Effect of Silver Reflective Mulch and a Summer Squash Trap Crop on Densities of Immature *Bemisia argentifolii* (Homoptera: Aleyrodidae) on Organic Bean

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ABSTRACT Polyethylene mulch with a reflective silver stripe and a yellow summer squash, *Cucurbita pepo* L., trap crop were tested alone and in combination as tactics to reduce densities of *Bemisia argentifolii* Bellows & Perring eggs and nymphs, and incidence of bean golden mosaic geminivirus on snap bean, *Phaseolus vulgaris* L. Egg densities were consistently higher on squash than on bean, but egg densities and virus incidence were not lower on bean grown with squash than on bean grown in monoculture. Silver reflective mulch reduced egg densities compared with bean grown on bare ground during the first week after crop emergence for 2 of the 3 yr that the study was conducted. However, egg suppression by silver mulch was not enhanced by the presence of a squash trap crop when both tactics were combined. The obstacles to suppressing *B. argentifolii* through the use of trap crops are discussed.

KEY WORDS pest management, intercropping, polyculture, cultural control, geminivirus, vector management

Bemisia argentifolii BELLOWS & PERRING, also known as the B strain of *Bemisia tabaci* (Gennadius), has become a serious pest of horticultural and agronomic crops throughout warm regions of the world (Brown et al. 1995). In addition to causing mechanical damage, *B. argentifolii* has been associated with several new plant disorders, and dozens of new geminiviruses (Polston and Anderson 1997). In 1993, bean golden mosaic virus, vectored by *B. argentifolii*, reached epidemic proportions for the first time in Florida (Blair et al. 1995), the foremost producer of snap bean (*Phaseolus vulgaris* L.) in the United States (National Agricultural Statistics Service 1998). *Bemisia* has developed resistance to most classes of pesticides (Dittrich et al. 1990, Denholm et al. 1996), forcing conventional growers to seek nonchemical alternatives for *Bemisia* management. The systemic insecticide imidacloprid (Bayer) is currently effective for controlling *B. argentifolii* (Polston et al. 1994), but is not an option for organic growers, who face special challenges in the management of virus vectors.

Reflective plastic mulches contribute to whitefly management programs by providing early-season protection from whitefly-vectored viruses (Suwwan et al. 1988, Kring et al. 1990, Csizinsky et al. 1999). Whitefly adults are apparently repelled by UV wavelengths reflected by silver and aluminum pigments on the plastic mulch (Stansly and Schuster 1999). However, reflective mulches tend to lose their efficacy as shade from the crop canopy increases (Csizinsky et al. 1997). Reflective mulches alone do not provide sufficient protection from whitefly damage (Schuster et al. 1989), and so have been tested in combination with other tactics such as pesticides (Powell and Stofella 1993, Natwick and Mayberry 1994).

Bemisia argentifolii has demonstrated differential rates of oviposition on various hosts (Blua et al. 1995, Chu et al. 1995, Tsai and Wang 1996). Certain hosts have been tested as trap crops for management of *B. argentifolii* because of these apparent crop preferences. The results of trap cropping with soybean (*Glycine max* L., McAuslane et al. 1995), melon (*Cucumis melo* L., Perring et al. 1995), and Wright's ground cherry (*Physalis wrightii* Gray, Ellsworth et al. 1994) have been inconclusive. However, Schuster et al. (1996) delayed the onset of *Bemisia*-transmitted virus by intercropping tomato, *Lycopersicon esculentum* Mill., with squash, *Cucurbita pepo* L. In addition, Al-Musa (1982) reduced levels of *B. tabaci* nymphs and the onset of tomato yellow leaf curl symptoms by intercropping tomato with cucumber, *Cucumis sativus* L.

The current study was conducted as part of an effort to find methods for managing *B. argentifolii* that are suitable for organic growers. Silver reflective mulch and a squash trap crop were tested alone and in combination to determine their effect on virus incidence, yield, and densities of immature *B. argentifolii* on snap bean compared with bean grown in monoculture on bare ground. We hypothesized that squash might draw

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females of *B. argentifolii* away from the bean, and so offer a less expensive method than reflective mulch for suppressing whitefly densities and virus incidence. Furthermore, we wished to determine if squash and reflective mulch used in combination could suppress whitefly densities to levels below those measured with either method alone.

Materials and Methods

The study was carried out on a 4-ha certified organic farm, 6 km northwest of Gainesville, FL (29° 40′ N, 82° 30′ W). Four treatments were compared: (1) bean grown on bare soil (bean), (2) bean grown with reflective polyethylene mulch (mulch), (3) bean mixed with squash on bare soil (squash), and (4) bean mixed with squash and grown with reflective mulch (squash/mulch). 'Espada' garden bean seed and 'Multipik' yellow summer squash seed from Harris Seed (Rochester, NY) were used. Seeds had been treated previously with captan, metalaxyl, streptomycin, and chloroneb. It is acceptable for organic growers to use treated seeds if nontreated seeds are unavailable (Organic Materials Review Institute 1998).

To ensure uniformity among covered and exposed beds, all beds were formed using a Rainflo plastic mulch layer (model no. 560, Rainflo Irrigation, East Earl, PA). Plastic mulch and drip irrigation tubing were laid over all beds, which were 1.22 m wide. After planting, plastic mulch was removed from the bare-soil treatments.

Beans were planted 15 cm apart within the row. Squash replaced every fifth bean plant in the squash treatments. Beds were 3.5 m long, and the space between beds was 2 m. Each treatment plot contained two beds with two rows of plants per bed. The reflective mulch was a white polyethylene mulch with a central stripe of silver pigment, 61 cm wide (product 60–64S/W125PR, North American Film, Bridgeport, PA).

Treatments were arranged in a 4 × 4 Latin square design. Plots were irrigated as needed using drip irrigation. Plants were fertilized 3 wk after emergence and at flowering with ≈250 g per row of 3–2–3 (N–P_2O_5–K_2O) North Florida Brand composted chicken manure. Plots were hand-weeded as needed. No pest control products were applied to the experimental area. The study was repeated in 1995, 1996, and 1997. In 1995, crops were planted on 15 October. The following years, crops were planted on 2 September.

Sampling. Sampling for whiteflies began 1 wk after crop emergence. Four or five bean and squash plants were sampled per plot each week. The sample unit was a 3.35-cm^2 leaf disc cut from upper and lower leaves by using a cork borer (McAuslane et al. 1995). Discs were taken from the abaxial side of the leaf, in the lower half of the leaf to the right of the midvein. Samples were examined using a dissecting stereoscope (model SMZ-1B, Nikon, Japan) set at 20× and the numbers of whitefly eggs, nymphs, and parasitized nymphs were recorded. Sampling was stopped after 4 wk in 1995 because of a freeze. Beans and squash were sampled for 6 wk in 1996 and 1997. Pods were harvested weekly and fresh weight was recorded.

Virus Screening. Visual observations of the symptoms of viral infection were recorded. After harvest, leaf tissue from six plants from each plot was collected and tested with a dot blot hybridization technique for the presence of geminivirus (Rojas et al. 1993). Analysis was conducted by the laboratory of E. Hiebert, Department of Plant Pathology, University of Florida. Bean tissue (50 mg) was extracted in 200 mM NaOH with 1% SDS. Geminivirus DNA-A component was amplified by polymerase chain reaction (PCR) with Maxwell degenerate primers (PAL1v1978 and PAR1c496). The amplified DNA was used for a ^{32}P random-primed labeling reaction (RTS RadPrime DNA Labeling Systems, Life Technologies, Grand Island, NY). The membrane was hybridized with ^{32}P labeled probe in 6× SSC, 5× Denhardt's solution, and 0.5% SDS at 65°C for 16 h. The membrane was then washed under high stringency conditions with 0.2× SSC and 0.1× SDS at 65°C. Finally, the membrane was exposed to X-ray film for 16 h.

Statistical Analysis. Whitefly counts were transformed by $\log_{10}(x + 1)$ because of low counts during the first year and unequal variance over time. Comparisons of immature counts on bean under the four treatments and between squash and bean in the trap crop treatments were made based on the average of upper and lower leaf disc counts. Treatments were compared by individual week with analysis of variance (ANOVA) (PROC MIXED, SAS Institute 1996). When appropriate, treatment means were compared with the Tukey studentized range mean separation procedure. Yield data were analyzed with the same ANOVA and mean separation procedures. Bean samples that tested positive for the presence of bean golden mosaic virus were assigned a value of one, and negative responses were assigned a value of zero. Responses were then analyzed with logistic regression.

Results

A November freeze killed all crops in 1995 after only 4 wk of sampling. During the next 2 yr the study was initiated during the first week of September to reduce the risk of freezes. Egg densities of *B. argentifolii* were highest during the first weeks of sampling and declined over subsequent weeks. However, egg densities varied from year to year. For example, overall egg densities on bean during the first week of sampling were low in 1995 (1.01 ± 0.85/cm^2 foliage, mean ± SD), slightly higher in 1996 (4.16 ± 3.28/cm^2), and high in 1997 (24.71 ± 19.74/cm^2).

Treatment Comparisons. Egg densities were consistently higher on squash foliage than on bean foliage in the two squash treatments (Fig. 1; $4.48 \leq F \leq 69.54$; df = 1, 3; $P < 0.05$). However, other than during a few weeks in 1995 (Fig. 2a), egg densities were not lower on bean intercropped with squash than on bean grown in monoculture, with or without reflective mulch (Fig. 2 b and c). Egg densities were significantly lower on bean in the mulch and squash/mulch treatments than

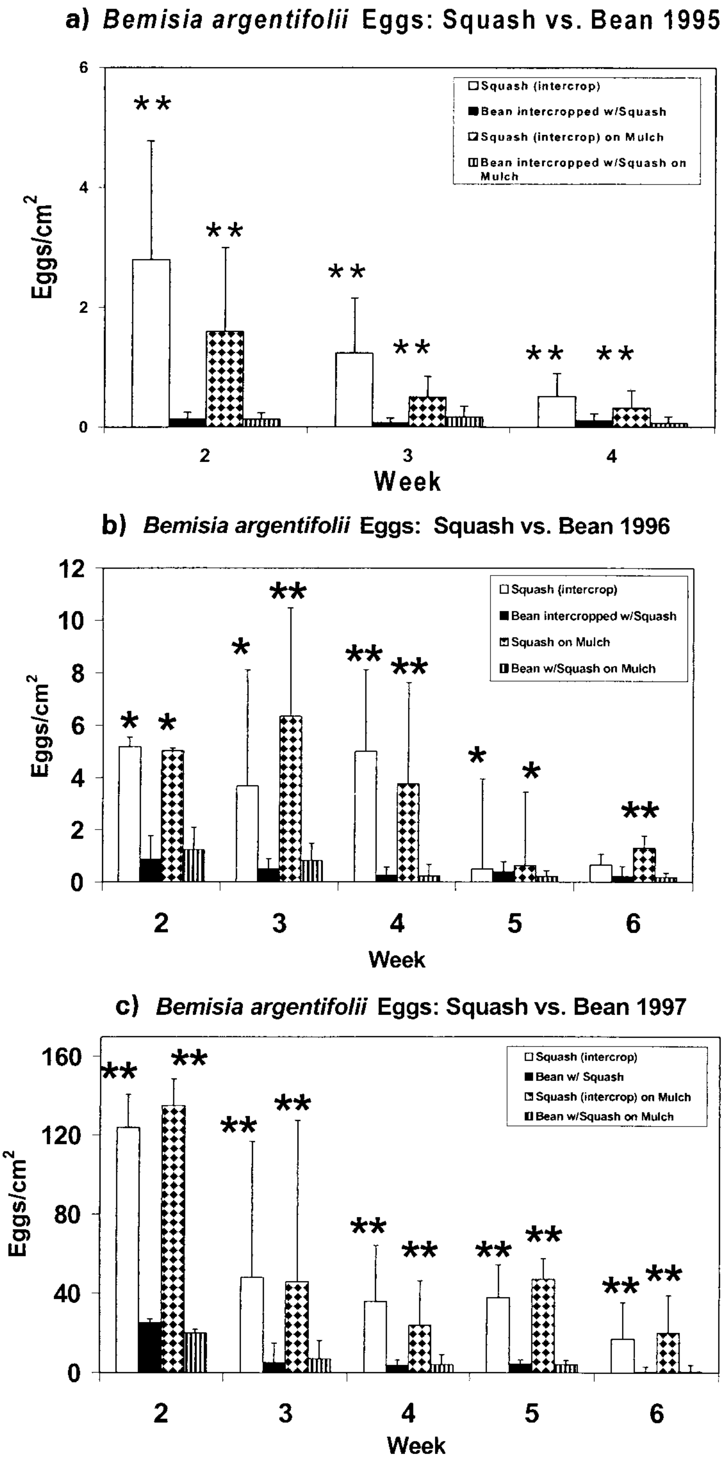

Fig. 1. Densities of *B. argentifolii* eggs on squash and bean intercropped on bare ground, and squash and bean intercropped with reflective mulch, for 1995, 1996, and 1997 seasons. Asterisks indicate that egg densities on squash foliage are significantly higher than densities on bean in the same treatment at $P < 0.01$ (**) or $P < 0.05$ (*).

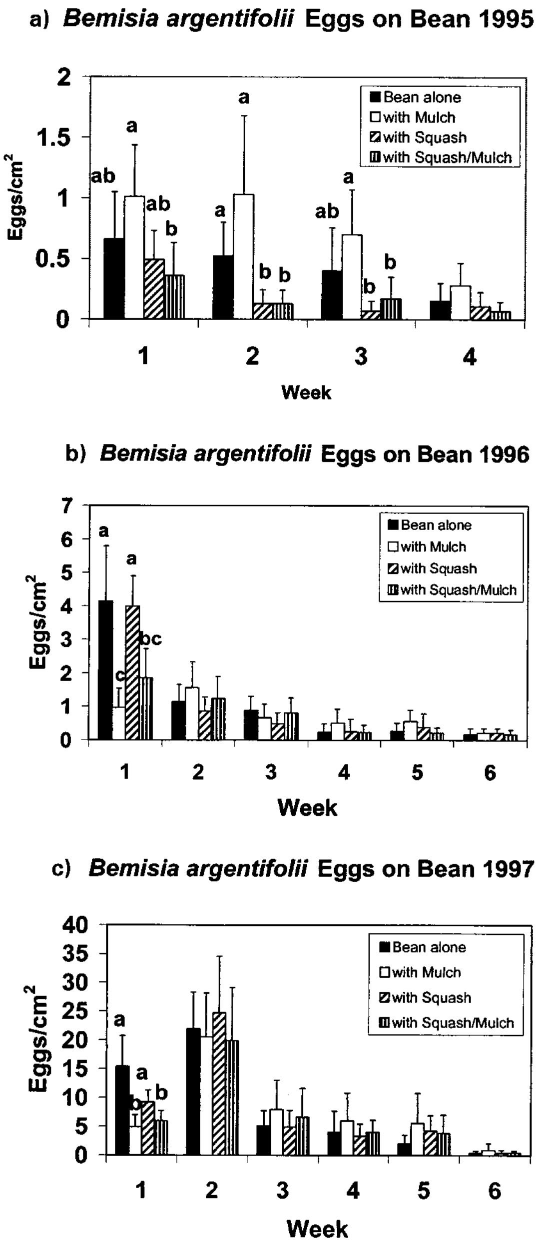

Fig. 2. Densities of *B. argentifolii* eggs on bean grown on bare ground, with silver reflective mulch, intercropped with squash on bare ground, and intercropped with squash on silver reflective mulch for 1995, 1996, and 1997 seasons. Columns with the same letter are not significantly different at $P < 0.05$ according to Tukey studentized range test.

on bean grown in monoculture during the first week of 1996 (Fig. 2b; $F = 6.20$; df = 3, 6; $P < 0.03$) and 1997 (Fig. 2c; $F = 6.59$; df = 3, 6; $P < 0.03$). There were no treatment differences on subsequent weeks during either of these years.

During the first 3 wk of sampling in 1995, egg densities were lower on bean in the squash/mulch treatment than on bean grown on bare ground and bean grown with mulch (Fig. 2a). Egg densities were lower on bean in both of the squash treatments (bare ground and reflective mulch) than on bean grown on reflective mulch during week 2 and 3 of 1995 (week 1: $F = 5.34$; df = 3, 6; $P < 0.04$; week 2: $F = 9.24$; $P < 0.01$; week 3: $F = 5.55$; $P < 0.04$). Given the low egg densities in 1995, the significance of these treatment differences is unclear.

Table 1. Bean yield

Year	Treatment	Total bean yield, kg/plot
1996	Bean	6.22b[a]
	Mulch	15.68a
	Squash	4.52bc
	Squash/Mulch	11.42ab
1997	Bean	0.57
	Mulch	0.68
	Squash	0
	Squash/Mulch	1.24

[a] Means in the same column with the same letter are not significantly different according to Tukey studentized range test. The absence of letters in a column indicates the lack of significant differences among means.

Differences in nymphal densities between squash and bean and across bean treatments were rare each year, and showed no consistent pattern (data not shown). Densities of parasitized nymphs were significantly higher on bean grown in monoculture ($0.34 \pm 0.57/cm^2$ foliage) than on the bean grown with reflective mulch ($0.13 \pm 0.37/cm^2$) on week six of 1997 ($F = 4.77$; df = 3, 6; $P < 0.04$). Parasitism was not high enough during other years for statistical comparison.

Yield. Crops froze in 1995 before yield could be harvested. In 1996, yields were highest in the mulched treatments (Table 1). Yields were extremely low in 1997, presumably because of high whitefly pressure.

Virus. In 1996, only one plant (in the squash treatment) tested positive for the presence of bean golden mosaic geminivirus. Virus presence was much higher in 1997. There were no statistical differences in the average number of plants (±SD) testing positive for the presence of virus among the 24 plants sampled per treatment (bean: 9.0 ± 0.5; mulch: 10.0 ± 0.5; squash/mulch: 8.0 ± 0.5; squash: 6.0 ± 0.5).

Discussion

Squash consistently elicited a higher oviposition response than bean in the two treatments that contained both squash and bean. However, the presence of squash did not reduce egg densities or virus incidence on adjacent bean compared with bean grown in monoculture. That is, squash apparently did not draw whitefly females away from the bean, and so did not function as a trap crop. The silver reflective mulch reduced egg densities compared with bean grown in monoculture during the first week of sampling for 2 of the 3 yr of the study. However, the suppressive effect of the reflective mulch, which is known to be insufficient as a solitary control measure, was not enhanced by the presence of squash in the squash/mulch treatment.

The early season protection that we observed with the reflective mulch was consistent with results from other studies (Csizinsky et al. 1997, Stansly and Schuster 1999). Trap crop studies using soybean (McAuslane et al. 1995), melon (Perring et al. 1995), and Wright's ground cherry (Ellsworth et al. 1994) also produced similar results: statistically lower whitefly densities on a few sampling dates, but no indication that the presence of the preferred crop consistently reduced whitefly numbers over time.

Because the trap crop mechanism relies on influencing herbivore behavior as the arthropod approaches or moves through a cropped area, it is possible that *B. argentifolii* may be a poor candidate for manipulation through trap cropping. *B. tabaci* apparently does not rely on host-specific visual or olfactory cues, but responds rather to the greenish-yellow range of light spectra reflected by most vegetation (Mound 1962, Byrne et al. 1996). Adults will move with air currents from crop to crop until they find an acceptable host (Byrne and Bellows 1991). Hunter et al. (1996) examined the precibarial and cibarial chemosensilla of *B. tabaci* and suggested that adults may be able to taste plant sap without ingesting it. If *B. argentifolii* adults require gustatory information to determine host suitability, as seems be true for *Trialeurodes vaporariorum* (Westwood) (van Lenteren and Noldus 1990, Lei et al. 1998), then adults may not make decisions related to host selection at a stage that would be influenced by the presence of a trap crop.

Schuster et al. (1996) required six rows of squash to observe a delay in virus incidence on adjacent tomato. We might have observed greater treatment differences if we had used larger plots and planted higher densities of squash. However, because the research was carried out on an active farm, only a limited area could be devoted to investigation. The work of Schuster et al. (1996) and Al-Musa (1982) suggests that certain crop combinations may be useful for reducing densities of *Bemisia* and *Bemisia*-vectored viruses. Our data indicate that squash was not useful as a trap crop in small plots, and that combining reflective mulch with a preferred crop such as squash offered no advantage over using reflective mulch on its own.

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