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Development and reproduction of *Neoleucinodes elegantalis* (Lepidoptera: Crambidae) on tomato (*Solanum licopercum*) cultivars

Desarrollo y reproducción de *Neoleucinodes elegantalis* (Lepidoptera: Crambidae) en cultivares de tomate (*Solanum licopercum*)

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Abstract: The small tomato borer *Neoleucinodes elegantalis* (Lepidoptera: Crambidae) is one of the most representative pests of Solanaceae in Brazil and other South American countries. Due to the restricted knowledge about the influence of tomato cultivars on the biology of the species, the development and reproduction of *N. elegantalis* was evaluated on three tomato cultivars. In the hybrid Giuliana the development from oviposition to adult emergence was longest (56.2 days) in comparison to the cultivar Santa Clara (50.2 days) and the hybrid Paronset (50.7 days). Pupal weight was significantly lower in Giuliana. The mean fecundity of females fed on Giuliana during the larval stage was 88.6 eggs per female, 82.3 in Paronset and 37.0 on Santa Clara, but egg viability was higher for females fed during the larval stage on Santa Clara (79.0%). The hybrid tomato Giuliana had the highest impact on the development and reproduction of *N. elegantalis.* These findings can be useful as a plant resistance factor in the field to reduce the number of generations of the pest and to favour the action of natural enemies by decreasing the fitness of the pest.

Key words: Small tomato borer. Biology. Solanaceae.

Resumen: El pequeño barrenador del tomate *Neoleucinodes elegantalis* (Lepidoptera: Crambidae) es una de las plagas más destructivas de las solanáceas en Brasil y otros países suramericanos. Debido al conocimiento limitado acerca de la influencia de los cultivares de tomate sobre la biología, se evaluó el desarrollo y la reproducción de *N. elegantalis* en tres cultivares de tomate. En el híbrido Giuliana el desarrollo de la oviposición hasta la emergencia del adulto fue más larga (56,2 días) en comparación con el cultivar Santa Clara (50,2 días) y el híbrido Paronset (50,7 días). El peso pupal fue significativamente menor en Giuliana. La fecundidad media de hembras alimentadas con Giuliana, Paronset y Santa Clara durante la etapa larval fue de 88,6; 82,3 y 37,0 huevos por hembra, respectivamente. Sin embargo, la viabilidad de los huevos fue mayor para las hembras alimentadas durante la etapa larval en Santa Clara (79,0%). El híbrido de tomate Giuliana tuvo el mayor impacto en el desarrollo y la reproducción de *N. elegantalis*. Estos hallazgos pueden ser útiles como un factor de resistencia de la planta en el campo para reducir el número de generaciones de la plaga y para favorecer la acción de los enemigos naturales por la disminución del "fitness" de la plaga.

Palabras clave: Perforador del fruto del tomate. Biología. Solanaceae.

Introduction

Tomato crops (*Solanum licopercum* L.) have a great economic and social importance worldwide (Jordão and Nakano 2002). The small tomato borer *Neoleucinodes elegantalis* (Guenée) (Lepidoptera: Crambidae) is a pest of economic importance due to the damage caused in the fruits and thus reducing the commercial value of the crop (Leiderman and Sauer 1953; Picanço *et al.* 2007). The distribution of *N. elegantalis* is strictly Neotropical including both Central and South America (Capps 1948).

The damage caused by *N. elegantalis* is direct, a single larva is sufficient to depreciate and render the fruit unsuitable for consumption (Toledo 1948). Yield losses are estimated at as much as 50% (Gallo *et al.* 2002), 79% (Miranda *et al.* 2005) and 90% (Carneiro *et al.* 1998). According to Blackmer *et al.* (2001) under field conditions the females lay their eggs preferably beneath the sepals of small (23.0 mm diameter) green fruits and soon after hatching the larva penetrates the fruit and causes the irreversible damage.

The knowledge on the biology of the small tomato borer is scarce; Marcano (1991a) evaluated the development time and reproduction of *N. elegantalis* on tomato fruits of the cultivar Rio Grande. Other studies include the ecology and behavior of *N. elegantalis* (Salas *et al.* 1991) and the role of pheromones (Badji *et al.* 2003). Blackmer *et al.* (2001) and Barbosa *et al.* (2010) evaluated the preference for oviposition of *N. elegantalis* on tomato and Jaffe *et al.* (2007) investigated the role of chemical stimulus on the sex attraction of *N. elegantalis*. Benvenga *et al.* (2010) conducted field samplings of the small tomato borer for decision making on the control in staked tomato plants.

Apart from tomato, *N. elegantalis* is able to develop and reproduce in other cultivated Solanaceae, green peppers, eggplant and scarlet eggplant (Toledo 1948, Zucchi *et al.* 1993). On scarlet eggplant Picanço *et al.* (1997) found all stages of *N. elegantalis* and described the tunneling caused by the larvae in the fruits. Uncultivated solanaceous plants also host the pest (Toledo 1948, Zucchi *et al.* 1993).

Due to the limited efficiency of chemical control measures against the damage of boring insects such as *N. elegantalis*, research on cultivar performance is a sound alternative to reduce the larval damage on tomato yield. This work was conducted to evaluate the effect of three tomato cultivars on the growth, reproduction and survival of *N. elegantalis*.

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Materials and methods

A colony of *N. elegantalis* was established in laboratory from larvae collected on tomato hybrid Alambra crops in Southern Paraná state (25°19'S 49°18'W). Larvae were reared on tomato fruits of the group Salad. Field collected larvae were introduced into the colony every week in order to maintain the genetic diversity of the population. One cultivar (Santa Clara) and two hybrids (Giuliana and Paronset) were employed to compare the development cycle, reproduction and longevity of *N. elegantalis*. These materials were chosen for being commonly employed by local tomato growers and their high commercial value. Tomato plants were grown in a greenhouse in 5 L pots. Green tomato fruits, 23 to 40 mm in diameter from each cultivar were used in the experiment. For surface disinfection, the fruits were immersed in water + 1% sodium hypochlorite for five minutes, rinsed under running water and dried with paper towels. Three neonate larvae were incubated in each fruit totaling 99 larvae per tomato cultivar. The experiment was conducted in a climatic chamber at a constant temperature of 20 ± 1 ºC, relative humidity of 60 ± 10% and photoperiod of 12:12 h (light: dark). At the end of the larval period, the insects left the tomatoes to pupate outside the fruits. Pupae were kept in 7x4 cm polyethylene vials and after 72 hours were weighed and checked for the presence of deformities. Sex determination was made after adult emergence according to Muñoz *et al.* (1991) and Carneiro *et al.* (1998).

The biological parameters evaluated were development time, larval and pupal viability, pupal weight and sex-ratio (females / males + females). To evaluate the influence of the cultivar consumed in the larval stage on adult performance, twenty pairs were formed from each treatment. Each pair was kept in polyethylene tubes 20 cm high x 10 cm diameter closed at the bottom and on the top with a polyethylene cover. Tomato leaves of each cultivar were placed inside the cages as stimulants for oviposition. The adults were fed with 10% honey diluted in water, provided in a cotton ball soaked in the solution. Food renewal and egg collection were carried out daily. The eggs were separated according to the female and kept in moistured paper towel to assess the incubation period and viability.

The effect of the cultivars consumed by larvae on the adult stage of *N. elegantalis* was evaluated through longevity, length of pre-oviposition and oviposition periods, number of eggs laid and their viability.

The experimental design was completely randomized, the condition of normality of the data was submitted to Shapiro-Wilks test. Results were subjected to analysis of variance and the means were compared by Tukey's test or nonparametric Kruskal-Wallis test ($P < 0.05$). Data were analyzed with Statistica 7.0 software (Statsoft 2004).

Results and discussion

The incubation period of *N. elegantalis* at 20 ± 1 °C lasted from 7.3 to 7.4 days, did not differ significantly among the cultivars (Table 1). There are no other references on the development of *N. elegantalis* other than the work of Marcano (1991a) which describes an incubation period of 7.1 days at 20 ºC.

Larval survival was not significantly different among the treatments ($P = 0.162$) (Table 1). The larval stage was significantly longer when the larvae were reared on Giuliana fruits (29.5 days) in comparison to Paronset (26.1 days) and Santa Clara (25.8 days) (Table 1). There are no data in the literature comparing the influence of different tomato cultivars on the biological traits of *N. elegantalis*. Marcano (1991a, 1991b) estimated the development of *N. elegantalis* on tomato and eggplant; when feeding on tomato, the larval period lasted 22.7 days compared to 31.4 days for larvae reared on eggplant fruits. According to Muñoz (1991) newly hatched larvae move by means of a silk thread until a point chosen for penetration through the epicarp and then feed on the mesocarp and later reach the endocarp where the rate of feeding is higher. In the endocarp the larvae tunnel galleries and feed on the placenta and seeds provoking most of the damage seen on the fruits.

Between the end of the larval stage and the beginning of the pupal phase *N. elegantalis* remained over five days in the pre-pupal stage (Table 1). No deformed pupae were recorded among the three cultivars. Likewise the larval development, adults took longer to emerge from the pupae proceeding from Giuliana in comparison to the other cultivars (Table 2). These values are similar to the ones reported by Marcano (1991a, 1991b) for eggplant and tomato fruits for the pupal stage of *N. elegantalis* (13.9 days). Paredes *et al.* (2010) also recorded a pupal period of 13 days for the species.

The sex-ratio of *N. elegantalis* was 0.4 in all cultivars. A predominance of males was also recorded by Salas (1992), Carneiro *et al.* (1998) and Jaffe *et al.* (2007). Fernández and Salas (1985) found in 688 sexed pupae 370 females and 318 males.

During the reproductive period, no statistical differences were observed among the treatments in relation to the pre-oviposition ($P = 0.510$) and oviposition ($P = 0.299$) periods, oviposition rate ($P = 0.495$) and egg viability ($P = 0.813$). Mating success of *N. elegantalis* is low in laboratory; only 10 out of 20 pairs evaluated laid eggs on Giuliana, 11 on Santa Clara and 13 on Paronset (Table 2). According to Jaffe

Table 1. Mean development time (± SE) of the immature stages and pupal weight (milligrams) of *N. elegantalis* in three tomato cultivars. 20 ± 1 ºC, relative humidity of 60 ± 10% and photoperiod of 12:12 hours (Light: Dark).

		Development Time (days)					Pupal
Treatment	N	Incubation^{NS}	Larva	Pre-Pupa^{NS}	Pupa	Total	weight (mg)
Santa Clara	64	7.4 ± 0.14	25.8 ± 0.41b	5.1 ± 0.04	11.9 ± 0.15b	50.2 ± 0.43b	39 ± 1.19a
Paronset	55	7.3 ± 0.10	26.1 ± 0.40b	5.2 ± 0.05	12.1 ± 0.13b	50.7 ± 0.36b	41 ± 1.24a
Giuliana	51	7.4 ± 0.12	29.5 ± 0.38a	5.1 ± 0.10	14.2 ± 0.21a	56.2 ± 0.40a	36 ± 1.02b
P value	0.162	0.579	< 0.001	0.874	< 0.001	< 0.001	0.043

* P values < 0.05 differ according to test non-parametric Kruskal-Wallis. n - Survival larval stage; NS – not significant.

Table 2. Mean (± SE) of the duration of pre-oviposition and oviposition in days, fecundity and fertility of *N. elegantalis* in three varieties of tomatoes stored at 20 ± 1 ºC and relative humidity of 60 ± 10% and photoperiod of 12:12 hours (Light:Dark).

Treatment	Couple (n)	Pre-oviposition	Oviposition	Fecundity	Fertility
Santa Clara	11	6.5 ± 1.11	3.9 ± 0.86	37.0 ± 14.13	79.0 ± 8.38
Paronset	13	6.5 ± 0.87	5.8 ± 0.84	82.3 ± 16.68	67.6 ± 10.83
Giuliana	10	5.8 ± 1.22	4.9 ± 1.08	88.6 ± 33.97	70.8 ± 11.88
p value	---	0.510	0.299	0.495	0.813

* P values > 0.05 differ according to test non-parametric Kruskal-Wallis.

et al. (2007) *N. elegantalis* copulates monogamously, more vigorous males have a faster answer to the sexual pheromone released by females, thus functioning as a process of natural selection. This behavior explains the low number of females ovipositing, since the couples were randomly formed and no option were given for the choice of partners by the females.

The pre-oviposition period (Table 2) was more than twice longer than the values reported by Marcano (1991a) and Muñoz *et al.* (1991). These authors used tomato leaves as stimulants for oviposition instead of fruits as we did and the longer pre-oviposition period may be due to the less stimulant effect of tomato fruits in comparison to leaves. The oviposition period in *N. elegantalis* lasted less than a week for females emerged from the three cultivars (Table 2), similar to the value found by Carneiro *et al.* (1998), but longer than the two days of oviposition described by Marcano (1991a).

The mean number of eggs laid by each female was more than twice as high in Giuliana and Paronset than in Santa Clara (Table 2). However due to the wide amplitude in the number of eggs laid per female, no statistical difference was detected among the treatments, indicating that the cultivar had no effect on the fertility of *N. elegantalis*. Marcano (1991a) obtained a mean of 52.3 eggs per female, also with a large variation in the number of eggs per female among the replicates (19 to 77 eggs/female).

No differences were recorded on egg viability among treatments, which ranged from 67.6% in females reared in the larval stage on Paronset to 79.0% in Santa Clara (Table 2). Similar values were described by Fernández and Salas (1985) in tomatoes, while Muñoz *et al.* (1991) recorded 98.0% of egg viability when the larvae were reared on nightshade fruits, a perennial solanaceaus species, *Solanum quitoense* grown in Northwestern South America.

The longevity of males and females was longer than 15 days, and no differences were found both among cultivars and between the sexes (Table 3). These values are more than twice the longevity recorded by Marcano (1991a, 1991b) for *N. elegantalis* kept at the same temperature and reared during the larval stage on eggplant and tomato fruit.

Table 3. Mean (± SE) in days for the longevity of females, males, longevity and total time from egg to adult of *N. elegantalis* in three varieties of tomatoes stored at 20 ± 1 ºC and relative humidity of 60 ± 10% and photoperiod of 12:12 hours (Light: Dark).

Treatment	Longevity	
	Female	Male
Santa Clara	17.7 ± 1.07a	17.7 ± 1.02a
Paronset	17.7 ± 1.17a	17.6 ± 1.45a
Giuliana	16.3 ± 0.98a	15.2 ± 0.90a

* Means followed by the same letter in the column do not differ according to the Tukey test at 5% probability.

Development from egg hatching to adult emergence resulted significantly longer when larvae were reared on the cultivar Giuliana (56.2 days) in comparison to Paronset and Santa Clara (50.2 and 50.7 days, respectively). This difference may be due to the effect of antibiosis caused by cultivar, because according to Smith (2005) antibiotic effects may be expressed in a moderate way, increasing the duration of development and reducing weight or decreasing the adult life span. These results differ from those found by Marcano (1991a), which reported the development cycle in 43.7 days using cultivar Rio Grande. In the field the cultivar Santa Clara is considered as susceptible to the major insect pests and diseases of tomato (Bettiol *et al.* 2004) and thus is employed in laboratory tests as a pattern for susceptibility. According to the companies responsible for the hybrids (Sakata Seeds for Giuliana and Syngenta Seeds for Paronset), these hybrids possess genetic resistance to numerous diseases, but little is known about their tolerance to insect attack.

Previous results showed significant differences in the performance of *N. elegantalis* developing on different tomato cultivars (Lara *et al*. 1980, Moreira *et al*. 1985, Lyra Netto and Lima 1998). According to Viáfara *et al.* (1999) increased susceptibility of *N. elegantalis* to industrial tomato cultivars, is assigned to the higher content of soluble solids, which promote the feeding habits of the larvae.

Conclusion

The increased development time and the lower pupal weight of the small tomato borer reared on the hybrid Giuliana may reduce larval fitness, and thus varietal control can function as a strategy in programs of integrated pest management of *N. elegantalis* in tomato.

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