

New records of predation on eggs of Bemisia tabaci (Hemiptera: Aleyrodidae) by Chrysopodes (Chrysopodes) lineafrons (Neuroptera: Chrysopidae) in Northwestern Argentina

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Bemisia tabaci has become a major economic importance pest, affecting several crops worldwide. Among their natural enemies, species of Chrysopidae family, with larvae predators of different pests, are a very effective biological control agent. The developmental time and survival of the immature stages of Chrysopodes (Chrysopodes) lineafrons, and the longevity and oviposition of adults fed with eggs of B. tabaci was determined. C. (C.) lineafrons adults were collected in tomato crops in Lules department, Tucumán province. To determine the developmental duration of each instar, and larvae survival, 90 eggs of C. (C.) lineafrons were randomly selected, of which only 71 eggs hatched; of these, 34 larvae were fed with B. tabaci eggs and 37 with Sitotroga cerealella eggs, used as control. Oviposition and longevity of adults fed with the two preys were recorded. C. (C.) lineafrons larvae consumed an average of 127.04 B. tabaci eggs and 44 S. cerealella eggs per day. Mean developmental time of C. (C.) lineafrons fed with B. tabaci eggs was 45 days; while for those fed with S. cerealella eggs it was 35 days. Immature stages survival, number of eggs per adults and longevity were higher when C. (C.) lineafrons were fed with S. cerealella eggs than with B. tabaci eggs. C. (C.) lineafrons proved to be an efficient predator, thus representing an excellent tool for the biological control of B. tabaci in tomato crops.

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ABSTRACT

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Bemisia tabaci has become a major economic importance pest, affecting several crops 25 worldwide. Among their natural enemies, species of Chrysopidae family, with larvae predators 26 of different pests, are a very effective biological control agent. The developmental time and 27 survival of the immature stages of Chrysopodes (Chrysopodes) lineafrons, and the longevity and 28 29 oviposition of adults fed with eggs of B. tabaci was determined. C. (C.) lineafrons adults were collected in tomato crops in Lules department, Tucumán province. To determine the 30 developmental duration of each instar, and larvae survival, 90 eggs of C. (C.) lineafrons were 31 32 randomly selected, of which only 71 eggs hatched; of these, 34 larvae were fed with B. tabaci eggs and 37 with Sitotroga cerealella eggs, used as control. Oviposition and longevity of adults 33 fed with the two preys were recorded. C. (C.) lineafrons larvae consumed an average of 127.04 34 B. tabaci eggs and 44 S. cerealella eggs per day. Mean developmental time of C. (C.) lineafrons 35 fed with B. tabaci eggs was 45 days; while for those fed with S. cerealella eggs it was 35 days. 36 37 Immature stages survival, number of eggs per adults and longevity were higher when C. (C.) lineafrons were fed with S. cerealella eggs than with B. tabaci eggs. C. (C.) lineafrons proved to 38 be an efficient predator, thus representing an excellent tool for the biological control of B. tabaci 39 40 in tomato crops. 41

- 42 Subjects Entomology, Ecology
- 43 **Keywords** Chrysopodes, Developmental time, Survival, Longevity, Oviposition, Ingest capacity.

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INTRODUCCIÓN

48	The whitefly Bemisia tabaci Gennadius (Hemiptera: Aleyrodidae) is a serious pest of several
49	annual, ornamental and industrial crops, fruit plantations and weeds worldwide (Byrne et al.,
50	1990; Brown et al., 1995; Viscarret, 2000; López-Ávila, 2005). It causes direct damage through
51	sucking sap and excreting sugary substances, which produces the growth of sooty mold, reducing
52	photosynthetic capacity of the plant; and indirect damage through viruses and bacteria
53	transmission (Berlinger, 1986; Viscarret, 2000). Bemisia tabaci has caused significant losses in
54	America since 1981, reducing crop productivity of tomato, sweet pepper, beans and textiles
55	(Brown, 1993). In Argentina, the first record of <i>B. tabaci</i> arises from specimens found in an
56	unspecified host plant in Tucumán province (Viscarret, 2000). Subsequently, its presence has
57	been reported in greenhouses and field crops such as cotton, tobacco, citrus, sugar cane, soybean,
58	forestry and horticultural crops of Solanaceae, Cucurvitaceae, Cruciferaceae and Compositae
59	families (Polack, 2005).
60	Currently, the most used control method against B. tabaci is chemical control; however,
61	alternative methods based on biological control with natural enemies of the pest can reduce its
62	density (Reguilón et al., 2011), as well as environmental impact, and improve product quality
63	(López et al., 1999). Among the natural enemies of this species, the genus Chrysopodes Navás
64	(Neuroptera: Chrysopidae) has been mentioned, with a cosmopolitan distribution and with about
65	40 species distributed in two subgenera: <i>Chrysopodes</i> s. str. and <i>Neosuarius</i> Adams & Penny
66	(Adams & Penny, 1987). In Argentina, Chrysopodes (Chrysopodes) lineafrons Adams & Penny,
67	Chrysopodes (Chrysopodes) polygonicus Adams & Penny, Chrysopodes (Neosuarius) divisus
68	Walker and Chrysopodes (Neosuarius) porterinus Navás have been cited (Adams & Penny,



69	1987; Gonzalez Olazo et al., 1999; Monserrat & Freitas, 2005; Gonzalez Olazo & Reguilón,
70	2008; Ortega et al., 2014).
71	Chrysopodes (C.) lineafrons is considered an effective predator for biological control. Thus,
72	research focusing on its preference for certain pest species or even for certain pest stages, and its
73	possible interaction with other natural enemies is of great need. Since such knowledge is scarce
74	in literature, a study of the life cycle of C. (C.) lineafrons fed with eggs of B. tabaci as prey was
75	set up in this paper. Developmental time, survival, longevity and oviposition were analyzed as
76	biological parameters of C. (C.) lineafrons, and their predation ability over B. tabaci eggs under
77	laboratory conditions was assessed.
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79	MATERIALS AND METHODS
80	Study area and specimens collection
81	Entomological sampling was performed during the 2009-2010 period in two greenhouses and
82	one tomato crop field in Lules department (26 °55 '60"S-65° 20' 60"W, 382 m.a.s.l), Tucumán
83	province, northwestern Argentina (Fig. 1).
84	Chrysopodes (C.) lineafrons specimens were collected in tomato crops and the surrounding
85	vegetation using manual aspirators for adults; and manually with a brush for immature stages.
86	Subsequently, adults were placed in 500 cm³ plastic containers covered with voile; and larvae
87	were kept in Petri dishes with paper accordions, used to avoid cannibalism, with Sitotroga
88	cerealella Oliver eggs for their feeding.
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90	Laboratory assay

Laboratory assay



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Rearing of C. (C.) lineafrons took 12 months. Adults were placed in 5 liter plastic containers covered with voile, secured with an elastic band and properly labeled with collection date and number of individuals. A circular paper was placed inside the container, for the females to lay the eggs. Specimens were fed daily with a mixture of yeast, pollen, honey and water in a 10:1:5:/ proportion and provided of water with moistened cotton. To evaluate the ingestion of C. (C.) lineafrons, eggs breeded in the laboratory were selected randomly and placed in 2 cm diameter individual plastic containers with an hermetic seal, maintained at 27°C, 65% humidity and a photoperiod of 12:12 (L:D) until hatching. Once lacewing larvae emerged, they were separated in two groups. A known number of B.tabaci eggs were offered to one of the groups while S. cerealella eggs were fed to the other group, used as control. After 24 hours, the number of predated eggs and C. (C.) lineafrons larvae survival were recorded. When the larvae of C. (C.) lineafrons achieved the pupal stage, they were placed in 1 liter plastic containers covered with voile. The number of males and females which emerged was recorded in each repetition. Emerging individuals were fed and maintained under the same breeding conditions of the adults. Number of eggs laid and adults longevity was registered every 24 hours (Fig.2). **Data analysis** Data obtained in the laboratory, such as number of offered and consumed preys, emergence date of C. (C.) lineafrons larvae; oviposition, number of adult individuals placed in each container and numbers of eggs laid daily were registered in spreadsheets.



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Mean, average and percentage of eggs predated by C. (C.) lineafrons were calculated. In addition, the different life stages survival (egg, larva and pupa) of lacewing was determined, as well as adults longevity and number of eggs laid per female per day fed both with B. tabaci eggs and S. cerealella eggs. **RESULTS** Of the 90 eggs of C. (C.) lineafrons selected at the beginning of this study, only 71 hatched, of which 34 were fed with B. tabaci eggs and 37 with S. cerealella eggs. During the ingest assay, it was observed that a total of 34 individuals reached the adult state; 28 of these having been fed with S. cerealella eggs, and 6 with B. tabaci eggs. **Ingestion assay** In general, larvae of C. (C.) lineafrons consumed an average of 127.04 B.tabaci eggs and 44 S. cerealella eggs per day. When each larval stage was evaluated separately, it was observed that larvae I consumed a maximum of B. tabaci 556 eggs and 189 S. cerealella eggs, while larvae II consumed 531 B. tabaci eggs and 240 S. cerealella eggs, and larvae III consumed 619 B. tabaci eggs and 150 S. cerealella eggs per day (Table 1). On the other hand, the developmental time of the different larval stages was similar between larvae I and II, whereas it was shorter for larvae III. Regarding C. (C.) lineafrons developmental time, an average of 45 days was recorded when the larvae fed with *B. tabaci* eggs and 35 days when they fed with *S. cerealella* eggs (Table 2).

Survival, longevity and oviposition



In general, C. (C.) lineafrons eggs survival was 81.1%. The immature stages of C. (C.) lineafrons 135 fed with B. tabaci eggs exhibited lower survival (8.2%) than those fed with S. cerealella eggs 136 (30.1%). Lacewings survival significantly decreased from the larval to the adult stage, for 137 individuals fed both with B. tabaci eggs and S. cerealella eggs (Fig. 3). 138 In relation to C. (C.) lineafrons adults longevity, both a greater number of specimens and a 139 140 greater longevity (41 days approximately) was observed from larvae fed with S. cerealella eggs, while for those fed with B. tabaci eggs longevity was 20 days (Fig. 4a). Lastly, C. (C.) lineafrons 141 oviposition was also higher in females fed with S. cerealella eggs (Fig. 4b). 142 143 **DISCUSSION** 144 This study represents the first record of C. (C.) lineafrons predation capacity over B. tabaci eggs 145 in Argentina, considering this specie as a potential biological control agent. It is worth remarking 146 that this lacewing specie was recently cited for Tucumán province, in northwestern Argentina 147 (Ortega et al., 2014). 148 In relation to ingest capacity, C. (C.) lineafrons larvae consumed a higher number of B. 149 tabaci eggs than S. cerealella eggs per day, with larvae II standing out. These results are 150 151 comparable to those reported by Legaspi et al. (1994), who determined that a greater number of B. tabaci eggs were necessary for the development of Chrysoperla rufilabris Bumeister larvae 152 III. Other studies reported that *Chrysoperla carnea* Stephens was able to eat up to 200.5 B. 153 154 tabaci nymphs and 171.8 Amrasca devastans Distant nymphs (Nisar Syed et al., 2005); and that the larvae could consume about 8000 S. cerealella eggs and 510 B. tabaci pupae throughout its 155 156 development (Gallardo et al., 2005). Also, Legaspi et al. (1994) determined that C. rufilabris 157 larvae consumed an average of 531.55 B. tabaci eggs per day, whereas Avila et al. (2009)



reported that Chrysoperla argentina Steimann larvae ingested an average of 275 B. tabaci eggs 158 per day. This behavior might be attributed to the hypothesis suggested by Nisar Syed et al. 159 (2005), who pointed out that prey density exhibits a strong influence on predation potential; i.e. 160 the increase in egg consumption could be due to increases in larvae density. 161 Furthermore, the developmental time of C. (C.) lineafrons was 45 days when they were fed 162 163 with B. tabaci eggs and 35 days when they were fed S. cerealella eggs. In relation to this, studies performed by Ramirez-Delgado et al. (2007) with Ceraeochrysa sp. nr. cincta determined that 164 the total developmental time, from egg to adult emergence, was 29 days when the species was 165 fed with S. cerealella eggs. Moreover, Legaspi et al. (1994) recorded a longer duration of larval 166 developmental time in C. rufilabris fed with B. tabaci eggs compared to those fed with S. 167 cerealella eggs. However, Nisar Syed et al. (2005) recorded a shorter developmental time on C. 168 cornea fed with B. tabaci eggs compared to those fed with A. devastans eggs. 169 C. (C.) lineafrons survival decreased as the life cycle progressed; both for individuals fed 170 with B. tabaci eggs, and for those fed with S. cerealella eggs. These results are similar to those 171 obtained by Ramirez-Delgado et al. (2007), who determined a steady decrease of C. sp. nr. 172 cincta longevity when it was fed with S. cerealella eggs. The longevity of C. (C.) lineafrons 173 174 adults fed with S. cerealella eggs was 41 days; while for those fed with B. tabaci eggs it was 20 days, results that agree with those reported by Ramírez-Delgado et al. (2007), who determined a 175 176 greater longevity for C. sp. nr. cincta adults fed with S. cerealella eggs. 177 In general, C. (C.) lineafrons survival, longevity and number of eggs laid per female were greater when they were fed with S. cerealella eggs than with B. tabaci eggs. In relation to this, 178 179 Giffoni et al. (2007) determined that C. externa completed its life cycle only when being fed



180	with S. cerealella eggs; while Legaspi et al. (1994) recorded that C. rufilabris larvae showed a			
181	greater preference for S. cerealella eggs than for B. tabaci eggs.			
182	It has been observed that C. (C.) lineafrons exhibits a similar behavior to that of other			
183	lacewing species; with the peculiarity that their predation efficiency is much higher, thus turning			
184	this species into an excellent tool for efficient biological control of <i>B. tabaci</i> in tomato crops.			
185	Therefore, we conclude that the larval stages of C. (C.) lineafrons require a greater number of B.			
186	tabaci eggs than of S. cerealella eggs to complete their life cycle; larvae II and III of C. (C.)			
187	lineafrons consume more B. tabaci eggs; larval I stage lasts longer than the two other larval			
188	stages; survival and longevity of C. (C.) lineafrons adults was greater in individuals fed with S.			
189	cerealella eggs, and the number of eggs laid per female was also higher.			
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- 226 Brown J. 1993. Evaluación crítica sobre los biotipos de mosca blanca en América, de 1989 a
- 227 1992, pp. 1-9 *In* Memorias del taller Centroamericano y del Caribe sobre Moscas Blancas.
- 228 CATIE, Turrialba, Costa Rica.
- 229 Brown J, Coats SA, Bedford ID, Markhan PG, Bird J, Frohlich DR. 1995. Caracterization
- and distribution of esterase electromorphs in the Whitefly, Bemisia tabaci (Genn.)
- 231 (Homóptera: Aleyrodidae). Biochemical Genetics 33: 205-213.
- Byrne ND, Bellows TS Jr, Parrella MP. 1990. Whiteflies in agricultural systems, pp. 227-261
- 233 In Gerling D [ed.], Whiteflies: their bionomics, pest status and management, Intercept Ltd.,
- Andover, U.K.
- 235 Gallardo J, Vargas Camplis JJ, Lopez Arrovo JI, Reves Rosas MA. 2005. Uso y manejo de
- 236 Crhysoperla carnea. Patronato para la investigación fomento y sanidad vegetal. Centro
- regional de estudios y reproducción de organismos benéficos. H. matamoros, Tamaulipas, pp.
- 238 1-9.
- 239 Giffoni J, Valera N, Diaz F, Vasquez C. 2007. Ciclo Biológico de Chrysoperla externa
- 240 (Hagen) (Neuroptera: Chrysopidae) alimentada con diferentes presas. *Nota Técnica, Bioagro*
- 241 19: 109-113.
- 242 Gonzáles Olazo EV, Toledo S, Zaia G. 1999. Nuevas citas de Chrysopidae (Neuroptera:
- Planipennia) para la Argentina. Acta Zoológica Lilloana 45: 151-152.
- González Olazo EV, Reguilón C. 2008. Orden Neuroptera, pp. 235-248 In Claps LE, Debandi
- G, Roig Juñent S [eds.], Biodiversidad de Insectos de la Argentina II. Sociedad
- 246 Entomológica Argentina.



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270	Reguilón C, Alderete G, Flores GC. 2011. Chrysoperla argentina (Neuroptera: Chrysopidae)			
271	un enemigo natural promisorio para el control de la mosca blanca (Trialeurodes			
272	vaporariorum) en cultivo de pimiento en invernaderos – Lules, Tucumán. Horticultura			
273	Argentina 30: 55.			
274	Viscarret M. 2000. Estudios biológicos sobre Aleyrodidae de importancia económica (Insecta:			
275	Hemiptera) con énfasis en el complejo Bemisia tabaci (Gennadius) y su posible control			
276	biológico. Tesis doctoral, Facultad de Ciencias Exactas y Naturales, Universidad Nacional o			
277	Buenos Aires, pp. 131.			
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283	FIGURE CAPTIONS			
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285	Figure 1 Geographical location of the tomato crops in Tucumán Province, northwestern			
286	Argentina.			
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288	Figure 2 A . Rearing of <i>C</i> . (<i>C</i> .) <i>lineafrons</i> ; B . Eggs; C .and D . Larvas; E . Pupal stage; F . Adults.			
289				
290	Figure 3 Viability of different developmental stages of C. (C.) lineafrons fed with B. tabaci eggs			
291	and S. cerealella eggs.			
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293	Figure 4 (a) Longevity and (b) Oviposition of <i>C. (C.) lineafrons</i> adult fed with <i>B. tabaci</i> and <i>S.</i>
294	cerealella eggs.
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296	TABLE CAPTIONS
297	
298	Table 1. Daily egg consumption of <i>C.</i> (<i>C.</i>) lineafrons larvae fed with <i>B. tabaci</i> eggs and <i>S.</i>
299	cerealella eggs.
300	
301	Table 2. Developmental time of different stages of <i>C. (C.) lineafrons</i> fed with <i>B. tabaci</i> eggs and
302	S. cerealella eggs.
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Table 1(on next page)

Daily eggs consumption of C. (C.) lineafrons larvae fed with B. tabaci eggs and S. cerealella eggs.



- 1 Table 1 Daily eggs consumption of C. (C.) lineafrons larvae fed with B. tabaci eggs and S.
- 2 cerealella eggs.

	Number of individuals evaluated	Eggs consumption			
Stage		Total	Average/individuals	Average/individuals/day	Rank
B. tabaci					
L1	34	33607	988.4	88.01	0-556
L2	31	43188	1393.2	124.8	0-531
L3	23	27446	1193.3	168.3	0-619
S. cerealella					
L1	37	8125	219.6	22.02	0-189
L2	31	10367	334.4	55.2	0-240
L3	30	6020	200.6	54.8	0-150

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Table 2(on next page)

Developmental time of different stages of C. (C.) lineafrons fed with B. tabaci eggs and S. cerealella eggs.

- 1 Table 2 Developmental time of different stages of C. (C.) lineafrons fed with B. tabaci eggs and
- 2 S. cerealella eggs.

Stage	Mean \pm SD	Life cycle duration
B. tabaci		
L1	11.23 ± 0.56	45.2
L2	11.16 ± 1.03	
L3	7.09 ± 0.74	
Pupa	15.75 ± 0.51	
S. cerealella		
L1	9.97 ± 0.69	34.9
L1 L2	6.06 ± 0.7	34.9
L3	$3,66 \pm 0,4$	
Pupa	$15,21 \pm 0,57$	

4 Means \pm SE followed by P < 0.05

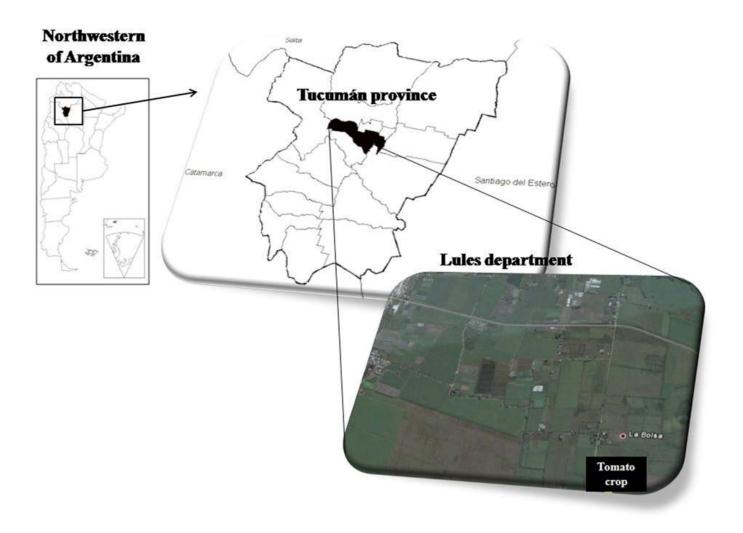
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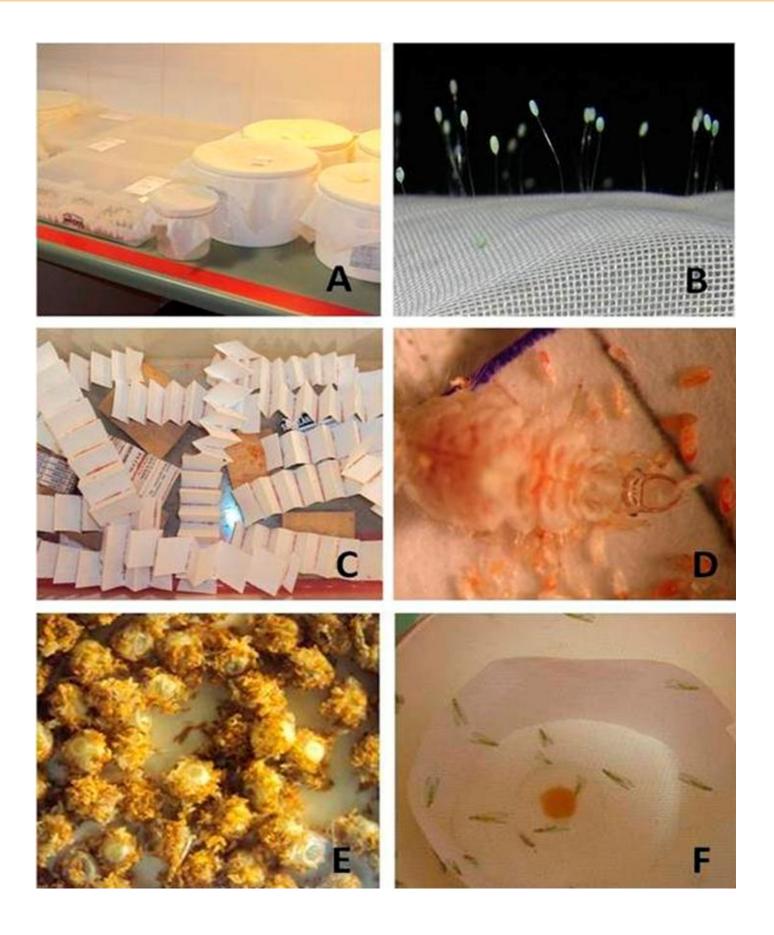
Geographical location of the tomato crops in Tucumán Province, northwestern Argentina.





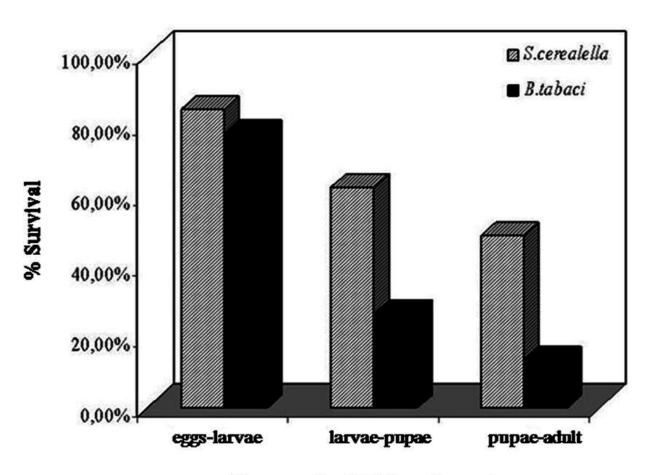
A. Rearing of C. (C.) lineafrons, B. Eggs, C.and D. Larvas, E. Pupal stage, F. Adults.







Viability of different developmental stages of *C. (C.) lineafrons* fed with *B. tabaci* eggs and *S. cerealella* eggs.



Chrysopodes (C.) lineafrons stage



(a) Longevity and (b) Oviposition of *C. (C.) lineafrons* adult fed with *B. tabaci* and *S. cerealella* eggs.

