THE CITRUS LEAFMINER *PHYLLOCNISTIS CITRELLA* (LEPIDOPTERA: GRACILLARIIDAE)IN SOUTH TEXAS: INCIDENCE AND PARASITISM

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Abstract

The citrus leafminer, *Phyllocnistis citrella* Stainton (Lepidoptera: Gracillariidae), was first reported in the Lower Rio Grande Valley of Texas in August, 1994. We surveyed about 40 orchards in 1995 and 20 in 1996. Percentage of leaf infestation by the leafminer was lowest on the spring flush, and increased significantly in the early summer (May-July) and late summer flushes (Aug.-Oct.) through to late fall (Nov.-Dec.). Numbers of citrus leafminer immatures usually ranged from 0-6.8 per leaf. Several native parasite species were identified from the surveys, including 9 species of parasites from 3 families, Eulophidae, Proctorupidae and Ceraphronidae. The most abundant native parasitoid was *Zagrammosoma multilineatum* (Ashmead) (Eulophidae). Less dominant parasitoids were the eulophids *Horismenus* sp., *Closterocerus* sp., *Neochrysocharis* sp., *Pnigalio* sp., and *Tetrastichus* sp. Percentage parasitoid *Ageniaspis citricola* Logvinoskaya (Encyrtidae) was released in February-April 1995 and August-October 1996.

Key Words: biological control, parasites, population dynamics

RESUMEN

El minador de los cítricos, Phyllocnistis citrella Stainton (Lepidoptera: Gracillariidae), fue reportado por primera vez en la región llamada "Lower Rio Grande Valley" de Texas en agosto de 1994. Inspeccionamos aproximadamente 40 huertas en 1995 y 20 en 1996. El porcentaje de hojas infestadas por el minador tuvo su nivel más bajo durante el crecimiento del follaje de la primavera y aumentó significativamente a principios de verano (mayo-julio) y a finales de verano (agosto-octubre) hasta finales de otoño (nov.-dec.). El número de larvas del minador generalmente varió de 0 a 6.8 por hoja. Se identificaron varias especies nativas de parásitos durante estas inspecciones, incluyendo a 9 especies en 3 familias, Eulophidae, Proctotrupidae y Ceraphronidae. La especie de parasitoide nativa más abundante fue Zagrammosoma multilineatum (Ashmead) (Eulophidae). Entre los parasitoides menos dominantes estuvieron los Eulophidae Horismenus sp., Closterocerus sp., Neochrysocharis sp., Pnigalio sp., y Tetrastichus sp. El porcentaje de parasitismo por parasitoides nativos generalmente varió del 5 al 10%. El parasitoide exótico Ageniaspis citricola Logvinoskaya (Encyrtidae) fue liberado en los meses de febrero y abril de 1995 y en agosto y octubre de 1996.

The citrus leafminer, Phyllocnistis citrella Stainton (Lepidoptera: Gracillariidae), is a serious pest of citrus and related plant species throughout Southern Asia, Australia and east Africa, and is native to eastern and southern Asia (Beattie 1993). In May 1993, citrus leafminer was discovered to have invaded southern Florida with over 90% infestation reported in Dade County (Heppner 1993a, see also Hoy 1996). By late 1994, the invasion had spread beyond Florida to Louisiana and Texas. The leafminer begins to damage the host plant as soon as its eggs hatch. The larvae bore through the leaf epidermis, ingesting the sap and producing a chlorotic leaf patch. Citrus leafminer may prevent young leaves from expanding, causing them to remain curled and twisted. Citrus leafminer may also attack succulent stems and fruits. After the miner has finished feeding, other insects such as the aphids; e.g., Aphis gossypii Glover and mealybugs, Planococcus citri (Risso) may continue feeding on the damaged area. Secondary effects of citrus leafminer damage may also include leaf desiccation or invasion by fungi and bacteria (Achor et al. 1997). Spatial distribution of eggs and larvae have been found statistically aggregated within the canopy. Egg and larval densities are also significantly higher on apical leaves on young branches compared with older, middle and bottom leaves on young branches of lime (Peña & Schaffer 1997).

Citrus leafminer was first reported in the Lower Rio Grande Valley of Texas in August 1994 (French et al. 1994). Surveys conducted immediately following the discovery revealed that the pest was already well established in several orchards and in a large citrus nursery. Practically all Texas citrus is currently grown in the Lower Rio Grande Valley, with about 35,000 acres estimated at an economic value of \$130-150 million. The leafminer invasion could constitute a serious setback to an industry still recovering from devastating freezes in 1983 and 1989 during which acreage declined from ~70,000 to 12,000. Acreage then increased to about 35,000 acres. Furthermore, citrus leafminer could spread throughout the southeast United States, probably as far north as Georgia.

Biological control may be a useful tool in suppressing populations of this insect (Hoy & Nguyen 1994a). Effective chemical control is difficult because this pest can develop resistance to pesticides, and its larvae are protected from insecticides by the leaf cuticle. The pupae are protected by the rolled leaf margins. A number of natural ene-

mies have been found for citrus leafminer, including 39 species of parasites from 7 families, mostly Chalcidoidea (Heppner 1993b). Ten species of chalcidoid parasites were reared from the citrus leafminer in Thailand (Hoy & Nguyen 1994a), with *Ageniaspis citricola* Logvinoskaya probably being the most significant. Schauff (1998) described two new species of Eulophidae reared from the citrus leafminer from Puerto Rico and Colombia. Currently, nearly 80 species of parasitoids have been reared from citrus leafminer throughout the world (LaSalle & Peña 1997, citing Schauff et al. [submitted]). Many are indigenous parasitoid species that have parasitized citrus leafminer as it has spread from the tropics—over 20 such species are listed by LaSalle & Peña (1997, citing Schauff et al. [submitted]).

In February-April, 1995, and August-October, 1996, the parasitoid *Ageniaspis citricola* Logvinoskaya (Encyrtidae) was released in the Lower Rio Grande Valley in collaboration with J. Goolsby (APHIS Mission Biological Control Center, TX). The objective of this study is to survey the citrus leafminer incidence and native complex parasitism, and determine the degree of establishment of *A. citricola*.

MATERIALS AND METHODS

Survey for citrus leafminer and native parasites.

We surveyed about 40 commercial orchards in the Lower Rio Grande Valley in 1995 and 20 in 1996, over a survey area of ~100 mi. in diameter (Fig. 1). No samples



Fig. 1. Sample locations in the Lower Rio Grande Valley (1995-1996).

were collected in January of both years due to the cold conditions (below freezing temperatures at certain periods) in the area. Six-26 leaf terminals were randomly collected per site to determine leaf infestation from February to December, 1995. In 1996, twelve leaf terminals were collected from 1 orchard from the east, central and western regions of the valley every 3 weeks. Leaf terminals were also collected from another 3-5 commercial orchards across the valley that were sampled randomly every 1-2 months. Percentage parasitism by the endemic parasite complex was calculated from a collection of 25-100 leaves per site at various times throughout 1995-1996. Parasitized citrus leafminer were reared through in the laboratory for species identification and to determine seasonal distribution of their native and exotic parasites. Parasites reared in the laboratory were identified by Dr. Michael E. Schauff (USDA Systematics Laboratory, Beltsville, MD) and Dr. James B. Woolley (Department of Entomology, Texas A&M University, College Station, TX). Voucher specimens were kept in the insect museums of the latter institutes.

Release and evaluation of an exotic parasite.

At several sites in the 3 regions of the Lower Rio Grande Valley, the exotic parasitoid *Ageniaspis citricola* was released (obtained from M. A. Hoy, University of Florida and R. Nguyen, Division of Plant Industry, FL) in February-April, 1995 and August-October 1996. Orchards that had a substantial population of first instar citrus leafminer on very young flushes were selected as release sites. In 1995, a total of 868 and 85 adult female parasites were released in the central (February, April, July) and western (July) parts of the valley. In 1996, 626 (August) and 480 (October) adult female parasites were released in the central regions, respectively. Subse-

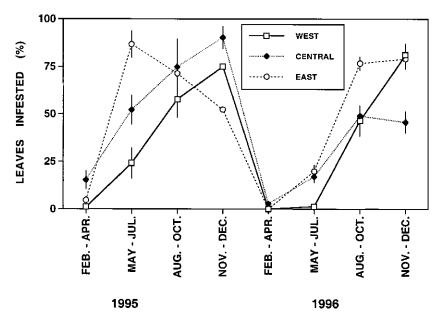


Fig. 2. Mean percentage leaves infested $(\pm$ SEM) by citrus leafminer in 3 regions of the Lower Rio Grande Valley (1995-1996).

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quent evaluation involved a random collection of up to 140 older leaves per site at various times after release. The number of A. *citricola* pupae and adults that emerged were counted and recorded.

RESULTS AND DISCUSSION

Survey for citrus leafminer and native parasites.

In 1995, percentage leaf infestation was lowest on the spring flush, and increased significantly on the early summer (May-July) and late summer (Aug.-Oct.) flushes through to late fall (Nov.-Dec.) (Fig. 2). Mean percentage leaf infestation (average across the 3 regions) increased steadily throughout 1995 until the winter: 7.03% (Feb.-Apr.), 54.3% (May-Jul.), 67.6% (Aug.-Oct.), and 72.5% (Nov.-Dec.). Mean percentage infestation (across the 3 regions) began to increase by the following spring: 12.8% (May-Jul.), 57.3% (Aug.-Oct.), and 68.6% (Nov.-Dec.). The total numbers of leaf-

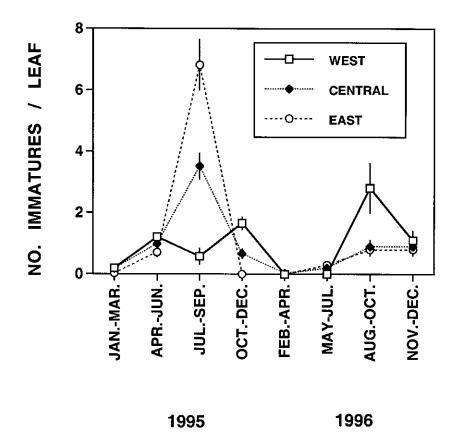
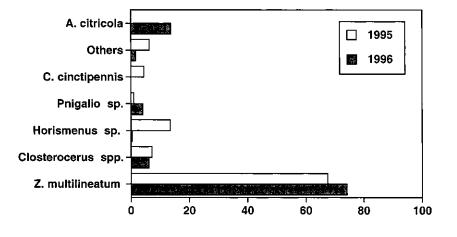


Fig. 3. Mean number of immature citrus leafminer per leaf (\pm SEM) in 3 regions of the Lower Rio Grande Valley (1995-1996).

Date	West			Central			East		
	Eggs	Larvae	Pupae	Eggs	Larvae	Pupae	Eggs	Larvae	Pupae
Jan-Mar '95	$\begin{array}{c} 0.048 \\ (0.022) \end{array}$	$\underset{(0.036)}{0.145}$	$\begin{array}{c} 0.000 \\ (0.000) \end{array}$	$\begin{array}{c} 0.003 \\ (0.003) \end{array}$	$\underset{(0.024)}{0.174}$	$\begin{array}{c} 0.000 \\ (0.000) \end{array}$	$\begin{array}{c} 0.000 \\ (0.000) \end{array}$	$\underset{(0.012)}{0.016}$	$\begin{array}{c} 0.008 \\ (0.008) \end{array}$
Apr-Jun '95	$0.348 \\ (0.050)$	0.822 (0.080)	$0.040 \\ (0.012)$	$0.195 \\ (0.034)$	$0.735 \\ (0.071)$	0.056 (0.022)	$0.026 \\ (0.026)$	0.684 (0.137)	0.000 (0.000)
Jul-Sep '95	$0.053 \\ (0.053)$	$0.526 \\ (0.246)$	0.000 (0.000)	$0.957 \\ (0.215)$	2.429 (0.353)	$0.121 \\ (0.036)$	3.389 (0.585)	$3.426 \\ (0.404)$	0.000 (0.000)
Oct-Dec '95	$0.452 \\ (0.118)$	0.839 (0.123)	(0.366) (0.088)	$0.061 \\ (0.030)$	$0.576 \\ (0.138)$	$0.030 \\ (0.021)$	0.000 (0.000)	0.000 (0.000)	0.000 (0.000)
Feb-Apr '96	0.000 (0.000)	0.000 (0.000)	0.000 (0.000)	0.000 (0.000)	0.024 (0.013)	$0.002 \\ (0.002)$	0.000 (0.000)	0.000 (0.000)	0.000 (0.000)
May-Jul '96	0.000 (0.000)	0.007 (0.003)	0.001 (0.001)	$0.034 \\ (0.014)$	$0.123 \\ (0.038)$	0.022 (0.009)	$0.107 \\ (0.037)$	0.187 (0.052)	0.030 (0.008)
Aug-Oct '96	$0.854 \\ (0.276)$	1.881 (0.711)	0.093 (0.023)	$0.055 \\ (0.021)$	0.691 (0.208)	$0.119 \\ (0.052)$	$0.152 \\ (0.062)$	0.522 (0.139)	$0.094 \\ (0.022)$
Nov-Dec '96	$\begin{array}{c} 0.022 \\ (0.019) \end{array}$	$\begin{array}{c} 0.798 \\ (0.305) \end{array}$	$\begin{array}{c} 0.223 \\ (0.069) \end{array}$	$\begin{array}{c} 0.310 \\ (0.145) \end{array}$	$\begin{array}{c} 0.553 \\ (0.154) \end{array}$	$0.029 \\ (0.020)$	$\begin{array}{c} 0.115 \\ (0.065) \end{array}$	$\begin{array}{c} 0.509 \\ (0.172) \end{array}$	$\begin{array}{c} 0.210 \\ (0.071) \end{array}$

TABLE 1. MEAN NUMBERS OF CITRUS LEAFMINER AT DIFFERENT STAGES PER LEAF (± S. E.) IN THREE REGIONS OF THE LOWER RIO	GRANDE VALLEY
OF TEXAS IN 1995 AND 1996.	



PARASITE SPECIES

ABUNDANCE (%)

Fig. 4. Overall percentage abundance of the native Zagrammosoma multilineatum, Closterocerus sp., Horismenus sp., Pnigalio sp., Closterocerus cinctipennis and the exotic (Ageniaspis citricola) parasitoids of the citrus leafminer. "Others" include the natives Neochrysocharis sp., Tetrastichus sp., Proctotrupidae (1995), and Ceraphronidae (1996).

miner immatures ranged from 0-6.8 per leaf, with peak samples collected in July to September, 1995 from the East region (mean immatures \approx 6.8) and in August to October, 1996 from the West (mean immatures \approx 2.8) (Fig. 3). Table 1 presents the mean numbers of citrus leafminer at different stages per leaf in the three regions of the Lower Rio Grande Valley in 1995 and 1996 (see Table 1). Similar trends were reported by Peña et al. (1996) for Florida, wherein leafminer densities were found to increase from spring through fall, and declined during the winter.

Several native parasite species were identified from the surveys, including 9 species of parasites from 3 families, Proctotrupidae, Ceraphronidae and the more common Eulophidae (Fig. 4). The most abundant native parasitoid was Zagrammosoma multilineatum (Ashmead), which comprised 68% and 74% of the parasitoid complex sampled in 1995 and 1996, respectively. Less dominant parasitoids were the eulophids Horismenus sp., Closterocerus sp., Neochrysocharis sp., Pnigalio sp., Tetrastichus sp. and a few others that were unidentified. In 1996, Ageniaspis citricola was collected at the site of release and this exotic species was found to be a substantial percentage (17%) of all the parasites recovered (see Fig. 4). Percentage parasitism by native parasitoids usually ranged from \approx 5-10%, but showed extremes in the samples taken from the east and central region, where parasitism ranged from 0-24.4% and 0-34.9%, respectively (Fig. 5). Seasonal distribution of the native parasites and the exotic species, A. citricola, are shown in Figs. 6A and 6B. In 1995 and 1996, the parasites were mostly abundant from August through December. Following the releases of A. citricola in August and September 1996, peak numbers of A. citricola were about as

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high as those recorded for the native parasite, *Z. multilineatum*, in late August 1996. As may be expected, parasitization patterns of citrus leafminer during the two years (Fig. 5) were broadly similar to percentage leaf infestation (Fig. 2). The native parasitoid complex of south Texas was similar to those at Colima (Perales-Gutiérrez et al. 1996a and 1996b), and perhaps to a lesser extent, to Tamaulipas, in Mexico (Martínez-Bernal & Ruíz-Cancino 1996). In contrast, the most dominant native parasitoid of citrus leafminer in Florida was the eulophid *Pnigalio minio* (Walker) which comprised \approx 80% of parasitoids which emerged from parasitized miners (Peña et al. 1996). Only 3 individual *Pnigalio* sp. was reared from our samples.

Release and evaluation of an exotic parasite.

The release of the exotic parasitoid *Ageniaspis citricola* in Feb.-Apr. 1995 was not successful because subsequent evaluations revealed that no parasitoids were recovered at the sites. The unsuccessful recoveries may be due to weather conditions and availability of suitable hosts in the valley at the time of release. Mean temperature, cumulative rainfall and relative humidity were: $18.9^{\circ}C$ (66.1°F), 0.4 cm (0.16 in),

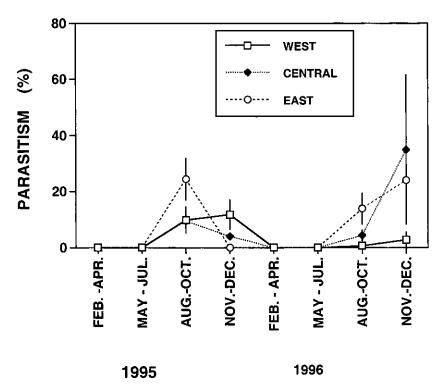


Fig. 5. Mean percentage parasitism (\pm SEM) by native parasites of citrus leafminer in 3 regions of the Lower Rio Grande Valley (1995-1996). Percentage parasitism by native parasitoids usually ranged from 5-10%, but showed extremes in the samples taken from the east and central region, where parasitism ranged from 0-24.4% and 0-34.9%, respectively.

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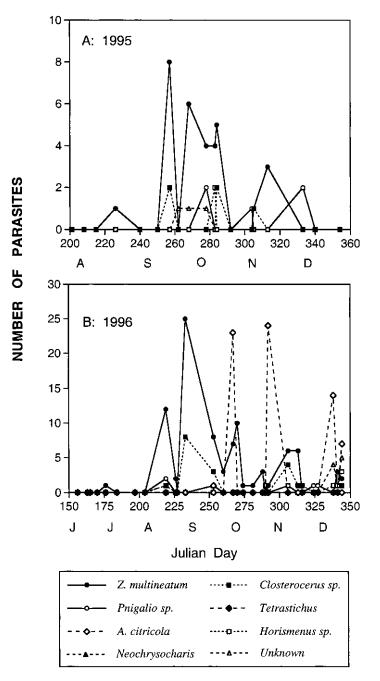
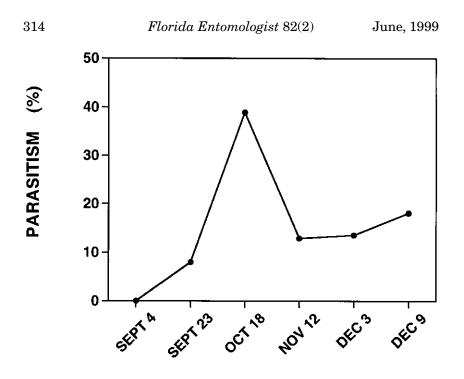


Fig. 6. Total number of parasites of citrus leafminer reared in the laboratory from leaf samples collected in the Lower Rio Grande Valley in 1995 (A) and 1996 (B).



SAMPLE DATE (1996)

Fig. 7. Overall percentage parasitism by *Ageniaspis citricola* (released in August 1996) of citrus leafminer in the eastern region of the Lower Rio Grande Valley. Recovery samples resulted in up to 39% parasitism in the field.

74.1%, respectively for Feb. 1995; and 23.8°C (74.9°F), 6.2 cm (2.46 in) and 75.6%, respectively for Apr. 1995. Maximum temperatures reached 31.7°C (89°F) in Feb. 1995 and 36.1°C (97°F) in Apr. 1995.

In August 1996, following a release of 626 adult parasite females in the east region of the valley, recovery samples resulted in up to 39% parasitism in the field (Fig. 7). Mean temperature was 29.3°C (84.7°F) and cumulative rainfall was 11.8 cm (4.66 in) in August 1996. Further surveys indicate that A. citricola may disperse from the site of release (French & Legaspi 1996). The higher parasitism rates may be due to the more humid conditions in August compared to the earlier releases in February to April. Rainfall also occurred about 10 minutes after the parasites were released. Precipitation is usually highest during the months of August to September in the Lower Rio Grande Valley. In addition, the eastern region is closer to the coast (Gulf of Mexico); conditions are more humid than the central and west parts of the valley. The presence of suitable hosts may also have contributed to the increased incidence of the leafminer parasites (Figs. 2 and 3). Although A. citricola were recovered soon after its release in 1996, no recoveries of A. citricola were made in 1997. Apparently, the parasites were not able to overwinter successfully. Moreover, high humidity and presence of host eggs and first-instars are essential in survival and reproduction (Edwards and Hoy 1998). Releases of A. citricola in Florida have produced promising results, with

some fields yielding almost 100% pupal parasitization during certain periods (Hoy & Nguyen 1994b). Further studies will include continued monitoring of the incidence of citrus leafminer throughout the Lower Rio Grande Valley and assessments of the impact of parasitoids.

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