Sampling *Eoreuma loftini* (Lepidoptera: Pyralidae) on Texas Sugarcane

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Environ. Entomol. 25(1): 7–16 (1996)

ABSTRACT Sugarcane fields of different types were sampled for 3 yr to determine the seasonal density, spatial dispersion, and within-field stratification of larvae and pupae of Mexican rice borer, *Eoreuma loftini* (Dyar). Population density never exceeded 1.0 larva per stalk in any year. Ratoon fields of sugarcane regrowth averaged more larvae per stalk than plant cane or hot water-treated fields. There was a trend for hot water-treated fields to contain a higher proportion of younger larvae than other fields, an indication that infestations in hot water-treated fields come from moths migrating into the fields rather than from localized infestations from planted billets. Greater numbers of older larvae and pupae were found later in the season than earlier. There was more stalk-to-stalk and field-to-field variation in larval dispersion than among blocks, tiers, or plots within fields. Sample sizes for use in estimating *E. loftini* larval density with a defined level of reliability are presented.

KEY WORDS Mexican rice borer, *Saccharum*, stalkborers, within-field distribution, dispersion

MEXICAN RICE BORER, *Eoreuma loftini* (Dyar), was first detected in the lower Rio Grande Valley of Texas in 1980 (Johnson 1981, Johnson and van Leerdam 1981) and is now the primary insect pest of sugarcane. Meagher et al. (1994) described the biology and geographical range of *E. loftini*. Larvae injure sugarcane and other gramineous crops by feeding on leaf sheaths, tunneling within stalks, and causing *deadhearts* (plants that have a dead apical meristem) (Browning et al. 1989). Injury and yield loss result from feeding on internodes and buds, lodging, and reduction of sugar juice quantity and quality (van Zwaluwenburg 1926, Flanders 1930, Johnson 1981, Rozeff 1981). Surveys in the lower Rio Grande Valley in 1989 and 1990 showed an average of nearly 19% bored internodes by *E. loftini* (Meagher et al. 1992).

In Texas, sugarcane is vegetatively propagated from 0.8-m stalk cuttings (billets) containing several growing points and leaf and root primordia (van Dillewijn 1952). Fields are planted using billets on 1.52-m row centers in late summer. A small percentage of these fields have billets treated in water at 50°C for 2 h before planting to control ratoon stunting disease, *Clavibacter xyli* subsp. *xyli* (Davis et al. 1984, Gillaspie and Teakle 1989). Sugarcane is harvested from late September to March or April, depending on weather. Regrowth (ratoons) fields may be cycled >5 yr before replanting. Therefore, 3 different types of fields may be present at any time in the lower Rio Grande Valley sugarcane agroecosystem: plant cane, hot water-treated plant cane, and ratoon cane.

Studies conducted in the Caribbean and in Florida have shown that other stalkboring pyralid larvae have random (des Vignes 1978) or aggregated (Hall 1986, Hughes and White 1987 [based on stalk injury, not larval density]) spatial patterns. The study in Florida with *Diatraea saccharalis* (F.) showed populations to be aggregated within small areas of sugarcane (0.4–0.8 ha) and across large sugarcane fields (Hall 1986). Spatial patterns of larvae and pupae of 3 stalkborers, including *E. loftini* and *D. saccharalis*, were described from corn planted in northern Mexico (Rodriguez-del-Bosque et al. 1990). All species were generally aggregated; however, aggregation was greater for the younger larvae, with older larvae and pupae tending to have random spatial patterns.

An understanding of the spatial patterns of an insect can aid in developing a sampling plan (Southwood 1978). Spatial patterns are affected by intrinsic factors such as oviposition pattern and immature dispersal; extrinsic factors such as host quality and environmental toxicants; and estimation procedures such as sampling plan parameters, sample unit size, number of samples, and sampler bias (Wilson 1994). Spatial patterns can depend on population density, with low densities tending to be uniform or random and high densities tending to be aggregated. The efficiency of a resulting sam-

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0046-225X/96/0007–0016$02.00/0

Table 1. Field size and configuration, number of sampling dates, plot size, and sample interval for 9 sugarcane fields sampled in Hidalgo and Cameron counties, TX, 1990–1992

Year	Field type, location	Size, configuration	Sample no.	Plot size	Sample interval
1990	HWT, Weslaco[a]	6.3 ha, 104 rows by 396.2 m	6	13 rows by 19.8 m	26 Feb.–15 May
	Plant, Weslaco[b]	16.0 ha, 264 rows by 396.2 m	6	13/14 rows by 19.8 m	26 Feb.–15 May
	Ratoon (1st), La Villa[c]	14.5 ha, 260 rows by 396.2 m	6	13 rows by 19.8 m	26 Feb.–28 May
1991	HWT, Primera[d]	14.1 ha, 259 rows by 362.4 m	7	13 rows by 18.3 m	28 Feb.–9 May
	Plant, Alamo	15.7 ha, 251 rows by 329.2 m	8	12 rows by 16.5 m	29 Jan.–13 May
	Ratoon (1st), Runn	14.4 ha, 271 rows by 349.0 m	8	13/14 rows by 17.4 m	30 Jan.–6 May
1992	HWT-R, Primera[e]	14.1 ha, 259 rows by 362.4 m	9	13 rows by 18.3 m	14 March–30 July
	Plant, Bluetown	24.8 ha, 317 rows by 512.7 m	9	15 rows by 25.6 m	12 March–28 July
	Ratoon (1st), Alamo[f]	15.7 ha, 251 rows by 329.2 m	7	12 rows by 16.5 m	30 April–26 Aug.

[a] HWT (hot water-treated plant field) and plant fields were part of 1 large field.
[b] A large residence occupied 2.5 ha of the field (30 plots); missing plots were in the following blocks; 17 (6), 18 (4), 22 (12), 23 (8).
[c] Eight plots in block 24 (1) and 25 (7) were missing because of a shed.
[d] The last 16 rows (1 ha; blocks 5, 10, 15, 20, and 25) were cultivar TCP 81-3058.
[e] Hot water-treated field sampled in 1991, sampled as a ratoon field in 1992.
[f] Plant field sampled in 1991, sampled as a ratoon field in 1992.

pling plan must balance cost considerations against the quality of the information provided. The 3 objectives of the current research were: (1) to examine seasonal densities of different-aged *E. loftini* larvae across different field types, (2) to quantify spatial patterns as affected by field type and larval size, and (3) to develop sampling plans for different *E. loftini* larval densities for use in estimating population densities with a defined level of reliability.

Materials and Methods

Fields and Sampling. Field sizes, configurations, plot sizes, and sample intervals are summarized in Table 1. All fields were planted to cultivar CP 70-321, except where noted. Each field was divided into 25 blocks, consisting of an outer tier of 16 blocks, an inner tier of 8 blocks, and a center tier of 1 block. Each block was subdivided into 16 plots. Exceptions to this arrangement were in a 1990 hot water-treated sugarcane field which had 20 blocks, 8 plots per block (14 outer, 4 inner, and 2 center blocks), and a 1990 plant field which had unequal replication because of a residence within the field. Plot size was variable, ranging from 12 to 15 rows (16.7–21.3 by 16.5–25.6 m) (Table 1). Sampling began in late winter–early spring and ended mid- to late summer (Table 1). Every effort was taken to maintain a biweekly timetable; however, weather conditions or irrigation schedules occasionally delayed sampling. Sampling dates were grouped into early-(January–April) and late-season (May–August) categories, characterized by plant growth and tillering before internode formation and by rapid plant growth and formation of harvestable internodes (Meagher et al. 1994).

On each sample date, 4 plots per block were randomly selected from each field. Ten stalks per plot were searched for *E. loftini* larvae or pupae. Randomly selected stalks were cut at ground level, dissected, and the presence of stalkborer larvae recorded. Larvae (< 3rd instar) were counted by removing the leaf sheaths and inspecting each leaf sheath for live larvae. Discoloration, superficial tunneling in the leaf sheaths, and frass were used to direct the search for stalkborer larvae (Meagher et al. 1994). Larvae were classified according to the sizes: small, medium, or large, which corresponded to 1st and 2nd (age class 1), 3rd and 4th (age class 2), and 5th and 6th (age class 3) instars and pupae, respectively.

Statistical Analysis. *Seasonal Density.* Analysis of variance (ANOVA) (PROC GLM, SAS Institute 1985) was used to assess effects of field type (hot water-treated, plant, or ratoon), and time of season on density (larvae per stalk) and larval age for 1990, 1991, and 1992. Larval age was calculated by averaging age class numbers by field type and time of season. The 1992 hot water-treated ratoon field was a ratoon of the 1991 hot water-treated field but in the analysis was classified as an hot water-treated field. Tier (outer, inner, center) was used as an independent variable in the analysis of density. Means were separated using the Waller-Duncan k-ratio t-test ($P < 0.05$) (SAS Institute 1985).

Aggregation. The aggregation characteristics of *E. loftini* were examined with Taylor coefficients (Taylor 1961, 1984) and variance to mean ratios (Wilson 1994). The mean and variance in larvae per stalk for small, medium, or large larvae and pupae across fields were used in estimating Taylor coefficients by fitting $\ln (S^2) = \ln a + b \ln (\bar{x})$ (PROC REG, SAS Institute 1985) for data grouped in single-stalk (n = 1,000 per field), 40-stalk (n = 40 per block), or 10-stalk (n = 10 per plot) sample unit sizes. Estimates for a and b were compared across sample years in ANOVAs (PROC GLM, mean separation by Waller–Duncan k-ratio t-test , SAS Institute 1985) to examine the effects of field type, larval size, and sample unit size.

The effect of larval density on aggregation was examined for each field type, larval size, larval density [low density (mean = 0.01 larva per stalk), av-

erage density (mean = 0.1344 larva per stalk) and high density (mean = 0.8 larva per stalk)], and sample unit size in an ANOVA. Aggregation was represented as variance to mean ratios which were estimated using Taylor coefficients derived for each of the independent variables ($S^2/\bar{x} = a\ \bar{x}^{[b-1]}$) (Wilson 1994).

Within-Field Stratification. A nested ANOVA (PROC NESTED, SAS Institute 1987) was used to partition the sources of variation in larval density each year (Snedecor and Cochran 1967, Hall et al. 1994). The following 2 hierarchical levels were constructed based on a single sugarcane stalk as the sample unit: (1) stalks nested in plots, plots nested in blocks, and blocks nested in fields; and (2) stalks nested in plots, plots nested in tiers, and tiers nested in fields. Mean densities per stalk were calculated across dates. Variance components and the corresponding percentage of total variation were calculated to assess origins of sampling variation among the hierarchical levels.

Estimated Sample Size. Wilson and Room (1982) presented the following equation for estimating sample size where the goal is to estimate population density with a defined level of reliability:

$$n = (t_{\alpha/2}/D_{\bar{x}})^2 a\bar{x}^{(b-2)},$$

where $t_{\alpha/2}$ is the standard normal variate for a 2-tailed confidence interval, $D_{\bar{x}}$ = a proportion defined as the ratio of half the desired confidence interval to the mean ($D_{\bar{x}}$ = [CI/2]/$\bar{x}$ for enumerative sampling), and a and b are Taylor coefficients. This equation permits one to estimate required sample size (n) over a range of densities for any species and sample unit whose Taylor coefficients are known. Required sample size was calculated using $t_{\alpha/2}$ = 1.645 and $D_{\bar{x}}$= 0.2. Sample size was then used as a dependent variable and examined for an influence of field type and larval size by ANOVA (PROC GLM) separately for each sample unit size and sample year.

Results

Seasonal Density. The population density of *E. loftini* remained <1.0 larva per stalk in all years (Figs. 1–3). Densities varied significantly with field type only in 1991 (F = 19.6; df = 2, 45; P < 0.0001), with ratoon fields containing more larvae per stalk than in plant or hot water-treated fields (mean ± SE, 0.076 ± 0.012, 0.045 ± 0.008, 0.009 ± 0.003, respectively). The trend for ratoon fields to contain more larvae than hot water-treated fields occurred in 1990 and 1992. More larvae per stalk were collected during the late season in 1991 and 1992, but the opposite was true for 1990 (P < 0.05) (1990: early 0.337 ± 0.04, late 0.169 ± 0.03; 1991: late 0.101 ± 0.02, early 0.04 ± 0.006; 1992: late 0.150 ± 0.04, early 0.014 ± 0.003). The difference in larval density between seasons was generally greater in ratoon fields than in hot water-treated or plant fields. Tier was not a significant factor in any year (P > 0.900).

Ages varied among field types only in 1991 (F = 6.6; df = 2, 15; P = 0.0088), with the youngest larvae found in the hot water-treated field compared with the plant or ratoon fields (1.14 ± 0.07, 1.35 ± 0.05, 1.36 ± 0.05, respectively). Larger numbers of older larvae and pupae were found in the late season compared with early season in all years (P < 0.001).

Aggregation. Estimates of Taylor a for 1990 showed differences among larval sizes (F = 8.1; df = 2, 7; P = 0.0152), with a greater a calculated when sampling for small larvae than for large larvae and pupae (small 1.59 ± 0.09, medium 1.18 ± 0.06, large 0.91 ± 0.09). Estimates for Taylor b showed differences among field types (F = 10.4; df = 2, 7; P = 0.0080) and larval sizes (F = 7.8; df = 2, 7; P = 0.0167). Samples taken in ratoon and plant fields had greater b values than in the hot water-treated field (1.08 ± 0.04, 1.05 ± 0.03, 0.95 ± 0.04, respectively), and sampling for small larvae produced a greater b than for large larvae and pupae (small 1.07 ± 0.06, medium 1.03 ± 0.02, large 0.97 ± 0.03). There was also a significant field type × larval size interaction (F = 6.2; df = 2, 7; P = 0.0281) because of a lesser b value for small larvae in the hot water-treated field than in other fields.

In 1991, Taylor a was greatest in samples taken from the hot water-treated field (hot water-treated 1.90 ± 0.38, plant 1.22 ± 0.08, ratoon 1.09 ± 0.06; F = 6.9; df = 2, 8; P = 0.0183). No other variables were significant for either of the Taylor coefficients (P > 0.05).

In 1992, Taylor a was different among larval sizes (F = 4.9; df = 2, 12; P = 0.0280) and sample unit sizes (F = 5.9; df = 2, 12; P = 0.0167). Large larvae and pupae had a greater a value than medium larvae (large 1.34 ± 0.08, small 1.24 ± 0.03, medium 1.17 ± 0.04), and the 40-stalk sample unit size had a greater a value than the single-stalk size (40, 1.35 ± 0.08; 10, 1.24 ± 0.04; single, 1.16 ± 0.04). There was a significant field type × larval size interaction (F = 4.5; df = 4, 12; P = 0.0185) because of a greater a value for medium larvae in the plant field than in other fields. Taylor b was different among larval sizes (F = 3.2; df = 2, 12; P = 0.0796) and sample unit sizes (F = 15.1; df = 2, 12; P = 0.0050). Large larvae and pupae had a greater b value than medium larvae (large 1.08 ± 0.02, small 1.05 ± 0.01, medium 1.04 ± 0.01), and the 40-stalk sample unit size had a greater b value than the 10- or single-stalk sizes (1.10 ± 0.02, 1.04 ± 0.01, 1.02 ± 0.01, respectively). Taylor coefficients for individual larval sizes and field types across sample year and sample unit sizes are shown in Table 2. Taylor b was generally >1.0 for only the 40- and 10-stalk sample unit sizes.

Aggregation, as measured by the variance to mean ratio, was significantly different among field types (F = 4.7; df = 2, 8; P = 0.0441) and larval

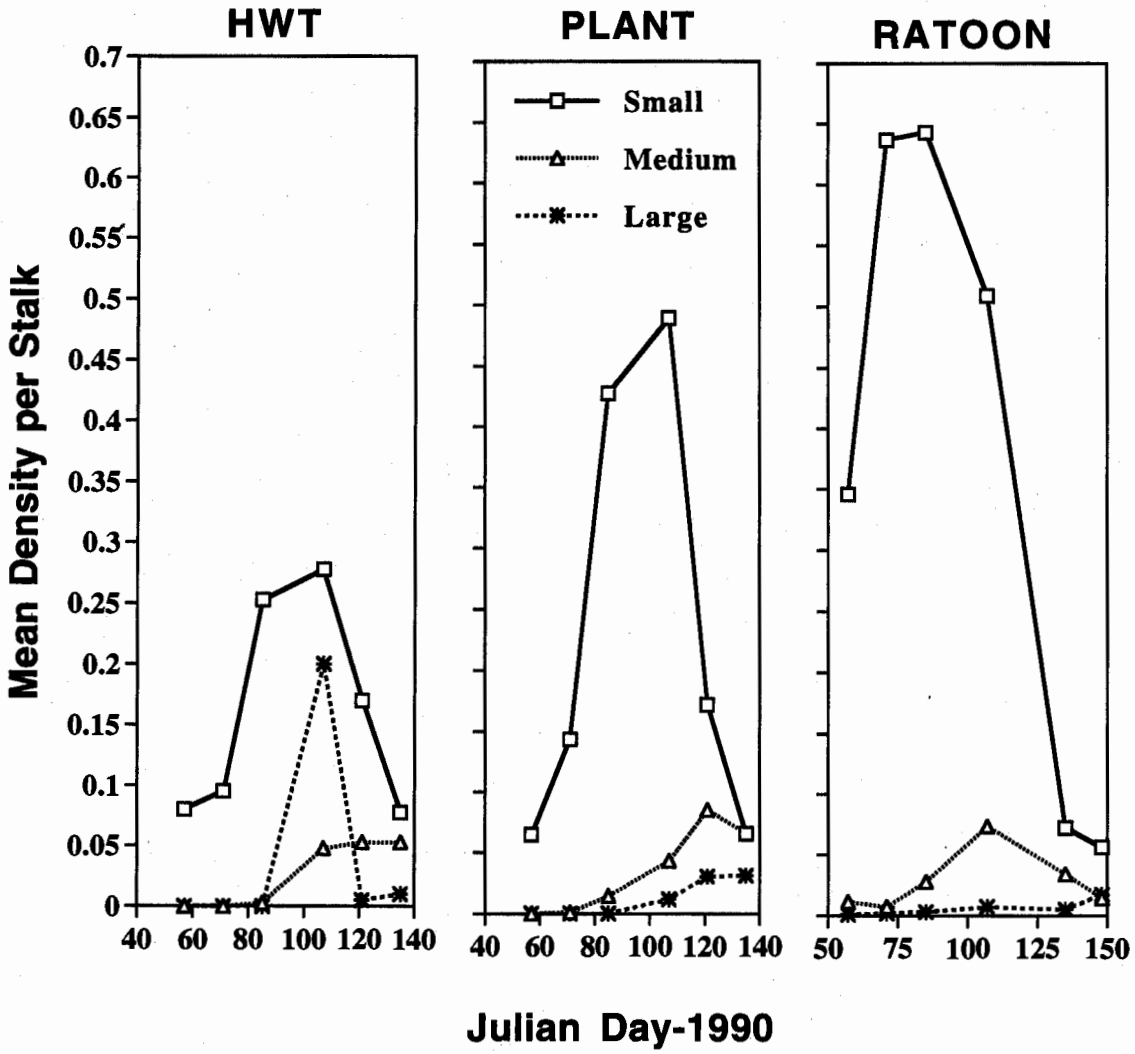

Fig. 1. Mean *E. loftini* small, medium, and large larvae per stalk for 3 sugarcane field types, Hidalgo and Cameron counties, TX, 1990.

sizes (F = 6.9; df = 2, 8; P = 0.0181) in 1990. Ratios ranged from 0.38 to 9.69. The hot water-treated field produced a greater ratio than the plant or ratoon fields (2.16 ± 0.52, 1.30 ± 0.14, 1.21 ± 0.12, respectively), and small larvae produced a greater ratio than medium or large larvae (2.06 ± 0.36, 1.14 ± 0.06, 0.95 ± 0.06, respectively). The interactions between field type and larval size (F = 6.0; df = 2, 8; P = 0.0261) and field type and larval density (F = 5.3; df = 4, 8; P = 0.0217) were significant. The field type × larval size interaction can be explained by a large variance to mean ratio estimated in the hot water-treated field for small larvae (3.23 ± 0.94) compared with the other field type–larval size combinations. The field type × larval density interaction is caused by a difference in direction of response of variance to mean ratios among field types. Greater variance to mean ratios were calculated for plant and ratoon fields with high density; the hot water-treated field had a large variance to mean ratio calculated with low density.

In 1991, aggregation was different only among larval densities (F = 12.9; df = 2, 8; P = 0.0031) because variance to mean ratio estimates calculated with high density were greater than those calculated with average and low density (1.69 ± 0.24, 1.07 ± 0.04, 0.91 ± 0.05, respectively). Ratios ranged from 0.55 to 5.01. In 1992, aggregation was significantly different among larval sizes (F = 4.3; df = 2, 16; P = 0.0331), sample units (F = 6.4; df = 2, 16; P = 0.0089), and larval densities (F = 37.9; df = 2, 16; P = 0.0001). Ratios ranged from 0.68 to 4.16. Large larvae and pupae were more aggregated than medium larvae (large 1.30 ± 0.13, small 1.18 ± 0.05, medium 1.12 ± 0.05); larvae

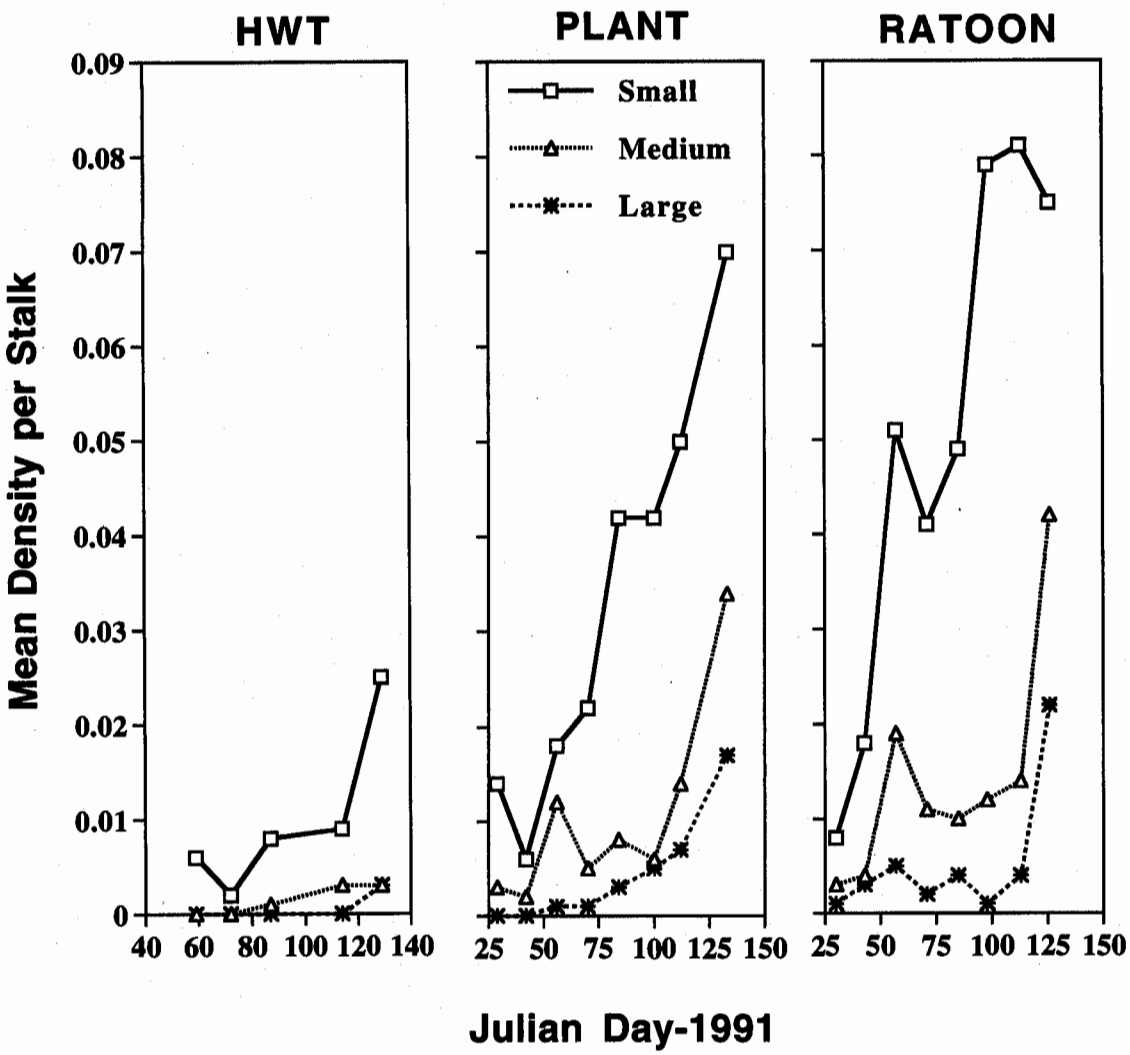

Fig. 2. Mean *E. loftini* small, medium, and large larvae per stalk for 3 sugarcane field types, Hidalgo and Cameron counties, TX, 1991.

collected from 40-stalk sample units were more aggregated than 10- or single-stalk sample units (1.33 ± 0.14, 1.16 ± 0.04, 1.10 ± 0.02, respectively); the estimate calculated with high density was greater than calculated with average density and low density (1.51 ± 0.12, 1.11 ± 0.02, 0.97 ± 0.03, respectively). The interactions between field type and larval density ($F = 3.4$; df = 4, 16; $P = 0.0341$) and sample unit and larval density ($F = 12.9$; df = 4, 16; $P = 0.0001$) were significant. These interactions can be explained by an increase in variance to mean ratios for high-density samples estimated from 40-stalk or ratoon field samples, respectively. The other larval density combinations showed similar variance to mean ratios across sample units and field types.

Within-Field Stratification. Hierarchical analysis disclosed that there was more stalk-to-stalk variation in *E. loftini* larval dispersion than any other hierarchical level (Table 3), followed by variation among fields, which supports the results of the initial ANOVA of seasonal density. Field-to-field variation was greatest in 1992 under both hierarchical levels. Block-to-block variation was significant in the 1990 and 1992 data, but explained only 1.7 and 3.6% of the total variation, respectively. Tier-to-tier and plot-to-plot variations were not significant in any year.

Estimated Sample Size. Only ANOVA of sample size estimates for single-stalk sample units in 1990 showed significant main effect variables in the models. Larger samples would be needed when sampling for small larvae compared with medium or large larvae and pupae (small 971.0 ± 48.2, medium 767.0 ± 20.3, large 661.2 ± 0; $F = 20.6$; df = 2, 2; $P = 0.0462$. All other main effects

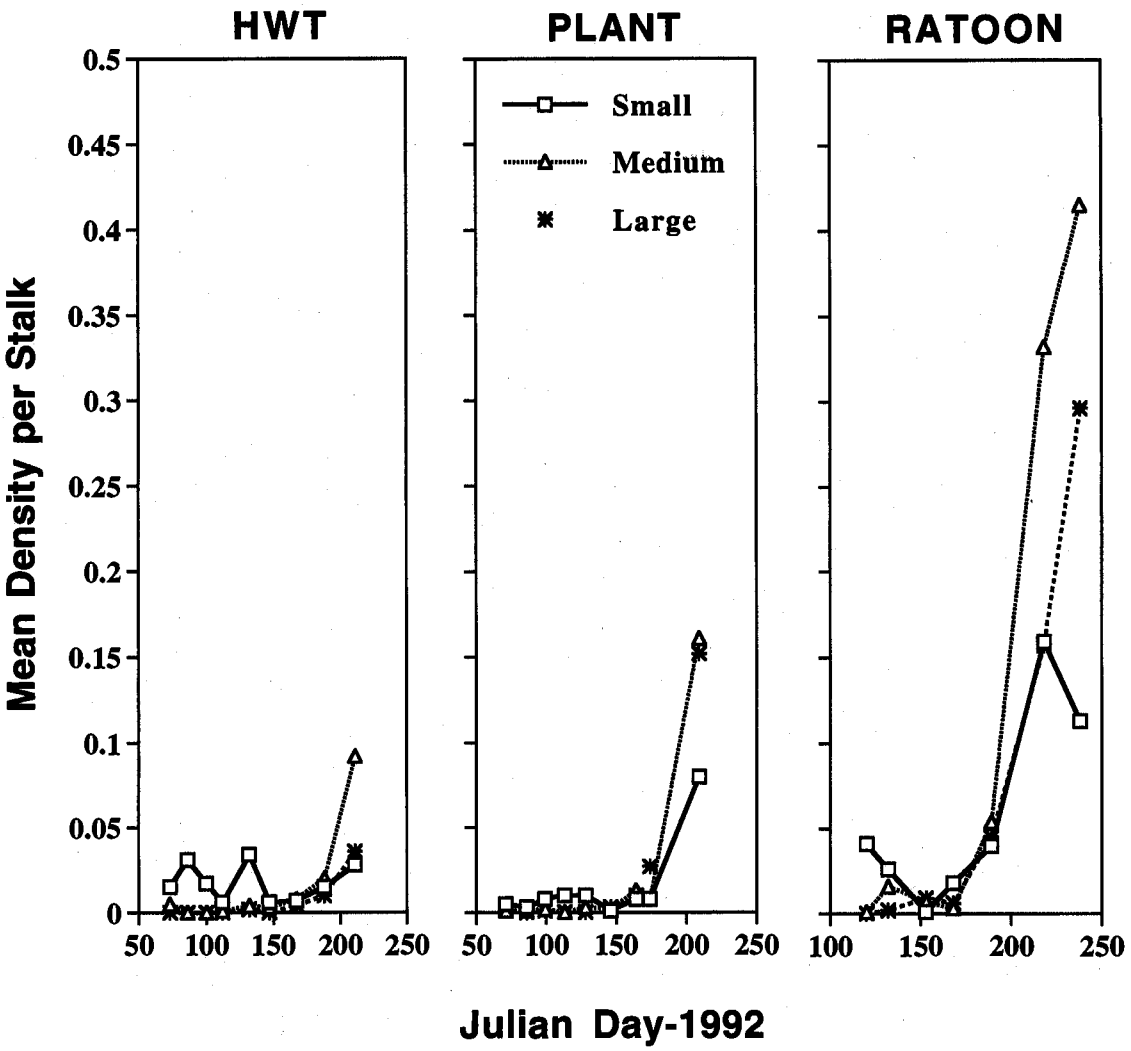

Fig. 3. Mean *E. loftini* small, medium, and large larvae per stalk for 3 sugarcane field types, Hidalgo and Cameron counties, TX, 1992.

were not significant ($P > 0.05$), although there were trends in other sample unit–sample year combinations for more samples needed when sampling for small larvae, or when sampling in hot water-treated fields. Density-dependent sample numbers per stalk across sample units are shown for the 3 sample years (Fig. 4).

Discussion

Although its biology of *E. loftini* is similar to that of other stalkborers, there are important differences that may influence its aggregation and spatial patterns. Females oviposit cryptically on dried plant material near the base of stalks by insertion of the egg mass into folded leaf crevices (van Leerdam et al. 1984, 1986). Most other stalkborers that attack sugarcane oviposit on leaves or stalks (Browning et al. 1989, Smith et al. 1993). Neonate *E. loftini* larvae disperse from soil-level locations vertically to plant leaves, and this behavior allows for mortality from ground predators. *Eldana saccharina* Walker, an African pyralid, has similar ovipositional and larval dispersal behavior (Girling 1978, Atkinson 1980). Research in Uganda and Ghana showed that by 3 wk after oviposition, almost 99.5% of the eggs and small larvae were attacked by ants, including *Tetramorium bicarinatum* (Nylander), *Camponotus sericeus* (F.), *Crematogaster* spp., and *Pheidole megacephala* F. (Girling 1978, Sampson and Kumar 1983). Early life stage mortality factors are unknown with *E. loftini*; however, ant predation is high when eggs are artificially deposited in the field (R.L.M and R.S.P, unpublished data). Several ant species are known to be active in the sugarcane agroecosystem

Table 2. Taylor coefficients for small, medium, and large *E. loftini* larvae, and for larvae found in HWT, plant, and ratoon sugarcane fields, across different sample unit sizes, Hidalgo and Cameron counties, TX, 1990–1992

Year	Larval size	*a* (±SEM)	*b* (±SEM)	r^2	*n*	Field type	*a* (±SEM)	*b* (±SEM)	r^2	*n*
					1,000 stalks field					
1990	Small	1.66 (1.12)	1.07 (0.06)	0.951	18	HWT	1.04 (1.27)	0.81 (0.13)	0.908	6
	Medium	1.19 (1.06)	1.02 (0.02)	0.997	15	Plant	1.17 (1.27)	0.91 (0.14)	0.913	6
	Large	0.96 (1.01)	0.99 (0.01)*	1.000	6	Ratoon	1.86 (1.22)	1.18 (0.14)	0.950	6
1991	Small	1.26 (1.11)	1.04 (0.03)	0.988	21	HWT	1.56 (1.26)	1.08 (0.05)	0.994	5
	Medium	1.04 (1.11)	1.00 (0.02)	0.993	16	Plant	1.13 (1.24)	1.01 (0.07)	0.977	8
	Large	1.05 (1.04)	1.01 (0.01)	0.999	16	Ratoon	1.19 (1.13)	1.05 (0.04)	0.991	8
1992	Small	1.21 (1.09)	1.03 (0.02)	0.992	25	HWT	1.19 (1.22)	1.03 (0.05)	0.982	9
	Medium	1.05 (1.06)	1.00 (0.01)	0.998	25	Plant	1.16 (1.10)	1.03 (0.02)	0.995	9
	Large	1.14 (1.02)	1.02 (0.01)***	0.997	25	Ratoon	0.94 (1.08)	0.96 (0.03)	0.995	7
					40 stalks/block					
1990	Small	1.58 (1.04)	1.14 (0.02)***	0.906	380	HWT	1.42 (1.11)	1.12 (0.06)*	0.799	103
	Medium	1.17 (1.08)	1.04 (0.02)	0.906	192	Plant	1.31 (1.06)	1.06 (0.03)*	0.895	141
	Large	1.12 (1.09)	1.03 (0.03)	0.954	83	Ratoon	1.62 (1.05)	1.15 (0.03)***	0.899	147
1991	Small	1.29 (1.05)	1.07 (0.02)***	0.932	317	HWT	2.11 (1.18)	1.21 (0.05)***	0.940	41
	Medium	1.03 (1.07)	1.01 (0.02)	0.945	137	Plant	1.13 (1.08)	1.03 (0.02)	0.927	146
	Large	1.04 (1.03)	1.01 (0.01)	0.997	63	Ratoon	1.08 (1.05)	1.02 (0.02)	0.948	166
1992	Small	1.22 (1.05)	1.05 (0.02)***	0.944	278	HWT	1.19 (1.07)	1.04 (0.02)*	0.947	129
	Medium	0.92 (1.03)	0.97 (0.01)**	0.972	196	Plant	1.14 (1.04)	1.04 (0.01)**	0.984	103
	Large	1.07 (1.04)	1.02 (0.01)	0.975	163	Ratoon	0.83 (1.03)	0.93 (0.01)***	0.969	136
					10 stalks/plot					
1990	Small	1.43 (1.02)	1.17 (0.02)***	0.836	983	HWT	1.40 (1.08)	1.16 (0.05)***	0.782	170
	Medium	1.28 (1.07)	1.10 (0.03)***	0.802	313	Plant	1.18 (1.09)	1.07 (0.03)**	0.791	417
	Large	1.06 (1.10)	1.02 (0.04)	0.850	101	Ratoon	1.44 (1.03)	1.20 (0.02)***	0.827	487
1991	Small	1.26 (1.04)	1.10 (0.02)***	0.863	512	HWT	1.50 (1.11)	1.18 (0.05)***	0.928	47
	Medium	1.08 (1.08)	1.03 (0.04)	0.826	183	Plant	1.12 (1.06)	1.05 (0.03)	0.844	258
	Large	1.13 (1.15)	1.05 (0.06)	0.808	75	Ratoon	0.99 (1.05)	0.99 (0.02)	0.837	347
1992	Small	1.27 (1.05)	1.10 (0.02)***	0.837	476	HWT	1.24 (1.07)	1.10 (0.03)*	0.834	226
	Medium	0.81 (1.03)	0.90 (0.02)***	0.827	452	Plant	1.12 (1.04)	1.06 (0.02)*	0.907	205
	Large	0.99 (1.04)	0.99 (0.02)	0.867	361	Ratoon	0.73 (1.03)	0.85 (0.02)***	0.805	362

HWT, hot water-treated. *, **, ***, Slope significantly different from 1.0, *F*-test, $P < 0.05, 0.01, 0.001$, respectively.

Table 3. Nested ANOVAs with block or tier as 2nd hierarchical variable, on the number of *E. loftini* larvae per sugarcane stalk, Hidalgo and Cameron counties, TX, 1990–1992

Source variation	df	MS	*F*	Variance component	% total
		1990			
Block					
Field	2	8.537	46.3[a]	0.011	8.1
Block	67	0.184	1.7[a]	0.002	1.7
Plot	162	0.106	0.8	−0.002	0.0
Stalk	2,088	0.128	—	0.128	90.2
Total	2,319	0.136	—	0.142	100.0
Tier					
Field	2	1.700	60.0[a]	0.017	15.7
Tier	6	0.028	0.5	−0.001	0.0
Plot	21	0.061	0.7	−0.003	0.0
Stalk	270	0.093	—	0.093	84.3
Total	299	0.100	—	0.111	100.0
		1991			
Block					
Field	2	0.853	66.7[a]	0.001	3.9
Block	72	0.013	0.7	−0.000	0.0
Plot	225	0.018	0.8	−0.000	0.0
Stalk	2,700	0.021	—	0.021	96.1
Total	2,999	0.021	—	0.022	100.0
Tier					
Field	2	0.133	23.0[a]	0.001	5.2
Tier	6	0.006	0.4	−0.002	0.0
Plot	27	0.013	0.7	−0.001	0.0
Stalk	324	0.019	—	0.019	94.8
Total	359	0.019	—	0.020	100.0
		1992			
Block					
Field	2	13.531	146.9[a]	0.013	29.4
Block	72	0.092	3.5[a]	0.000	3.6
Plot	225	0.026	0.9	−0.000	0.0
Stalk	2,700	0.031	—	0.031	67.0
Total	2,999	0.041	—	0.046	100.0
Tier					
Field	2	1.747	568.6[a]	0.015	38.4
Tier	6	0.003	0.2	−0.001	0.0
Plot	27	0.014	0.6	−0.001	0.0
Stalk	324	0.023	—	0.023	61.6
Total	359	0.032	—	0.038	100.0

[a] Significance ≤ $P = 0.001$.

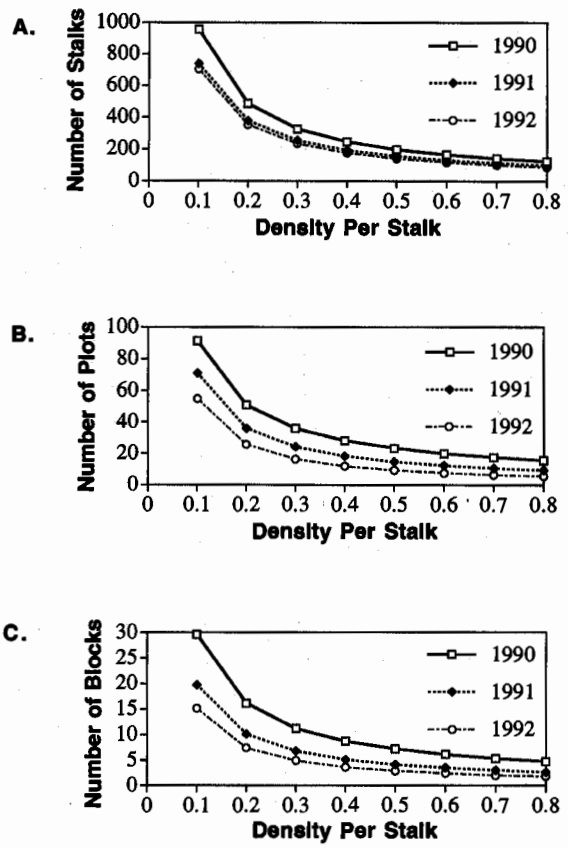

Fig. 4. Estimated number of stalks required to estimate *E. loftini* small, medium, and large larvae and pupae for sample unit sizes of 1,000 stalks per field, 10 stalks per plot, and 40 stalks per block using $\alpha = 0.2$, $D\bar{x} = 0.2$.

in Texas, including *Solenopsis geminata* (F.), *Crematogaster clara* Mayr, and *Pheidole* spp. (Breene et al. 1993).

Sugarcane fields were classified into 3 types for our study because of the importance of hot water-treated plant cane as a recommended practice in the management of ratoon-stunting disease and indirectly a mortality factor for Mexican rice borer. As expected, ratoon fields had higher larval densities, but plant cane and hot water-treated fields contained similar numbers. We believe that if more hot water-treated fields were sampled, a difference in larval density between plant cane fields and hot water-treated fields would have been detected. Ratoon fields contain populations from the previous season that survive the harvest process in the underground billets; billets used for planting plant cane fields can also be infested by Mexican rice borer larvae that will finish development and emerge as adults (R.L.M., unpublished data). Hot water-treated fields generally contained a higher average of younger larvae than other fields, an indication that infestations in hot water-treated fields come from moths migrating into the fields rather than localized infestation from planted billets.

More younger larvae were found in the earlier part of the season (January through March) than in the later part (April through August). The explanation for this result would be somewhat obvious for a stalkborer species that is found in temperate climates (seasonal larval development) but is not as obvious with a subtropical species such as *E. loftini*. Seasonal sampling suggests that this insect is multivoltine, and any life stage can be found during any part of the year (Johnson 1985, van Leerdam et al. 1986, Meagher et al. 1994), so young larvae should be present during the entire season. It is possible that some younger larvae were not collected during late-season sampling because they are more difficult to locate when plant biomass is large.

Taylor coefficients suggested small larvae to be more aggregated than other sized larvae in 1990 but large larvae and pupae to be more aggregated in 1992. Also, coefficients estimated from larvae collected in 1990 plant or ratoon fields were greater than in the hot water-treated field. Variance to mean ratios calculated under different larval density conditions removed the effect of larval density when comparing aggregation. This is important because calculations based on results taken in the hot water-treated field in 1990 were comparatively higher than in any other field–year combination. As with Taylor coefficients, the calculated variance to mean ratios for small larvae in 1990 had aggregated spatial patterns whereas medium and large larvae had random spatial patterns; the reverse was true for 1992.

Results of the seasonal density and nested ANOVAs suggest that important variables for a Mexican rice borer larval sampling plan are stalks and fields. Within-field strata such as blocks, tiers, or plots, as described, did not contribute significantly to sampling variation. This was unexpected, especially for the hot water-treated fields, because we believed the infestation of these fields was initiated from the border areas toward the center. The results suggest that random sampling of ≈1,000 stalks within fields of 6–25 ha is required to estimate larval density when it is near a mean of 0.1 larva per stalk, which is approximately equal to the *E. loftini* action threshold (Johnson et al. 1982). At higher or lower densities, considerably fewer stalks would need to be sampled if the goal were to classify the population as being above or below an action threshold (Wilson 1985). Sampling of young larvae is important because this is the target life stage for action thresholds under insecticidal management tactics (Johnson et al. 1982, Meagher et al. 1994). However, the large amount of sugarcane biomass makes sampling difficult and constrains the desired distribution of insecticide spray within the plant canopy, making insecticidal management as an economically viable practice questionable (Meagher et al. 1994).

Future research should concentrate on correlating larval densities with larval injury, because resulting larval damage is the important variable to growers and the industry. Previous studies with Mexican rice borer have documented percentage bored internodes (ratio of number of internodes with tunnels to total internodes present) as an indirect measure of larval density (Johnson 1985, Meagher et al. 1994). However, currently there is no known relationship between larvae per stalk and percentage bored internodes. Growers and industry agriculturalists speculate there is an economic effect of feeding by *E. loftini* only after a damage level of 10% bored internodes is attained (N. Rozeff, Rio Grande Valley Sugar Growers, Incorporated, personal communication). A relationship between population density (larval sampling or a related population parameter such as male captures in pheromone traps) and larval injury–damage could establish an improved early warning of pest population increase and provide a better indication of potential losses.

Acknowledgments

Special thanks are given to S. Alvarez, M. Barrosa, J. Huerta, H. Perez, and R. Saldaña, Texas Agricultural Experiment Station for their valuable assistance in the field, and to the sugarcane growers of Rio Grande Valley Sugar Growers, Incorporated. Thanks go also to R. Moon (University of Minnesota) and D. Riley (Texas Agricultural Experiment Station) for review of an early manuscript. This research was performed under Hatch Project 6796, with additional support from the Rio Grande Valley Sugar Growers, Incorporated, and the state of Texas under the Texas Agricultural Experiment Station Expanded Research Program.

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Received for publication 11 April 1995; accepted 2 October 1995.