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Influence of corn, *Zea mays*, phenological stages in *Diatraea saccharalis* F. (Lep. Crambidae) oviposition

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Abstract: *Diatraea saccharalis* F. is a major pest to maize, sorghum and sugarcane crops in Latin America and the most damaging insect pest of maize in Argentina. Female moths lay their eggs on lower and upper sides of leaves and sheaths of corn plants. Oviposition behaviour of different species of corn stem borers is influenced by host plant phenology in different ways. Host pubescence affects oviposition behaviour in different species of caterpillars. Oviposition preferences for phenological stages, leaf surface and vertical distribution in corn plants were investigated. Corn plots of 120 m^2 were sown with a Pioneer pubescent cultivar in six planting dates, each one replicated three times according to a completely randomized design. Throughout the season 14 samplings were carried out, during which plants were randomly chosen within each of the six treatments and exhaustively examined in order to count the number of egg masses and their location in the leaf surface and in the vertical structure of the plant. *Diatraea saccharalis* preferred laying their eggs on the lower sides of leaves located in the middle stratum of corn plants whose phenological stages were older than V10. Our study suggests that attention should be paid to the influence of crop phenological stage in the location of egg masses in futures studies, as oviposition preference can change through plant maturation.

Key words: *Diatraea saccharalis*, maize, oviposition preferences, plant–insect relationship

1 Introduction

Oviposition is particularly crucial in Lepidoptera because the hatching larvae are often relatively immobile and thus depend on the choice of food plant by the adult female (Renwick and Chew, 1994). A strong relationship between oviposition or settling choices of individual phytophagous insects and the subsequent performance of their larvae can significantly influence distribution, density and population dynamics (Rausher, 1979; Thompson and Pellmyr, 1991).

Diatraea saccharalis Fabricius (Lep. Crambidae) is the most damaging insect pest of maize in Argentina. Temporal occurrence of *D. saccharalis* is characterized by the succession of three to four adult flights throughout the growing season. Female moths lay their eggs on the under and upper sides of leaves and sheaths of corn plants. Two or 3 days after egg hatching, second instar larvae bore into the stalk where they stay until pupal development (Hensley, 1971; Leiva and Iannone, 1994).

The growth of a corn plant progresses through a series of vegetative stages characterized by the successive addition of up to 20 single primary leaves (V1–V20). Vegetative stages finish at tasselling (VT). Reproductive stages begin in silking (R1) and finish at physiological maturity (R5). Intermediate reproductive stages (R2–R4) are characterized by grain morphology (Ritchie et al., 1986).

Different authors demonstrated how phenological stage (Van Rensburg et al., 1987; Spangler and Calvin, 2000) and pubescence of the host (Ramaswamy, 1988; Sosa, 1988; Greco et al., 1998) affect oviposition behaviour in many lepidopterous species.

Spangler and Calvin (2000) observed that adult females of the European corn borer, *Ostrinia nubilalis* Hübner (Lep. Crambidae) prefer plants in reproductive to vegetative stages as oviposition sites. However, female moths of the maize stalk borer *Busseola fusca* Fuller (Lep. Noctuidae) prefer to lay their eggs on plants in vegetative stages (Van Rensburg et al., 1987).

Host pubescence affects oviposition behaviour in different species of Lepidoptera, stimulating (*Heliothis virescens* F., *Heliothis zea* Boddie) or deterring (*Chillo partellus* Swinhoe, *Diatraea grandiosella* Dyar, *D. saccharalis*) females to lay their eggs (Ramaswamy, 1988; Sosa, 1988).

Sosa (1988) observed that in glabrous cultivars of sugarcane the density of *D. saccharalis* egg masses per plant was higher than in pubescent cultivars. The distribution of the eggs between both sides of the leaf surface did not differ in glabrous cultivars but in pubescent cultivars abaxial leaf surface received more eggs. Instead, Greco et al. (1998) did not observe differences in the total number of egg masses per plant and number of eggs per plant laid by *D. saccharalis* between high pubescence and low pubescence cultivars of corn. However, higher densities of eggs and egg masses were found on the under side of leaves of high pubescence cultivars, while no differences were found between leaf sides in low pubescence cultivars.

In these previous studies, the influence of the host phenology in the oviposition preferences of females was not considered. The aim of this work was the identification of oviposition preferences in *D. saccharalis* for phenological stages, leaf surface and vertical distribution in corn plants.

2 Material and methods

The study was conducted at the Agricultural Experimental Station of the National Institute of Agricultural Technology (INTA) located in Manfredi, Córdoba province, Argentina (31°49′12″S and 63°46′00″W). The essay consisted in six corn plantings, which were sown on 30 October, 12 and 27 November, 11 and 23 December 1998, and 7 January 1999. A Pionner pubescent cultivar of maize was used. The purpose of this design was to provide *D. saccharalis* females with different phenological stages of corn simultaneously available for oviposition throughout the season.

The experiment was laid out according to a completely randomized design with the six planting dates as treatments and three replicates. The size of each plot was 12 × 10 m. In order to minimize the effect of nearby spray applications, a 10-m width border was sown on 11 October 1998 around the experimental plots.

Throughout the season 14 samplings were carried out. For each sampling session six to 12 sample units were randomly chosen within each of the six treatments. Each sample unit consisted in the exhaustive examination in the field of 10 consecutive plants in order to count the number of egg masses and their location on the leaf surface (abaxial, i.e. under side; adaxial, i.e. upper side; sheaths) and along the vertical structure of the plant (leaf number according to Ritchie et al., 1986 classification). A total of 3540 plants were sampled (Sosa, 1988; Van Rensburg et al., 1988; Greco, 1995).

Only white, newly laid eggs were recorded in order to avoid counting the same egg mass more than once between samplings and to be sure which phenological stage of corn plant was at the moment of oviposition (Prola et al., 2000).

Phenological stages were recorded on three plants per plot, once or twice a week, according to the scale proposed by Ritchie et al. (1986), and leaves were numbered accordingly.

Three different statistical approaches were employed to analyse the data. First, contingency tables were used to test whether or not the distribution pattern of total egg masses on the leaf surface of plants of two broad physiological stages followed a random pattern. Secondly, the mean number of egg mass per plant of the six treatments were compared for each of the 14 sampling sessions, previously transformed to $\sqrt{x + 0.5}$ using Student's *t*-test and analysis of variance. The variable was transformed because Kolmogorov–Smirnov analysis showed that the distribution of the original variable was not normal (Sokal and Rohlf, 1995). Finally, Friedman nonparametric test was used to evaluate whether or not females presented a hierarchy of preference for a particular corn phenological stage throughout the season. Plants were sorted into six categories according to their physiological development: VE–V5, V6–V9, V10–V14, V14–VT, R1–R2 and R3–R4. Mean egg mass per plant present on each of these categories was compared and Dunn *post-hoc* test was performed (Siegel, 1984; Zar, 1996).

3 Results

In phenological stages of corn plants up to 13 leaves (V13), the total number of egg masses found on adaxial (upper) and abaxial (lower) surfaces did not differ from a random distribution ($\chi^2_{(1d.f.)} = 1.33$; P > 0.05). However, in plants with phenological stages older than V14, the abaxial surface was preferred by females for egg laying ($\chi^2_{(1d.f.)} = 34.04$; P < 0.05) (fig. 1).

Leaves of the middle third received most of the egg batches, regardless of the phenological stage. In plants with phenological stage V6–V13, 97% of the egg masses were concentrated between leaves 5 and 14, whereas in plants whose phenological stages were V14–VT and R1–R4, 93% of the egg masses were located between leaves 9 and 16 (fig. 2).

Only in five sampling sessions (7 December 1997, 2, 6 and 28 January and 8 February 1998) did the mean number of egg mass per plant significantly differ between phenological stages (P < 0.05, F = 12.88, 6.54, 8.90, 3.56 and 3.73, respectively). At these

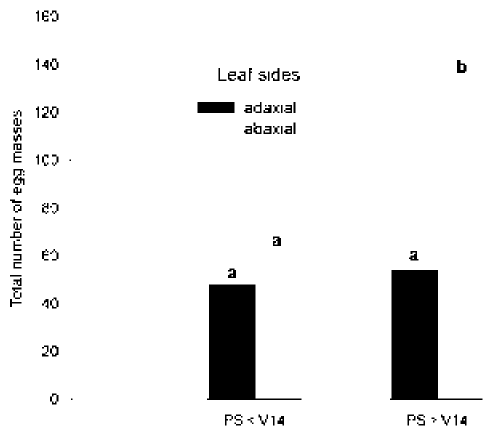

Fig. 1. *Distribution of* Diatraea saccharalis *egg masses on both leaf sides of corn plants in two broad phenological stages of the crop. Different letters represent significant differences between leaf sides according to* χ^2 *test (P < 0.05)*

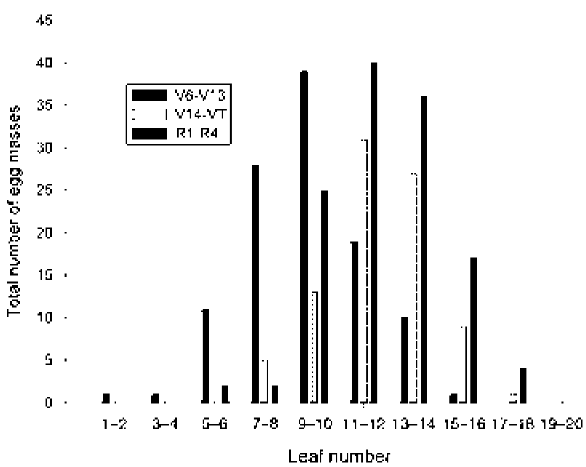

Fig. 2. *Within-plant distribution of* Diatraea saccharalis *egg masses on corn with different phenological stages (V6–V13, V14–VT and R1–R4).*

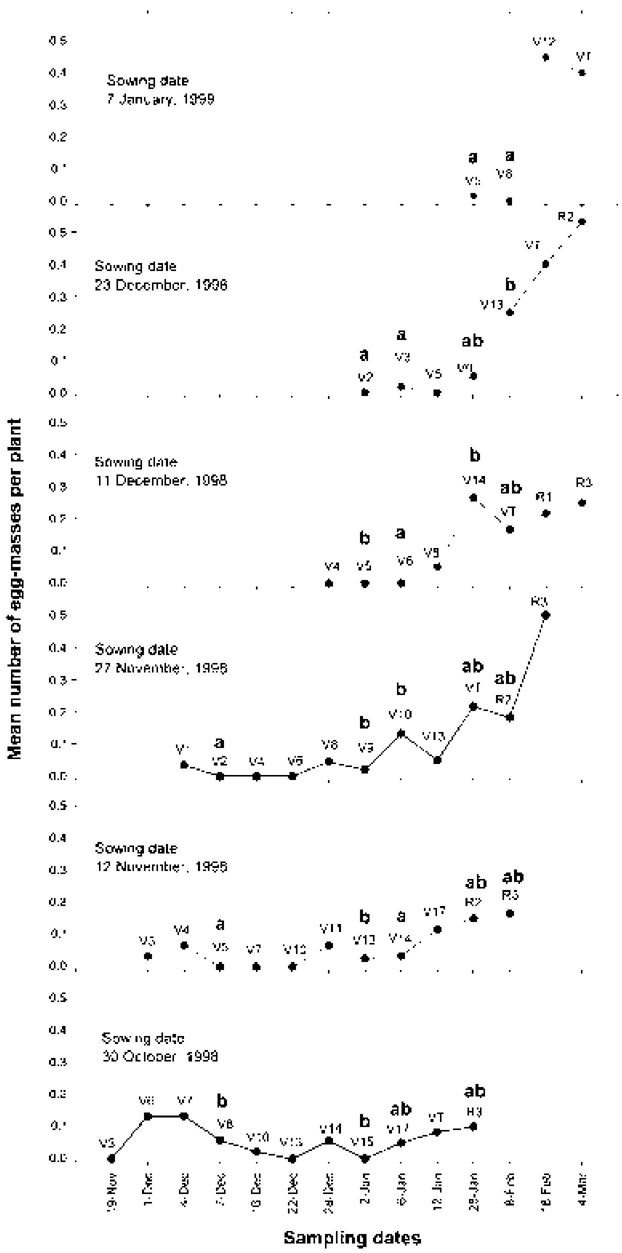

Fig. 3. *Mean number of* Diatraea saccharalis *egg masses per plant in six plantings during 1998/99 growing season. An* ANOVA *was performed in each of the 12 sampling sessions (on 1 December, a t-test was performed). Only five sampling dates showed significant differences between means (dotted lines). Different letters in each dotted line show significant differences between means (Tukey* post-hoc *test, P < 0.05). Phenological stages are indicated (see text for further information)*

samplings, plants in phenological stages between V10 and V14 received the highest density of egg batches. The mean number of egg masses per plant was low or null in plants at phenological stages earlier than V9 (fig. 3).

The fact that females showed, throughout the season, a hierarchy of preference to lay their eggs on plants older than V10 was evidenced by the Friedman test ($\chi^2_{r(2d.f.)} = 13.6$; P < 0.05). However, no single

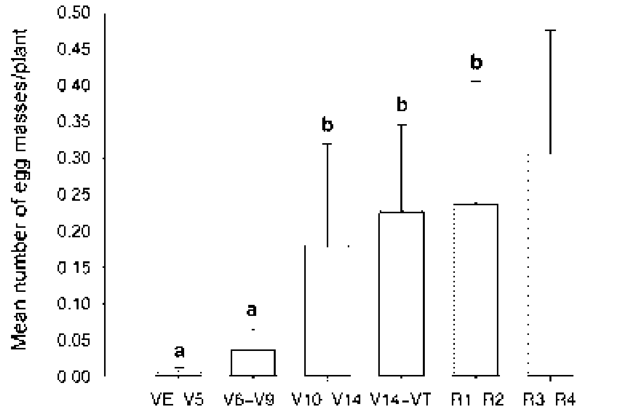

Fig. 4. *Mean number of* Diatraea saccharalis *egg masses per plant in different corn phenological stages. Different letters represent significant differences between phenological stages (Dunn* post-hoc *test, P < 0.05)*

class stage between V10 and R2 was preferred over any other. R3–R4 phenological stage could not be considered for Friedman test because this data set was unbalanced (fig. 4) (Siegel, 1984; Zar, 1996).

4 Discussion

In the present study we observed that females of *D. saccharalis* preferred laying their eggs on the lower sides of the leaves located in the middle stratum of corn plants whose phenological stages were older than V10. Sosa (1988) and Greco et al. (1998) found that in pubescent cultivars of sugarcane and corn, respectively, the greatest proportion of egg masses of *D. saccharalis* was found on the leaf underside (less hairy) in plants of advanced phenological stages. Instead, the similarity of egg mass incidence observed between under and upper sides of the leaves in plants of phenological stages earlier than V14 found in our work (where a pubescent cultivar was used) coincides with the results obtained by these authors for glabrous or low pubescence cultivars. The uniform distribution of egg masses between both leaf sides on plants through the first phenological stages (up to V14) could be associated to the fact that in young plants leaf hairs are not completely developed, and hence do not interfere with the oviposition behaviour of the females.

Sosa (1988) observed that pubescence in sugarcane leaves delay first instar larval movement, which should increase larval mortality by increasing exposure to adverse environmental factors and biological control organisms. He suggested that the use of pubescent clones might decrease the damage caused by sugarcane borers and possibly other pests.

However, Greco et al. (1998) found that egg mortality of *D. saccharalis* on the under side of leaves of low pubescence cultivars of corn was higher than on high pubescence cultivars because a higher percentage of parasitized eggs by *Trichogramma exiguum* Pinto, Platner and Oatman (Hymenoptera) was observed.

Our study suggests that attention should be paid to the influence of crop phenological stage in the location of egg masses in futures studies, as oviposition preference can change with the phenological stage of the crop.

Flynn et al. (1984) observed a definite gradient in leafsheath tightness with the upper leafsheaths of younger plants having the greatest and the lower leafsheaths of older plants the least appression. In reproductive phenological stages, ear leafsheaths present little impediment to larval entry because of openings provided by the developing ear shoots. They found a strong relationship between leafsheath appression to the stalk and larval survival. In our work, we observed that egg masses were concentrated in leafsheaths 13 and 12, harbouring primary and secondary ears, respectively, where appression is the lowest. The fact that the majority of the egg masses recorded in our study were found on the lower side (less hairy) of leaves located in the middle stratum of advanced phenological stage plants, where larval survival should be maximum (in agreement with Sosa, 1988 and Flynn et al., 1984), suggests a selection pressure favouring such oviposition behaviour in *D. saccharalis*.

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