Inhibition of Movement of Larvae of the Convergent Lady Beetle[1] by Leaf Trichomes of Tobacco[2]

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ABSTRACT

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Movement of all instars of *Hippodamia convergens* Guérin-Méneville was affected by the sticky exudates of leaf trichomes of tobacco cultivars. The younger the instar and the longer the larvae were on these leaves, the greater was the inhibition of movement. The alkaloid content of the plant had no effect on movement.

The green peach aphid, *Myzus persicae* Sulzer, became a serious pest of tobacco in the United States in 1946 and 1947 (Chamberlain 1958). It is capable of transmitting several virus pathogens to tobacco, including tobacco etch and tobacco vein mottling virus. In addition, the aphid can stunt and wither leaves of older plants and can harm tobacco plants by injecting toxic salivary secretions (Lawson et al. 1954). The honeydew produced by this aphid promotes growth of black molds and causes the leaves to stick together, both of which may hamper proper curing.

Predaceous coccinellid beetles feed on the green peach aphid and are commonly found on aphid-infested tobacco in Kentucky. Katanyukul (1973) found the most common species on tobacco in Fayette County, Ky., to be the convergent lady beetle, *Hippodamia convergens* Guérin-Méneville. Because the larvae of Coccinellidae do not use sight when locating prey, their ability to move on leaf surfaces is very important (Banks 1957, Dixon 1958, Fleschner 1950, Putman 1955). These larvae, although efficient predators in late instars, detect prey only by contact while randomly searching the plant (Dixon 1958).

Rabb and Bradley (1968) noted that exudates of tobacco trichomes limited the movement of *Trichogramma minutum* Riley on plant surfaces. Katanyukul's (1973) research supported their results. Elsey (1974) found that the searching speed of larvae of the lady beetle, *Coleomegilla maculata* (De Geer), and of the common green lacewing, *Chrysopa carnea* Stephens, was faster on cotton than on tobacco. He concluded that the large number of trichomes on tobacco seriously reduced the speed of movement of these insects compared with their movement on the leaves of cotton, which have few and nonglandular trichomes.

The purpose of this research was to determine the effect of characteristics of various leaf surfaces of four tobacco cultivars on the movement of the four instars of the convergent lady beetle.

Materials and Methods

Two U.S. tobacco cultivars, Burley 21 (By 21) and Low Alkaloid Burley 21 (LA by 21), and two foreign tobacco cultivars, Tobacco Introduction (TI)1068 and TI 1298, were transplanted on 13 June and 1 July 1979 at a University of Kentucky research farm in Lexington. At each time of transplanting 12 replicates of the four cultivars were planted in a randomized complete block design with each replicate consisting of a single plant of each cultivar. The plants were ca. 1.67 m apart in the row, and the rows were ca. 2 m apart. Tests were not run until the plants reached a height of ca. 1 m. When the plants from the first transplanting began flowering, plants from the second transplanting were used in subsequent tests.

The major emphasis was a comparison of the movement of each of the four coccinellid instars on the undersides of leaves from the middle part of By 21, LA By 21, TI 1068, and TI 1298 plants. Thirty leaves of each of the four cultivars were used for each of the four instars, making a total of 480 individual tests. The four cultivars in each replicate were tested, and then the four cultivars in the next replicate were tested. More than one leaf of plants from some of the 24 plants were necessarily used more than once to make the 30 tests for each instar. In tests on the movement of 1st instars on the undersurfaces of leaves from the top and bottom part of By 21, TI 1068, and TI 1298 plants, 15 tests were run, using one replicate for each test for each leaf position of each of the three cultivars. These results were compared with results from the 30 tests with 1st instars on the undersurface of middle leaves.

A leaf was cut from the plant to be tested and placed top-side down on a shaded table on the edge of the field containing the tobacco plots. All aphids on the leaf were removed manually with forceps to allow uninterrupted searching by the larva. The table was shaded to provide light conditions similar to those of the underside leaf surface when on the plant and to minimize any effect of sunshine on movement of the larva. The tests were run during the middle of the day to minimize the effect of any changes in temperature on succeeding tests.

A sheet of hard, transparent plastic (12 by 8 cm) supported by six 25-mm-long legs was placed on the table over the leaf, and the specified instar of convergent lady beetle was placed on the leaf. The movement of the larva was traced on the plastic sheet with a wax pencil. Movement was recorded for 30

[1] Coleoptera: Coccinellidae.

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sec immediately after the larva was placed on the leaf; the larva was left on the leaf, and the movement during 30 sec was again recorded at 10-, 20-, and 30-min intervals thereafter. The movement recorded on the plastic at each time interval was traced onto a piece of paper, and the plastic was cleaned before the next 30-sec measurement was made. The distance moved at each time interval was measured from the paper in the laboratory at the end of the test period. The measurement technique used was similar to that used by Rowlands and Chapin (1978).

For tests using 1st instars, egg masses collected on tobacco plants in the field were placed in cells of a tissue culture plate in the laboratory until they hatched. These larvae were less than 1 day old when used. When 2nd, 3rd, and 4th instars were used, they were collected from tobacco plants in the field, isolated in plastic dishes in the laboratory, and starved for 24 h before use so that they would search actively.

The four cultivars were chosen because of the differences in their leaf characteristics, especially the differences in their leaf trichomes and the trichome exudates. TI 1068 is very sticky to the touch and is much stickier than standard U.S. cultivars such as By 21; LA By 21 is similar to By 21 in this characteristic, whereas TI 1298 is not sticky. Large amounts of sticky exudates accumulate on the hands when LA By 21, By 21, and TI 1068 are handled. These observations are borne out by counts of the trichomes present on the leaf. LA By 21 and By 21 have as many trichomes as TI 1068 but substantially fewer of the glandular trichomes, which produce the exudates, whereas TI 1298 has significantly fewer glandular, as well as total, trichomes than By 21 or LA By 21 (Roberts et al. 1981). By 21 is grown commercially in Kentucky and has the usual alkaloid content present in other commercially grown burley cultivars, whereas LA By 21, which was developed from By 21, has been bred for very low alkaloid content (Legg et al. 1970).

Results and Discussion

The distance moved by the larvae varied with the instar and with the cultivar used (Table 1). The older the instar, the greater the distances they moved when on leaves of the same cultivar. These differences were especially marked for 1st and 2nd instars; 2nd instars moved more than twice as far as 1st instars on each of the cultivars, with the greatest differences occurring when on TI 1068. Each instar moved farthest on TI 1298, least on TI 1068, and intermediate distances on both By 21 and LA By 21. The longer the larvae stayed on TI 1068, the shorter the distance moved. After being on leaves of this cultivar for 30 min, the distance moved in 30 sec was ca. one-tenth that moved in 30 sec immediately after being placed on the leaf. There were reductions in movement after being on leaves of By 21 and LA By 21 for 30 min, but the differences were not nearly so great as those on TI 1068. There was no reduction in movement when on leaves of TI 1298 after being on the leaves for 30 min.

These observations are compatible with the concept that the trichome exudates are responsible for inhibition of movement. Elsey (1974) found that 1st and 2nd instars of another coccinellid, *Coleomegilla maculata* (De Geer), moved farther on the leaves of cotton which had nonglandular and few trichomes than on the leaves of a flue-cured tobacco cultivar. He found that searching speed was negatively correlated with the number of trichomes on the tobacco leaves, but he did not separate glandular from nonglandular trichomes. His observations by microscope of the interference of the trichomes with movement of the larvae were substantiated by our observations. The larvae, especially the 1st instars on TI 1068, were

Table 1.—Average distances moved (mm) in 30 sec by instars of the convergent lady beetle after being on the undersurface of leaves from the middle part of four tobacco cultivars for various periods of time[a]

Instar	Cultivar	Min after being placed on leaf[b]			
		0	10	20	30
1st	TI 1298	18.4a	18.2a	13.1a	14.7a
	By 21	9.4b	7.2b	4.7b	3.8b
	LA By 21	8.2b	6.0bc	5.2b	4.5b
	TI 1068	3.9c	2.4c	0.8c	0.3c
2nd	TI 1298	60.4a	59.3a	58.8a	62.7a
	By 21	19.5bc	14.4b	15.2b	12.3b
	LA By 21	24.7b	15.5b	12.9b	11.0b
	TI 1068	15.2c	3.2c	2.3c	1.9c
3rd	TI 1298	107.0a	107.5a	103.8a	104.3a
	By 21	49.4b	38.8b	36.4b	39.6b
	LA By 21	37.6b	21.4c	17.5c	19.2c
	TI 1068	18.5c	5.3d	3.4d	1.8d
4th	TI 1298	166.8a	169.9a	160.3a	166.7a
	By 21	59.1b	41.6b	41.4b	38.0b
	LA By 21	49.2bc	29.8b	24.7c	25.3b
	TI 1068	35.2c	15.4c	7.2d	3.5c

[a] $n = 30$.
[b] Means followed by the same letter are not significantly different at the 0.05 level by Duncan's new multiple range test.

Table 2.—Average distances (mm) traveled in 30 sec by 1st-instar *H. convergens* after being on the undersurfaces of leaves from the top and bottom stalk positions of three tobacco cultivars for various periods of time[a]

Leaves	Cultivar	Min after being placed on leaf[b]			
		0	10	20	30
Top	TI 1298	15.3a	17.1a	16.9a	16.3a
	By 21	3.3b	2.1b	1.9b	1.9b
	TI 1068	1.9b	0.9b	0.3b	0.2b
Bottom	TI 1298	10.1a	10.3a	12.2a	11.5a
	By 21	9.7a	8.3a	6.9b	4.9b
	TI 1068	3.3b	1.7b	0.8c	0.9b

[a] $n = 15$.
[b] Means followed by the same letter are not significantly different at the 0.05 level by Duncan's new multiple range test.

observed to be stuck to the trichome exudates, and they had to struggle to disengage their legs and the posterior end of the abdomen from the exudates. The legs often became stuck to the body, and the larvae then fell over to one side and had difficulty in righting themselves. This is similar to the reaction of larvae of the Colorado potato beetle, *Leptinotarsa decemlineata* (Say), which come in contact with the sticky exudates of trichomes of *Solanum polyadenium* (Gibson 1976).

Because the total number of trichomes on TI 1068 is similar to that of By 21 and LA By 21 but the number of glandular trichomes is significantly greater, it appears that the larger amount of exudate is responsible for the greater inhibition of movement on this cultivar. This effect on the movement of insects is also borne out by laboratory tests which showed that removing at least part of the exudates by washing the leaves of tobacco cultivars with water plus detergent significantly increased the parasitism of eggs of the tobacco hornworm, *Manduca sexta* (Johannson), by *Telenomus sphingis* (Ashmead) (W. Katanyukul and R. Thurston, unpublished data). The exudates of the glandular trichomes of tobacco are initially enclosed between the cell wall and an external cuticle, but as they accumulate they rupture the cuticle, whereupon they appear externally as blobs covering the heads of these trichomes (Bentley and Wolf 1945). When touched, strands of the sticky material can be pulled away from these blobs.

The amount of alkaloid in the plant apparently had little or no effect on the movement of the larvae. LA By 21 contains negligible amounts of alkaloid, whereas By 21 contains large amounts (Legg et al. 1970), but there were not consistent differences between the movement of larvae on these cultivars. In some instances, the larvae move farther on By 21 than on LA By 21, which is contrary to what would be expected if the alkaloid inhibited movement.

Larvae on the undersurface of leaves from the top part of By 21 and TI 1068 plants moved much shorter distances than when on the lower surface of leaves from the middle or bottom part of these plants (Table 2). This was expected because of the greater number of trichomes per unit of leaf area on the young, top leaves; the number of trichomes apparently does not increase as the leaf expands from a length of ca. 15 to 20 cm of the young top leaves to the ca. 50 to 60 cm of the middle and bottom leaves (Bentley and Wolf 1945). Surprisingly, the larvae on the bottom leaves of TI 1298 did not move farther than larvae on the top leaves of this plant, but even on the bottom leaves the 1st instars moved farther on TI 1298 than on TI 1068 or By 21.

The following conclusions were drawn from this study. (1) Larvae of the convergent lady beetle moved farthest on the leaves of TI 1298, intermediate distances on By 21 and LA By 21, and the shortest distances on leaves of TI 1068. The inhibition of movement is correlated negatively with the number of glandular trichomes which produce sticky exudates. (2) The longer the larvae stayed on the sticky leaves of By 21, LA By 21, and TI 1068 the greater was the inhibition of movement, but this was not so for the nonsticky leaves of TI 1298. (3) The older the instar the greater was the distance moved on leaves of all tobacco cultivars. (4) Movement on leaves from the upper part of plants of By 21 and TI 1068 was less than on leaves from the middle or bottom part of these plants, but the reverse was true for TI 1298. (5) Movement on LA By 21, a low-alkaloid cultivar, was similar to that on By 21 which contains much larger amounts of alkaloids.

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