the herbicide strip, whereas in the spring most adults were found in the trunk zone. Colburn and Asquith (1971) found 50 and 80%, respectively, of adults within 0.3 and 1.0 m of the trunks in spring samples of leaf litter when the litter was against the tree. Readshaw (1971) also observed *Stethorus* spp. concentrated in leaf litter and herbage around the base of apple trees.

The leaves in the trunk zone may be caught in the root suckers and offer a more stable overwintering site for the adults than in the other zones. The leaves in the other orchard zones may be raked into the drive rows during the pruning operation. Alternatively, some active movement of adults toward the trunks may occur during warm periods in the early spring. Asquith and Hull (1979) reported adults feeding on mites and mite eggs on the trunks during warm sunny days before moving into the trees.

Multiple regressions of variables of ground cover habitat within sample area and density of *S. p. punctum* adults had low coefficient correlations in most instances indicating little multicolinearity between variables. High densities of adults were positively correlated with presence of fallen leaves × apple root suckers and with fallen leaves alone in the autumn (Table 3). Although apple root suckers had a high beta value, this variable was not significant ($P > 0.05$). In the spring, when most of the suckers had been pruned, only the presence of fallen leaves was significant.

Table 2. Percentage (± SEM) per zone of adult *S. p. punctum* in the apple orchard ground cover during 3 seasons, Adams County, Pennsylvania, 1991–1993

Zone	Autumn	Spring	Summer[a]
Trunk	15.0 ± 4.7b	71.0 ± 18.6a	0.0
Herbicide strip	58.7 ± 11.9a	9.8 ± 9.8b	0.0
Border	20.8 ± 6.7b	19.3 ± 19.3b	100.0
Drive row	5.2 ± 4.3b	0.0 ± 0.0b	0.0

Means within columns followed by the same letter are not significantly different (Fisher protected LSD, $P > 0.05$). Values for individual orchards were calculated by multiplying density in each zone by relative area of each zone, dividing the total per zone by the total per all zones, and multiplying by 100%.

[a] Adults located in 1 orchard only.

Low density of adults in the April samples both years suggests relatively high overwintering mortality. Emergence from the overwintering sites was <15% when these samples were taken (Felland et al. 1995). In contrast to the orchard, density remained relatively high in the refugia, which tended to have a thicker layer of organic matter. Putman (1955) caged adults of *S. punctillum*, which overwinter in similar habitats as *S. p. punctum*, on dead leaves, soil, and sod and reported only 20.0–27.5% overwintering survival and found no survival of adults overwintering on an apple limb. Little snow cover occurred to modify temperature in the overwinterng sites. During the winter of 1991–1992 snow cover averaged 4.7 cm over a total of 13 d. In the winter of 1992–1993 snow cover averaged 1.7 cm for 3 d until 13 February, after which an average of 19.3 cm remained through 27 March.

The cues used by adults for entering overwintering sites are not well understood. Adults may move to the base of the trees and remain in a favorable habitat of fallen leaves trapped among the apple root suckers. The highest density of adults for the area of the ground cover under an entire tree was 36.3/m^2. The maximum number in a single 0.1-m^2 sample was 31 adults in the trunk zone. Adults have been found aggregated with up to 12 in a single fallen leaf (C.M.F., unpublished data). Whether this aggregation is a result of movement toward optimum habitat or of an aggregation pheromone is not known.

Little overlap occurs in the temporal and spatial distribution during the spring of overwintering adult *S. p. punctum* with that of larval tufted apple bud moth. The coccinellids leave overwintering sites several weeks before the tufted apple bud moth enters the pupal stage allowing a window of opportunity to manage the pest without disrupting the predator (Felland et al. 1995). Spatially, the coccinellids are concentrated around the trunks, whereas the tufted apple bud moth larvae tend to be most common in the herbicide strip and the border (Felland et al. 1996). Thus, habitat left undisturbed to increase overwintering of the coccinellid (e.g., leaves collected among root suckers) may not greatly increase the density of tufted apple

Table 3. Model coefficient table of ground cover habitat variables and density of adult *S. p. punctum*, Adams County, Pennsylvania, 1991–1993

Variable	Autumn		Spring		Summer	
	$\beta \pm$ SEM	*P*	$\beta \pm$ SEM	*P*	$\beta \pm$ SEM	*P*
Fallen leaves (g/m^2)	0.016 ± 0.007	0.0153	0.049 ± 0.006	0.0001	−0.001 ± 0.005	0.8867
Fallen leaves (g/m^2) × % apple root suckers	0.003 ± 0.001	0.0001	0.000 ± 0.001	0.6315	0.000 ± 0.000	0.8259
% apple root suckers	0.037 ± 0.020	0.0611	−0.001 ± 0.004	0.8191	−0.000 ± 0.001	0.7207
% broadleaf weeds	−0.001 ± 0.003	0.7604	−0.002 ± 0.001	0.1386	−0.000 ± 0.001	0.5666
% grasses	−0.004 ± 0.003	0.1461	0.000 ± 0.001	0.9862	−0.000 ± 0.001	0.9160
Intercept	0.372 ± 0.164	0.0242	−0.047 ± 0.062	0.4500	0.013 ± 0.012	0.2675

bud moth. Hibernal shelters have been used to conserve predators in other crops (Sengonca and Henze 1992).

Experimental studies should be conducted in which potential overwintering sites in the ground cover are manipulated. Pruning of root suckers and raking of leaves at different seasons could be investigated. The impact of rootstocks with little root sucker growth, which are preferred by growers, on *S. punctum punctum* overwintering should also be examined.

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