Evaluation of mortality factors using life table analysis of Gratiana boliviana, a biological control agent of tropical soda apple in Florida

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ABSTRACT

Tropical soda apple (TSA), Solanum viarum Dunal (Solanaceae), has invaded many pastures and natural areas in Florida. The biological control agent Gratiana boliviana Spaeth (Coleoptera: Chrysomelidae) is providing adequate control of TSA stands in South and Central Florida. However, poor or no establishment of this agent has occurred in northern Florida. The goal of this study was to examine the mortality factors that influence the population dynamics of G. boliviana in Florida. Horizontal life tables were constructed by following cohorts of individuals in the laboratory and inside closed and open cages at field sites in Central and North Florida. Fertility life table parameters were estimated using laboratory and field data. In addition, as part of a vertical life table analysis, TSA plants were sampled every two weeks in pastures in Central Florida, and counts of all G. boliviana and other herbivores and predators were recorded. Survival to adulthood was similar between Central and North Florida (open cages: 12–19%). Intrinsic mortality (laboratory data) and biotic factors (predation) together accounted for 75% of the mortality of immature stages. Survival of beetles in a natural population determined from vertical life tables was 5%. A complex of three mirid species (Engytatus modesta Distant, Tupiocoris notatus Distant, and Macrolophus sp.) were the most abundant predators found in the field, and are known to feed on G. boliviana eggs and larvae. Positive growth rates (rm = 0.3) during the summer and early fall allow the beetle population to increase and provide suppression of TSA in Central Florida.

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1. Introduction

Several studies suggest that the establishment of biological control agents is greatly affected by biotic and abiotic factors found in the introduced areas (e.g., Sutherst and Maywald, 1985; Byrne et al., 2003; Chosheh, 2005). Climate matching is commonly used to select biocontrol agents that are well adapted to the climate in the introduced range (Sutherst and Maywald, 1985; Byrne et al., 2003; Senaratne et al., 2006; Mukherjee et al., 2011). In addition, predation and parasitism have been shown to limit the success of many biological control agents (e.g., Goeden and Louda, 1976;
Briese, 1986; Semple and Forno, 1987; Ghosheh, 2005). Life table studies are often employed to examine the mortality factors affecting insect populations in the field (e.g., Royama, 1981; Carey, 1993, 2001; Kuhar et al., 2002; Zanuncio et al., 2006; Yang et al., 2010). Moreover, life tables provide estimations of demographic parameters including age structure, reproductive and intrinsic rates of increase, and generation times. These parameters are useful when comparing potential population growth of different agents and for the development of mass rearing methods. Horizontal life tables are constructed by following the fate of a cohort of individuals over the course of their lives, while vertical life tables examine the age structure of a population at a single moment in time (Bellows et al., 1992). The latter approach is commonly used for insects with overlapping generations.

Tropical soda apple (hereafter TSA), Solumon viarum Dunal (Solanaceae), native to South America, is considered a problematic weed of pastures and natural areas in Florida (Mullahey, 1996). TSA has spread rapidly throughout Florida since it was first reported in 1988 (Mullahey et al., 1993), and the plant is now present in several other states (e.g., Arizona, Arkansas, Georgia, Tennessee, Texas) (National Plant Data Center, 2010). It is a perennial weed characterized by having thorn-like prickles on leaves and stems, which are unpalatable to cattle, but has yellow fruits which are consumed by livestock, aiding in the dispersal of seed (Mullahey et al., 1993). Management practices against this weed are typically based on expensive herbicide treatments and mowing, costing Florida ranchers an estimated $6.5 to 16 million annually (Thomas, 2007). In addition, TSA harbors several viruses that can infect solanaceous crops (McGovern et al., 1994a, 1994b, 1996) and serves as an alternative host for key insect pests of cultivated crops (e.g., Leptinotarsa decemlineata Say and Manduca sexta L.) (Habeck et al., 1996; Sudbrink et al., 1999).

Foreign exploration for natural enemies of TSA was initiated in South America in 1994, and several potential biocontrol agents were identified (Medal et al., 1996). One of the most promising agents was a leaf-feeding beetle, Gratiana boliviana Speath (Coleoptera: Chrysomelidae), from Argentina and Paraguay. Host range studies demonstrated that this beetle is highly host specific, feeding and completing development only on TSA (Medal et al., 2002). G. boliviana was introduced into Florida in 2003, and since then it has successfully established throughout South and Central Florida (Medal et al., 2006; Overholt et al., 2009). Both larvae and adults feed on TSA foliage by making shot-gun holes in the leaves and eventually defoliating the whole plant (Medal et al., 2002, 2006). Adults of G. boliviana lay single eggs on the leaves and petioles, and the larvae complete five instars and pupate on the plant foliage (Diaz et al., 2008). Beetle populations increase during the spring-summer (May–Oct) and adults enter reproductive diapause during the coldest months (Nov–April) in Florida (Overholt et al., 2010; Kuhar et al., 2002; Zanuncio et al., 2006; Yang et al., 2010). Moreover, life tables provide estimations of demographic parameters including age structure, reproductive and intrinsic rates of increase, and generation times. These parameters are useful when comparing potential population growth of different agents and for the development of mass rearing methods. Horizontal life tables are constructed by following the fate of a cohort of individuals over the course of their lives, while vertical life tables examine the age structure of a population at a single moment in time (Bellows et al., 1992). The latter approach is commonly used for insects with overlapping generations.

2. Materials and methods

2.1. Insects and plants

G. boliviana used for the experiments were obtained from a mass rearing facility operated by the Florida Department of Agriculture & Consumer Services, Division of Plant Industry in Fort Pierce, FL. Insects were originally collected in Southeast Brazil in the early 2000s.

TSA plants were grown from seeds, planted in nursery pots (20 cm tall, 21 cm diameter) and fertilized with 15 g of a slow release fertilizer (19-6-12N-P-K Osmocote®, Scotts-Sierra Horticultural Products Co., Marysville, OH, USA). All plants were kept inside screen cages at the University of Florida, Indian River Research & Education Center (IRREC) located in Fort Pierce, FL. Plants were watered daily using an automatic drip irrigation system.

2.2. Data collection in the laboratory

G. boliviana pupae were held individually in Petri dishes at 24 °C with a fresh TSA leaf until adult emergence. After one week, adults were sexed by examining the ventral side of the abdomen. Males were distinguished from females by the presence of two rounded orange testes which were visible through the ventral side of the abdominal sternites (Medal et al., 2002). Single pairs of adults were placed inside cages (60 × 60 × 60 cm BugDorms, Bioquip, Rancho Dominguez, CA, USA) with one TSA plant and maintained at 24 ± 2 °C and a 14:10 L:D photoperiod (9 replicates total). Plants were inspected daily and all eggs were counted, removed and placed inside Petri dishes. If a male died before the female, the male was replaced. Lifetime fecundity was recorded for each female. Petri dishes containing fresh eggs (<24 h old) and moist filter paper were kept inside environmental growth chambers (24 ± 2 °C, 14:10 L:D photoperiod). TSA leaves were replaced as needed, and survival and date of molting were recorded daily. A total of 1247 eggs (<24 h old) were followed until adult emergence or death.

2.3. Data collection in the field

Life-history parameters of G. boliviana were studied in Central and North Florida during the summer and fall of 2010. Experiments were conducted at two field sites in Saint Lucie County (Central Florida), and a third site in Madison County (North Florida) (Table 1). Ten potted TSA plants (~25 cm tall) were caged with reproductive adults (15 females and 5 males per cage) of G. boliviana for 24–48 h to allow oviposition. All plants were inspected for eggs, and 30 eggs per plant were marked with a red indelible marker pen (red line next to, but not touching the egg) while extra eggs were removed from the plants. TSA plants with eggs were then placed in the field where they were enclosed inside a fine mesh cloth cage (46 cm diameter, 70 cm tall) placed over a wire tomato plant support. A 60-cm vertical zipper in the cage allowed access to the plant. Five TSA plants were randomly assigned to each treatment (open or closed cages). For the open cages, the bottom of the mesh was folded such that only the top half of the plant was covered. Pots were buried in the ground, straw mulch was placed around the pot, and pots were separated by 1.5 m and arranged in two rows (five plants per row). Data loggers measured temperature and humidity inside open and closed cages during the experiment. The experiment was replicated twice at the Central Florida sites and once at the North Florida site.
All plants were inspected twice per week, and the total number of eggs, small larvae (instars I, II, and III), large larvae (instars IV and V), pupae, and new adults were counted. All newly laid eggs and adults were removed from the plants in the open cages. In addition, the number of predators found on each plant was recorded during each sampling period (Central Florida only). Newly emerged adults were counted and placed together for 10 days on TSA plants in cages to obtain sexually mature adults (9–12 d pre-oviposition period, Medal et al., 2003). Two pairs of mature adults were placed in closed field cages (10 cages total, 40 adults in total) and the number of adults alive (females and males) was recorded weekly. All TSA plants were replaced every 3 weeks, and the total number of eggs per plant was recorded. The experiment was terminated when all adults had died (2 replicates in Central Florida only).

2.4. Calculations of horizontal life table parameters

Life tables were constructed from the laboratory and field data (open and closed cages). The following parameters were calculated: number of individuals alive at the beginning of each stage \(L_0\), the number individuals that died during each age interval \(d_0\), and the percent of individuals that died during each age interval \(100d_0\). The causes of mortality were separated into intrinsic factors (laboratory mortality), abiotic factors (difference between mortality in closed cages and in the laboratory), and biotic factors (difference between mortality in closed and open cages). Comparisons of mortality factors were made using the Wilson method for two independent proportions (Newcombe, 1998). Significant differences between mortality were obtained if the 95% confidence interval (CI) did not include zero, while differences between proportions were significant \((P < 0.05)\) if there was no overlap between 95% CIs.

2.5. Calculations of fertility life table parameters

Fertility life tables were constructed using data from the laboratory and the field (open cages). The following demographic parameters were calculated: net reproductive rate \(\lambda\), intrinsic rate of increase \(r_m\), finite rate of increase \(R_m\), and doubling time \(T_D\) (weeks). Net reproductive rate is the sum of eggs, small larvae (instars I, II, and III), large larvae (instars IV and V), pupae, and new adults. The experiment was terminated when all adults had died (2 replicates in Central Florida only). The jackknife method was used to estimate variance and bias of estimators in order to quantify uncertainty associated with demographic parameter estimates (for more details see Maia et al., 2000).

Longevity of adults was analyzed using two factor analysis of variance (ANOVA) with set-up (laboratory vs. field) and gender (female, male) as factors. Significant interaction was obtained \((F_{1,127} = 6.4, P = 0.01)\), therefore, single ANOVA’s were used to compare longevity between laboratory and field \((P < 0.05)\).

2.6. Calculations of vertical life table parameters

Vertical life tables of \(G. boliviana\) were constructed using naturally occurring overlapping generations at two field sites in Central Florida. One TSA infested pasture located in Okeechobee Co. (N 27.283, W 81.042) and one in St. Lucie Co. (N 27.363, W 80.560) were sampled every two weeks from June to October 2010. Beetles were originally released in 2006 at both locations and populations have been established since then. In each pasture, fifteen randomly selected TSA plants separated by at least 5 m were sampled along a longitudinal transect. Each plant was cut at the ground level, placed in a plastic bag and transported to the laboratory for data collection. The numbers of eggs, larvae, pupae and adults of \(G. boliviana\) were counted on each plant. In addition, all predators and herbivores found on plants were recorded. Leaves from each plant were harvested, oven-dried at 70°C for one week, and weighed to measure leaf biomass. The mean development times for eggs (6 d), small larvae (13 d), large larvae (9 d), and pupae (7 d) were obtained from the laboratory horizontal life table, while adult longevity (70 d) was obtained from the fertility life tables constructed from the field, and used to calculate survivorship curves. The relationship of survival to adult stage and sampling date was compared between sites using linear regression, and pooled after it was found that the slopes of the regression lines were not different (slope Okeechobee = 0.00039, slope St. Lucie = 0.00025; \(t = 1.3, P = 0.224)\).

A multiple linear regression with backward elimination of non-significant \((P > 0.05)\) independent variables was performed to examine the relationship between the number of mirid adults and the following variables: sampling date, leaf biomass, and numbers of spiders, ants, \(G. boliviana\) adults and immature \(G. boliviana\) (sum of eggs, larvae and pupae). Sampling date was included in the model to allow estimation of the effect of leaf biomass and number of beetles and predators on mirid counts independent of time. The analysis was performed with PROC REG procedure of SAS (SAS Institute, 2008).

3. Results

3.1. Horizontal life table parameters

Data from the laboratory environment (constant favorable conditions with no natural enemies) allowed the construction of a horizontal life table in which the mortality obtained during each life stage was attributed to intrinsic mortality (Table 2). The use of open and closed cages in the field allowed mortality measurements

<table>
<thead>
<tr>
<th>Location</th>
<th>GPS coordinates</th>
<th>Habitat description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Site 1 CF</td>
<td>N 27.3455, W 80.4966</td>
<td>Mosaic of open pastures and wooded areas. TSA density has been reduced by (G. boliviana)</td>
</tr>
<tr>
<td>Site 2 CF</td>
<td>N 27.4262, W 80.4050</td>
<td>Open field at research station next to field crops. Previously used for TSA-(G. boliviana) studies</td>
</tr>
<tr>
<td>Site 3 NF</td>
<td>N 30.4025, W 83.4560</td>
<td>Mosaic of open pastures and wooded areas. (G. boliviana) has not reduced the density of TSA</td>
</tr>
</tbody>
</table>

Table 1 Description of three Florida field sites used for \(G. boliviana\) horizontal life table studies. CF refers to Central Florida, NF refers to North Florida.
of abiotic factors (climatic conditions) and biotic factors (predation) (Tables 3 and 4). Mean temperature in the field varied between 26 and 30°C, and no differences were detected between open and closed cages ($F_{1,9181} = 2.1, P = 0.14$). Two closed cages were discarded because plants desiccated at site 1, and one closed cage at site 2 (Central Florida) was discarded due to the presence of the red imported fire ant, Solenops invicta. Buren. The egg stage experienced the highest mortality in both laboratory and in the field (Tables 2–4). As expected, mortality was much higher in open cages compared to closed cages (Tables 3 and 4). There was no difference in overall mortality between North and Central Florida (Wilson Independent Proportion Test, $P > 0.05$). However, egg mortality due to predation (open–closed cages) was greater in Central Florida (35%) (Table 3), while larval predation was higher in North Florida (40%) (Table 4). Time of development from egg to adult was similar in the field (30.6 ± 0.9 d) and the laboratory at 24°C (33.6 ± 0.7 d) ($F_{1,56} = 1.61, P = 0.21$).

Overall mortalities from egg to adult were as follows: 85% in open cages > 50% in closed cages > 40% in the laboratory. Independent tests of proportions showed that mortalities due to biotic factors and intrinsic factors were similar, and both were higher than mortality due to abiotic factors (Table 5). A complex of three mirid species (E. modesta, T. notatus, and Macrolopus sp.) comprised 95% of the predators found during the field studies. Repeated measures analysis of mirid densities showed no differences between replicates in Central Florida ($F_{3,83} = 1.82, P = 0.16$), but differences were detected among months ($F_{5,83} = 6.9, df = 9, 83, P < 0.0001$). Overall, mirid densities tended to increase during the season and were highest 21 days after initiating the experiment (Fig. 1).

3.2. Fertility life table parameters

Fertility life tables of G. boliviana were constructed using data from the laboratory and field (Central Florida only). Adults were followed from July to February in the field, but most adults died before the winter started. Two adults turned brown and migrated to the bottom of the cage, indicative of entering reproductive diapause (R. Diaz, unpublished results), and were excluded from data analysis. The net reproductive rate ($R_0$), intrinsic rate of increase ($\lambda$), and finite rate of increase ($r_m$) were significantly greater in the laboratory compared to the field, while no differences were detected in mean generation time ($T$) (Table 6). In contrast, doubling time (Dt) was higher in the field than the laboratory (Table 6). Life-time fecundity was similar in the laboratory (271.86 ± 43.74 eggs) and the field (232.86 ± 37.52 eggs) ($F = 0.38, df = 1, 27, P = 0.54$). Females lived longer in the field (80.4 ± 7.5 d) compared to the laboratory (60.8 ± 5.2 d) ($F = 4.4, df = 1, 75, P = 0.03$), but no differences were detected for male longevity (ca.70 d) ($F_{1,52} = 2.6, P = 0.1$). In the field, females lived longer than males ($F_{1,77} = 4.5, P = 0.03$).

3.3. Vertical life table parameters

A total of 1909 eggs, 2148 larvae, 182 pupae and 1109 adults were collected during the five month sampling period. Calculated mortality was particularly high for eggs and large larvae.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Egg</th>
<th>L1–L3</th>
<th>L4–L5</th>
<th>Pupa</th>
<th>Egg-adult</th>
</tr>
</thead>
<tbody>
<tr>
<td>$l_0$</td>
<td>1247</td>
<td>966</td>
<td>918</td>
<td>870</td>
<td>846</td>
</tr>
<tr>
<td>$d_0$</td>
<td>281</td>
<td>48</td>
<td>48</td>
<td>24</td>
<td>24</td>
</tr>
<tr>
<td>$100q_x$</td>
<td>22.5</td>
<td>5.0</td>
<td>5.3</td>
<td>2.8</td>
<td>2.9</td>
</tr>
</tbody>
</table>

$l_0$ = number of individuals alive at the beginning of each stage, $d_0$ = number individuals that died during each age interval, and $100q_x$ = percent of individuals that died during each age interval.

### Table 3

Horizontal life table for Gratiana boliviana constructed from open and closed cage data from two sites in Central Florida (mean ± SE).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Egg</th>
<th>L1–L3</th>
<th>L4–L5</th>
<th>Pupa</th>
<th>Egg-adult</th>
</tr>
</thead>
<tbody>
<tr>
<td>$l_0$</td>
<td>127.5</td>
<td>84.0</td>
<td>73.2</td>
<td>58.2</td>
<td>127.5</td>
</tr>
<tr>
<td>$d_0$</td>
<td>46.3</td>
<td>13.2</td>
<td>12.2</td>
<td>10.0</td>
<td>49.9</td>
</tr>
<tr>
<td>$100q_x$</td>
<td>33.4</td>
<td>8.8</td>
<td>8.8</td>
<td>8.8</td>
<td>5.1</td>
</tr>
</tbody>
</table>

### Table 4

Horizontal life table for Gratiana boliviana constructed from open and closed cage data from one site in North Florida.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Egg</th>
<th>L1–L3</th>
<th>L4–L5</th>
<th>Pupa</th>
<th>Egg-adult</th>
</tr>
</thead>
<tbody>
<tr>
<td>$l_0$</td>
<td>150</td>
<td>87</td>
<td>86</td>
<td>86</td>
<td>150</td>
</tr>
<tr>
<td>$d_0$</td>
<td>63</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>65</td>
</tr>
<tr>
<td>$100q_x$</td>
<td>42</td>
<td>1</td>
<td>0</td>
<td>1</td>
<td>43</td>
</tr>
</tbody>
</table>

### Table 5

Comparison of mortality factors of Gratiana boliviana reared under different conditions using the Wilson method for two independent proportions (Newcombe, 1998).

<table>
<thead>
<tr>
<th>Comparisons</th>
<th>Proportion difference</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Closed vs. open cage</td>
<td>0.317 (biotic factors)</td>
<td>0.214–0.414</td>
</tr>
<tr>
<td>Lab vs. open cage</td>
<td>0.481 (abiotic + biotic)</td>
<td>0.413–0.531</td>
</tr>
<tr>
<td>Lab vs. closed cage</td>
<td>0.163 (abiotic factors)</td>
<td>0.072–0.250</td>
</tr>
<tr>
<td>Laboratory only</td>
<td>0.399 (intrinsic mortality)</td>
<td>0.372–0.427</td>
</tr>
</tbody>
</table>

a Simple proportion.

### Table 7

A negative exponential model fitted the relationship between the number of individuals per life stage and cumulative developmental time (Fig. 2). Only 5.1% of eggs reached the adult stage.

G. boliviana was the most common herbivore found on TSA plants followed by the Colorado potato beetle Leptinotarsa decemlineata (Say) (Coleoptera: Chrysomelidae) (Fig. 3). Other herbivores included Lepidoptera larvae, stinkbugs (Pentatomidae) and long horn beetles (Cerambycidae). Mirids (E. modesta, T. notatus, and Macrolopus sp.) were the most common predators on TSA plants followed by spiders (Fig. 3). Other predators included assassin bugs (Reduviidae), red fire ants (Formicidae), big-eyed bugs (Lygaeidae: Geocorinae), ground beetles (Carabidae) and tree crickets (Grillidae) (Fig. 3). Sampling date, number of ants and G. boliviana adults were not significantly related to mirid numbers ($P < 0.05$) and therefore removed from the multiple regression model. The number of adult mirids per plant was positively related to plant biomass (partial correlation coefficient = 0.45, $P < 0.0001$), the number of immature G. boliviana (eggs, larvae and pupae) (partial correlation coefficient = 0.03, $P = 0.004$) and spiders (partial correlation coefficient = 0.03, $P = 0.0001$).

### Table 6

Horizontal life table for Gratiana boliviana constructed from open and closed cage data from two sites in Central Florida (mean ± SE).
4. Discussion

Life table studies provide measurements of stage-specific mortality factors and population parameters that are critical to understanding insect population dynamics in the field (Royama, 1981; Carey, 1993, 2001; Kuhar et al., 2002; Zanuncio et al., 2006; Yang et al., 2010). Surprisingly, this study is one of very few that has examined life table parameters of a weed biological control agent in the field (Briese, 1986; Jayanth and Bali, 1994). G. boliviana immature stages, in particular the egg and larval stages, suffered significant mortality with only 5–19% survival to adulthood. Horizontal life tables were constructed using laboratory and field data, which permitted the separation of different mortality factors. The intrinsic mortality (laboratory data) and biotic factors (predation) together accounted for 75% of the mortality of G. boliviana immature stages, while abiotic factors were less important. According to Cornell and Hawkins (1995), attack by natural enemies is the major source of mortality inflicted on immature stages of insect herbivores. Moreover, developmental stage and feeding biology of herbivores influence susceptibility to natural enemies (Cornell and Hawkins, 1995; Hawkins et al., 1997). Higher mortality rates by predators were found on ectophagous herbivores, such as G. boliviana, compared to endophagous (Hawkins et al., 1997). A complex of three mirid species (E. modesta, T. noratus, and Macrolopus sp.) was the most abundant group of predators found in the field, and they are known to feed on G. boliviana eggs and larvae (R. Diaz, personal observation). These mirids are sap-feeders as well as predators in several crop systems (Alomar et al., 1991; Sampson and King, 1996; Letourneau and Goldstein, 2001; Wheeler, 2001). This study was conducted after 4–6 years of field releases in Florida and could serve as a reference for comparison of mortality factors evaluated in the future.

Several studies have shown that G. boliviana significantly reduces TSA growth and reproduction, and has resulted in a decline of TSA populations in Central Florida (Overholt et al., 2009, 2010; Medal

### Table 6

<table>
<thead>
<tr>
<th>Parameter</th>
<th>True calculation</th>
<th>Jackknife estimate</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>( R_0 )</td>
<td>Laboratory</td>
<td>79.489 a</td>
<td>79.489</td>
</tr>
<tr>
<td>( r_m )</td>
<td>Laboratory</td>
<td>0.430 a</td>
<td>0.423</td>
</tr>
<tr>
<td></td>
<td>Field</td>
<td>0.319 b</td>
<td>0.319</td>
</tr>
<tr>
<td>( i )</td>
<td>Laboratory</td>
<td>1.537 a</td>
<td>1.525</td>
</tr>
<tr>
<td></td>
<td>Field</td>
<td>1.377 b</td>
<td>1.376</td>
</tr>
<tr>
<td>( T )</td>
<td>Laboratory</td>
<td>10.175 a</td>
<td>10.310</td>
</tr>
<tr>
<td></td>
<td>Field</td>
<td>8.260 a</td>
<td>8.288</td>
</tr>
<tr>
<td>( D_t )</td>
<td>Laboratory</td>
<td>1.611 b</td>
<td>1.631</td>
</tr>
<tr>
<td></td>
<td>Field</td>
<td>2.160 a</td>
<td>2.168</td>
</tr>
</tbody>
</table>

### Table 7

<table>
<thead>
<tr>
<th>Stage</th>
<th>Field counts (n)</th>
<th>Duration of each stage (d)</th>
<th>Number of individuals (( l_x ))</th>
<th>Number dying (( d_x ))</th>
<th>Proportion dying (( q_x ))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eggs</td>
<td>212.1</td>
<td>6</td>
<td>35.4</td>
<td>22.3</td>
<td>0.6</td>
</tr>
<tr>
<td>L1–L3</td>
<td>169.4</td>
<td>13</td>
<td>13.0</td>
<td>5.3</td>
<td>0.4</td>
</tr>
<tr>
<td>L4–L5</td>
<td>69.2</td>
<td>9</td>
<td>7.7</td>
<td>4.8</td>
<td>0.6</td>
</tr>
<tr>
<td>Pupae</td>
<td>20.2</td>
<td>7</td>
<td>2.9</td>
<td>1.1</td>
<td>0.4</td>
</tr>
<tr>
<td>Adults</td>
<td>123.2</td>
<td>70</td>
<td>1.8</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Survival to adult = 5.1%
and Cuda, 2010). However, poor establishment of G. boliviana has been reported in North Florida (Overholt et al., 2009). According to this study, the overall mortality to adulthood (85%) was similar between Central and North Florida. Although predation rates of immature stages varied between locations with higher egg predation in Central Florida (35%) and higher larval predation in North Florida (40%), overall mortality was similar (88% and 81% in Central and North Florida, respectively). As mortality was approximately equal in North and Central Florida, other factors must be responsible for the poor performance of G. boliviana in North Florida. Differential survival during winter is a likely explanation, either through direct effects of cold on beetles, or indirect effects through the host plant. Adult G. boliviana enter reproductive diapause at low temperatures and short photoperiods during winter months, which coincides with decreased host availability. Above ground parts of TSA die back in response to freezing temperatures, but roots survive and resprout when temperatures increase (Patterson et al., 1997). Based on our field observations in Central and North Florida, TSA is absent for longer periods in North Florida than further south, which may result in a mismatch in the seasonal phenologies of the beetle and its host-plant. Beetle diapause may terminate in North Florida before TSA has had sufficient time to recuperate from repeated freezes, leading to starvation of beetles. Additionally, host-plant quality may vary between locations, and this has been shown to affect performance of G. boliviana (Overholt et al., 2009). Additional studies are needed to elucidate the factors which negatively influence beetle performance at locations above 29° latitude (Overholt et al., 2009).

Fertility life tables have frequently been used to estimate population growth of biological control agents of insect pests, as they provide insight into the agent’s efficacy and potential for success in controlling pest populations (Bellows et al., 1992; Medeiros et al., 2000; Kuhar et al., 2002; Zanuncio et al., 2006; Gitau et al., 2007). Fertility life tables of G. boliviana were constructed using laboratory and field data. The net reproductive rate ($r_n$), intrinsic rate of increase ($r_m$), and finite rate of increase ($\lambda$) were greater in the laboratory compared to the field, and doubling time was longer in the field. These results were expected since laboratory data were obtained under putatively ideal conditions for population growth (favorable environmental conditions and absence of natural enemies). The intrinsic rate of increase for G. boliviana was converted to days for comparison with other studies on chrysomelid beetles, and was higher (0.06 in laboratory) than Octodonta nipae (Maulik) ($r_m = 0.01–0.03$ in the laboratory) (Hou and Weng, 2010), but lower than Gastrophysa viridula (DeGeer) ($r_m = 0.1$ in the laboratory) (Hon-ek et al., 2003). Overholt et al. (2010) derived $r_m$ from sampling a naturally occurring population of G. boliviana over three years and found that it varied between $–0.13$ and 0.15 depending on the time of year, which agrees well with the current finding. It was quite surprising that female G. boliviana lived longer in the field than in the laboratory. One likely explanation is that some adults in the field entered diapause in the late fall (November–December), resulting in increased longevity in the field.

Vertical life tables are often employed for organisms with overlapping generations, and assume stable age distribution and constant mortality factors acting on the populations (Bellows et al., 1992; Carey, 1993, 2001; Munga et al., 2007). The construction of vertical life tables provided estimates of age-specific mortality of G. boliviana at a larger population scale than our open and closed cage study. High mortality rates were found for all immature stages, particularly eggs and larvae, with an overall survival to adulthood of 5%, which falls in the lower survival range of other introduced insect herbivores (see review by Cornell and Hawkins, 1995). A possible explanation for the higher survival (12–19%) found in open cages was cage design; the half cages may have negatively influenced predator searching behavior and provided some protection from abiotic factors (e.g., rain and wind).

As indicated previously, mirids (76%) were the most common predators found on TSA plants throughout this field study. Moreover, the number of adult mirids per plant was positively correlated with plant biomass, G. boliviana immature stages and spiders. These mirid species are known to be both phytophagous and predaceous (Letourneau and Goldstein, 2001; Wheeler, 2001), which may explain the positive correlation with plant biomass and beetle densities. Both mirids and spiders may share a common prey such as G. boliviana larvae, which resulted in their increased abundance in the field. In addition, G. boliviana was the most common herbivore found during this study, supporting evidence from previous studies (Overholt et al., 2009, 2010; Medal and Cuda, 2010) indicating that this biocontrol agent exerts a strong top-down impact on TSA populations.

In summary, this study provides a better understanding of the different mortality factors affecting the population dynamics of G. boliviana, a biocontrol agent of TSA in Florida. Even though significant mortality to adulthood (81–95%) was found in the field, positive growth rates result in a reduction of TSA densities in the field (Overholt et al., 2010). In addition, herbivore damage in combination with other factors such as plant diseases may further reduce plant performance (see Caesar, 2005). TSA is known to host numerous plant viruses including the TSA mosaic virus (TSAMV) that affects plant growth and is widespread in Florida (Adkins et al., 2007). A complex of three mirid species was the most common predator group found on TSA plants, but further studies are needed to investigate individual predation rates of each mirid species on immature stages of G. boliviana. Finally, life table studies provide useful measurements of population performance that can provide insight into the establishment and population growth of introduced biological control agents.

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