Functional Response of the Azalea Plant Bug (Heteroptera: Miridae) and a Green Lacewing Chrysoperla rufilabris (Neuroptera: Chrysopidae), Two Predators of the Azalea Lace Bug (Heteroptera: Tingidae)

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ABSTRACT Azalea plant bug (Rhinocapsus vanduzeei Uhler) fifth instars and a commercially obtained green lacewing (Chrysoperla rufilabris Burmeister) first and second instars exhibited a type II functional response when caged with varying densities of fourth or fifth instar azalea lace bug, Stephanitis pyrioides (Scott), prey. Attack coefficients for combined fourth and fifth instar prey were statistically similar for R. vanduzeei and C. rufilabris (0.052 and 0.057, respectively). The handling time was significantly greater for R. vanduzeei (3.96 h) than C. rufilabris (2.41 h). Search efficiency generally declined for both predators as initial azalea lace bug density increased. C. rufilabris killed significantly more fourth and fifth instar prey than R. vanduzeei (8.0 and 6.0, respectively) in 24 h. Results indicate that C. rufilabris is a more suitable candidate for augmentative, not inoculative, release for azalea lace bug control than R. vanduzeei. However, R. vanduzeei can effect reductions in azalea lace bug populations in the landscape as a component of the guild of lace bug’s natural enemies and should be considered in conservation efforts.

KEY WORDS Augmentative release, Chrysoperla rufilabris, functional response, Stephanitis pyrioides, Rhinocapsus vanduzeei, urban landscape


FEW NATURAL ENEMIES OF THE AZALEA LACE BUG HAVE BEEN REPORTED (NEAL ET AL. 1991, BALSDON ET AL. 1996). THESE INCLUDE THE MYMARID, Anagrus takeyanus Gordh (an egg parasitoid), and three predatory mirids Dieomyzus rhododendri Dolling, Rhinocapsus vanduzeei Uhler (Braman and Beshar 1994), and Stethocnous japonicus Schumacher (Henry et al. 1986, Neal et al. 1991). D. rhododendri and R. vanduzeei are generalists. S. japonicus, an obligate lace bug predator (Neal and Haldemann 1992), has been reported only in Beltsville, MD, and in Rockland County, NY (Schwartz 1989).

The azalea plant bug, R. vanduzeei, described in 1890 (Uhler 1890), is a generalist predator found on azaleas (Wheeler and Herring 1979) and raspberry (Van Duzee 1904). It is univoltine, and egg hatch is synchronized with azalea bloom. R. vanduzeei has been observed feeding on the azalea lace bug, whiteflies, leafhoppers, aphids, small flies, thrips (Braman and Beshar 1994), fall armyworm [Spodoptera frugiperda (J. E. Smith)] eggs and larvae, twospotted spider mite (Tetranychus urticae Koch), azalea leafminer [Caloptilia azaleella (Brants)] pupae, each other (Stewart and Braman 1999), and azalea pollen (Wheeler and Herring 1979). This predator has been observed on Alabama (Rhododendron alabamense Rehder), Flame [Rhododendron austrinum (Small) Rehder], Korean azaleas [Rhododendron yedoense Maximowicz variety poukhanense (Leveille) Nakai], Piedmont [Rhododendron calendulaceum (Michaux) Torrey], and wild swamp Rhododendron viscosum (L.) Torrey, as well as the Ghent hybrids of the flame
azalea. Its recorded range is from Quebec (Moore 1907) and Maine to Florida and as far west as Missouri (Wheeler and Herring 1979). It has also been reported in Manitoba, Canada (Henry and Wheeler 1988).

Green lacewings feed on a diverse array of arthropod prey including azalea lace bugs (Principi and Canard 1984, and references therein; Ehler and Kinsey 1995; Shrewsbury and Smith-Fiola 2000). Although little data for relative abundance of lacewing species on azaleas exists, studies in Georgia indicate that C. rufilabris is the dominant species in Georgian pecan orchards (Dinkins et al. 1994, Smith et al. 1995). Furthermore, C. rufilabris seems better adapted than Chrysoperla carnea Stephens to the more humid conditions in the southeastern United States (Taubers and Tauber 1983).

The functional response, first discussed by Solomon (1949), describes the relationship between prey density and the number of prey consumed by an individual predator. Holling (1959) described three possible responses in which the number of prey consumed rises linearly, hyperbolically, or sigmoidally, for a type I, type II, and type III response, respectively. The generalist predatory mirid Deraeocoris nebulosus Uhler exhibited a type II response, characteristic of most arthropod predators, when challenged with varying levels of oak lace bug [Corythucha arcuata (Say)] fourth and fifth instars (Wheeler et al. 1975). C. rufilabris exhibited a type I or a type II functional response when fed cotton aphids (Aphis gossypii Glover) or Heliothis virescens (F.) eggs or larvae (Norstdlund and Morrison 1990).

In our study, the functional response, handling time, and attack coefficient of R. vanduzeei and C. rufilabris to fourth or fifth instar azalea lace bugs, Stephanitis pyrioides, on azaleas were determined, as was the number of azalea lace bugs killed in 24 h.

Materials and Methods

Azalea lace bugs were collected in Griffin, GA, and were kept in 1.0 m² screened cages at 27 ± 1°C and a photoperiod of 14:10 (L:D) h, and fed azaleas (various cultivars) as needed.

Immature R. vanduzeei were collected from native azaleas, Rhododendron canescens (Michaux) Sweet, and Rhododendron austrinum at Callaway Gardens (Pine Mountain, GA) on 9, 13, 20 April 1999 by beating the foliage over a 40 cm × 24 cm enamel pan. They were placed in individual 11 cm diameter × 2.4 cm petri plates containing one fall armyworm egg mass, an azalea leaf, and piece of damp paper towel. The dishes were placed in an environmental chamber (Percival Co., Boone, IA) at 21°C, 91% RH, and a photoperiod of 14:10 (L:D) h. The predators were checked every 24 h to determine if they had molted. Fall armyworm eggs, chrysanthemum aphids [Macrosiphoniella sanborni (Gillette)], thrips (Echinothrips americanus Morgan), and fourth and fifth instar azalea lace bugs were supplied as food. Neonate fifth instar R. vanduzeei (as determined by the length of the wing pads) were held without food for 24 h in individual petri dishes with only a moist paper towel immediately before testing.

Problems with comparing functional response studies in petri dishes with whole-plant field studies have been discussed in detail elsewhere (O’Neil 1997). Cages containing azalea terminals were used instead of petri dishes in our studies to more closely simulate plant architecture and thus produce a more field-applicable functional response curve (Messina and Hanks 1998). Mesh screen (Chicopee Manufacturing Co., Cornelia, GA) [4.96 × 4.96 apertures/cm²] was attached to cover the 3.2-cm diameter hole in the base of the 11.0 cm × 5.0 cm plastic cage (Thorton Plastic Co., Salt Lake City, UT) (Klingeman et al. 2000a). Inner cage surface area equaled 212.1 cm². An azalea terminal (Rhododendron obtusum ‘Hinodegiri’) with four to seven leaves was inserted through a 2.0-cm long section of 6.4-mm i.d. Nalgene Grade VI Premium NonToxic Tubing (Nalge Co., Rochester, NY) and sealed using Parafilm “M” Laboratory Film (American Can Co., Greenwich, CT) (Klingeman et al. 2000a). The stem was inserted through a 0.95-cm diameter hole in the lower half of a Falcon 1007 60 × 15 mm petri dish (Becton Dickinson, Lincoln Park, NJ), which served as the bottom of the cage. Azalea lace bugs do not feed on new growth; thus terminal foliage was removed. The stem was in contact with 60 ml of water contained in a 120-ml plastic cup.

One, 3, 5, 10, 15, 20, 25, and 30 fifth instar azalea lace bugs were placed on the top of the foliage. One fifth instar R. vanduzeei was introduced on the bottom portion of the stem. Controls were maintained without the predator present. After 24 h, the predator was removed from the cage, and the number of live azalea lace bugs was determined. All tests were performed in an environmental chamber (Percival Co., Boone, IA) at 21°C, 91% RH, and a photoperiod of 14:10 (L:D) h. Stem length and diameter were measured, and leaf area was determined using a Li-Cor model 3100 leaf area meter (Li-Cor, Lincoln, NE). The experiment was repeated on seven occasions until a minimum of 15 replications at all densities were obtained. The experiment was also repeated simultaneously on seven occasions using fourth instar azalea lace bugs as prey at all densities.

Commercially obtained, “prefed” late first or early second instar green lacewings (Beneficial Insectary, Oak Run, CA) were tested using the system described, except the larvae were not starved for 24 h but were used within 18 h from the time they were received. Preliminary data indicated that the first and second instars were capable of capturing and killing azalea lace bug fourth and fifth instars. Tests were conducted using 1, 3, 5, 10, 15, 20, 30, and 40 azalea lace bug fourth or fifth instars per cage. Leaf area was determined as described above. The experiment was repeated on four occasions until a minimum of 10 replications at all prey densities were obtained.

Data on prey killed were analyzed using a two-way analysis of variance (ANOVA) of the percent prey killed at each density (Wells and McPherson 1999). The type of the functional response was determined.
by performing a logistic regression of the percent prey killed as related to their initial density (Trexler et al. 1988). The linear coefficient of the plot of the proportion of prey killed versus the initial number of prey killed is negative for a type II response and positive for a type III response (Juliano 1993). Data were fitted to both type II and type III models and compared with the expected values predicted by the “random-predator” equation (Rogers 1972) for a type II functional response:

\[ N_e = N_0 \left[ 1 - \exp \left\{ a (T_h N_e - T) \right\} \right] \]

\[ N_e = \text{number of azalea lace bugs killed} \]
\[ N_0 = \text{initial number of azalea lace bugs} \]
\[ a = \text{attack coefficient} \]
\[ T_h = \text{handling time in hours} \]
\[ T = \text{total time prey is exposed to the predator} \]

The equation for a type III response (Juliano 1993) is

\[ N_e = N_0 \left\{ 1 - \exp \left\{ \frac{(d + b N_0)(T_h N_e - T)}{(1 + c N_0)} \right\} \right\} \]

The constants b, c, and d relate the attack coefficient to the initial number of azalea lace bugs per cage. For all data sets, the criteria for simplifying the model were met (Juliano 1993); thus, the model reduces to

\[ N_e = N_0 \left\{ 1 - \exp \left\{ (b N_0)(T_h N_e - T) \right\} \right\} \]

The parameters of the functional response, the attack coefficient, and handling time were determined by performing a nonlinear least squares regression and a t-test used to determine significant differences. Parameters are not significantly different when confidence intervals include zero (Juliano 1993). Data sets for fourth and fifth instar prey were combined for a given predator when no significant differences were observed for the attack coefficient and the handling time, as determined by nonlinear least squares regression. A one-way ANOVA was used to determine significant differences in leaf area among the prey densities (SAS Institute 1985).

**Results**

**Prey Killed and Search Efficiency.** Control mortality was negligible. Survival of azalea lace bug nymphs was 99% \((n = 180)\) and 98% \((n = 50)\) after 24 h in the cages without *R. vanduzeei* or *C. rufilabris* present, respectively. *C. rufilabris* killed significantly more fourth and fifth instars than *R. vanduzeei* \((F = 12.94, df = 1, 408, P = 0.0004)\) (Table 1). *R. vanduzeei* killed between 0.43 and 5.55 fourth and fifth instar azalea lace bugs in 24 h depending on initial prey density (Fig. 1). At the upper asymptote, *R. vanduzeei* killed 6.0 fourth and fifth instar azalea lace bugs, *C. rufilabris* killed between 0.63 and 8.29 fourth and fifth instar azalea lace bugs in 24 h depending on initial prey density. At the upper asymptote, *C. rufilabris* killed \(~8.0\) fourth and fifth instar prey. Search efficiency for both predators, as measured by the proportion of prey killed, generally declined as initial azalea lace bug density increased (Fig. 2). At low prey densities, predators spend the majority of their time searching for scarce prey items. At the highest prey densities where the environment is saturated with prey, the search time approaches zero. Search efficiency declines because the predator is capable only of handling (subduing/consuming the prey and cleaning itself) a finite number of prey in a given amount of time (O’Neil 1990).

**Type of Functional Response.** For *R. vanduzeei*, the linear coefficients of the fourth and fifth instar data are slightly positive, but with the standard error, include zero (Table 2). When the fourth and fifth instar data are combined, the linear coefficient ± SE is positive

![Fig. 1. Number of fourth or fifth instar azalea lace bugs killed by *R. vanduzeei* and *C. rufilabris* during 24 h on caged azalea stems.](image-url)
and does not include zero. A type III functional response is suggested by the logistic regression because estimates of the linear coefficients were positive (i.e., the proportion of prey killed did not decline as sharply at lower densities than at higher densities) and the quadratic coefficient was negative (Table 2). The model fit the observed data similarly for type II and type III models with raw $r^2$ values of 0.36 and 0.38, respectively. However, the shape of the curve of the proportion of prey killed versus the initial prey concentration also indicates the type of functional response (Trexler et al. 1988, Wells and McPherson 1999). A negative slope along all parts of the curve indicates a type II response. A positive slope in the initial portion of the curve indicates a region of density-dependent predation and therefore a type III response. When the fourth and fifth instar data sets are combined (Fig. 2), the slope is negative, indicating a type II functional response.

For *C. rufilabris*, the linear coefficients are negative (Table 2), and the standard errors do not include zero. The slope increases between one and three azalea lace bug fifth instars per plant and then declines monotonically. When the fourth and fifth instar data sets are combined (Fig. 2), the slope of the line is negative. As with *R. vanduzeei*, the criteria for using the simplified type III model were met (Juliano 1993). The negative estimate of the linear coefficient and the positive estimate of the quadratic coefficient support the conclusion that *C. rufilabris* exhibits a type II functional response.

**Plant Area.** Plant areas differed significantly among initial fourth instar azalea lace bug concentrations tested ($F = 2.47, df = 6, 88, P = 0.030$) and were nearly significant in tests using fifth instar azalea lace bugs ($F = 2.06, df = 7, 128, P = 0.052$) as prey for *R. vanduzeei*. Total search area (plant plus cage) ranged from 225.5 to 245.7 sq. cm. Thus, there was no more than an 8.2% difference in total search area at the extremes. As a result, the number of azalea lace bugs killed by *R. vanduzeei* was plotted against the plant area at each initial azalea lace bug concentration. The generally low positive and negative correlation coefficients (Table 3) indicate little relationship between plant area and the number of azalea lace bugs killed for the range of areas used in this study. Although plant areas were significantly different statistically among treatments, these differences appear not to have influenced prey capture.

Plant areas among the azalea lace bug concentrations were not significantly different ($F = 0.76, df = 7, 94, P = 0.6260$ and $F = 0.96, df = 7, 95, P = 0.4726$) for *C. rufilabris* larvae challenged with fourth and fifth instar azalea lace bugs, respectively. No clear trends were apparent between the number of lace bugs killed and plant area (Table 3).

**Parameters of the Functional Response.** The attack coefficients for fifth instar *R. vanduzeei* ranged from 0.03 to 0.06 depending on prey instar (Table 4). The attack coefficients for *C. rufilabris* late-first and early second instars ranged from 0.04 to 0.09 depending on prey instar. Attack coefficients were statistically similar for the two predators at all prey stages. The handling time for *R. vanduzeei* ranged from 2.77 to 3.96 h depending on prey instar (Table 4). The handling time for *C. rufilabris* ranged from 2.37 to 2.41 h depending on prey instar. Handling time is significantly greater ($P < 0.05$) for *R. vanduzeei* (fourth and fifth instar data combined) than *C. rufilabris* at all prey instars evaluated.

**Discussion**

In petri dishes, *R. vanduzeei* fifth instars killed an average of 4.1 of the 20 azalea lace bug fifth instars provided in 24 h (Braman and Beshar 1994), which is similar to the 3.8 of 20 azalea lace bug fifth instars killed in our cage trials. Neonate first instar *Stethoconus japonicus* killed 1.8 fifth instar azalea lace bugs over a 2-d period (Neal et al. 1991). Fifth instar *Stethoconus japonicus* neonates killed 9.2 fifth instar azalea lace bugs over a 2.7-d period for a mean of 3.4/d. In both experiments using *Stethoconus japonicus*, 10 azalea lace bug nymphs were placed on a leaf and replaced every 24 h. In our study, *R. vanduzeei* killed 3.5 of the 10 fifth instar azalea lace bugs over a 24-h period. Neal did not recommend commercially culturing *Stethoconus japonicus* (Sanchez 1989). *R. vanduzeei* eggs eclosed in synchrony with azalea bloom, and *Stethoconus japonicus* eggs eclosed in synchrony with the second generation of azalea lace bugs (Neal et al. 1991).

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**Table 2.** Linear and quadratic coefficients of the curve of the mean proportion of azalea lace bugs killed by fifth instar *R. vanduzeei* nymphs and late-first/early second instar *C. rufilabris* larvae on caged azalea stems.

<table>
<thead>
<tr>
<th>Predator</th>
<th>Prey instar</th>
<th>No.</th>
<th>Linear coefficient + SE</th>
<th>Quadratic coefficient + SE</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>R. vanduzeei</em></td>
<td>4</td>
<td>95</td>
<td>+0.09 ± 0.11</td>
<td>-0.01 ± 0.01</td>
</tr>
<tr>
<td><em>C. rufilabris</em></td>
<td>5</td>
<td>129</td>
<td>+0.04 ± 0.00</td>
<td>-0.01 ± 0.01</td>
</tr>
<tr>
<td><em>R. vanduzeei</em></td>
<td>5</td>
<td>96</td>
<td>-0.11 ± 0.06</td>
<td>+0.00 ± 0.00</td>
</tr>
<tr>
<td><em>C. rufilabris</em></td>
<td>4.5</td>
<td>225</td>
<td>+0.10 ± 0.07</td>
<td>-0.01 ± 0.00</td>
</tr>
<tr>
<td><em>R. vanduzeei</em></td>
<td>4.5</td>
<td>191</td>
<td>-0.15 ± 0.04</td>
<td>+0.00 ± 0.00</td>
</tr>
</tbody>
</table>
Table 3. Correlation coefficients obtained by plotting mean azalea lace bug nymphs killed vs. the total plant area. Negative values indicate that the number of prey killed was inversely related to plant area. Positive values indicate that prey killed was directly related to plant area.

<table>
<thead>
<tr>
<th>Predator</th>
<th>Prey instar</th>
<th>Initial no. of azalea lace bugs per cage</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1</td>
</tr>
<tr>
<td>R. vanduzei</td>
<td>4</td>
<td>-0.14</td>
</tr>
<tr>
<td>R. vanduzei</td>
<td>5</td>
<td>+0.02</td>
</tr>
<tr>
<td>C. rufilabris</td>
<td>4</td>
<td>-0.03</td>
</tr>
<tr>
<td>C. rufilabris</td>
<td>5</td>
<td>0.00</td>
</tr>
</tbody>
</table>

Our results indicate that R. vanduzei would be a less effective candidate than C. rufilabris for augmentative release to control azalea lace bug. Significant differences between the species were observed in the time spent handling prey when fifth instar R. vanduzei nymphs were compared with C. rufilabris first and second instars. C. rufilabris third instars attacked more aphids than first or second instars (Burke and Martin 1956), and we expect that C. rufilabris third instars would kill more azalea lace bugs than observed in our study. Limitations to the use of R. vanduzei include difficulties in colonization. R. vanduzei is univoltine and can be collected only for a short time interval for study. Its distribution is spotty and poorly documented. Despite this, R. vanduzei is a valuable predator, often present in large numbers on azaleas, and should be conserved whenever possible. Predator numbers, including those of R. vanduzei and C. rufilabris, should be incorporated into decision-making guidelines for azalea lace bugs. Recently developed thresholds (Klingeman et al. 2000b) can be refined to incorporate biological control into management strategy.

Generalist predators, such as R. vanduzei and C. rufilabris, feed on a wide range of prey items, allowing them to survive when azalea lace bugs are scarce, a positive attribute in a biological control agent (Ehler and van den Bosch 1974, Nordlund and Morrison 1990). Ehler and Kinsey (1995) and Shrewsbury and Smith-Fiola (2000) found that green lacewing larvae (C. rufilabris and C. carnea, respectively) are suitable candidates for augmentative, but not inoculative, release for control of azalea lace bug and aphids (Manduris kinseyi Voegtlin), respectively. In both studies, once prey populations declined, few green lacewings could be found on the plant, perhaps because of cannibalism and dispersion (Ehler and Kinsey 1995, Shrewsbury and Smith-Fiola 2000). Another possibility is that green lacewings are being killed by other generalist predators, such as assassin bugs (Zelus spp.), damsel bugs (Nabis spp.), or big-eyed bugs (Geocoris spp.) (Rosenheim et al. 1993).

Although some differences may be expected between the number of azalea lace bugs killed in the lab and the field because of plant growth, temperature differences, and other variables (O’Neil 1997), Fig. 1 serves as a useful guideline for estimating the potential impact of R. vanduzei nymphs and C. rufilabris larvae on the azalea lace bug population. Applying a ratio of one C. carnea larva to 16 azalea lace bug third or fourth instars resulted in significant lace bug reductions within 2 d and was as effective as Orthene (acephate) in the nursery (Shrewsbury and Smith-Fiola 2000).

Although C. rufilabris is generally tolerant of pyrethrins, organochlorines, acetamides, and fungicides, it is intolerant of organophosphates (including chlorpyrifos, malathion, and dimethoate) and carbamates (including carbaryl) at the rates listed for aphid control in pecans (Mizell and Schiffsauer 1990). C. rufilabris is intolerant of acephate (Orthene), which is commonly used to control azalea lace bug (Shrewsbury and Smith-Fiola 2000). Similar studies have not been done for R. vanduzei. Other chemicals such as imidacloprid, and insecticidal oils and soaps, are currently being used for azalea lace bug control in ornamentals (Sparks and Hudson 1999). Laboratory and field studies are needed to determine compatibility of C. rufilabris and R. vanduzei with these compounds.

Acknowledgments

The authors thank C. Mazer and P. Andes for allowing us to collect azalea plant bugs from the native azaleas at Callaway Gardens. We also thank J. Davis and L. Wells for valuable assistance with the statistical analysis, and T. Henry

Table 4. Attack coefficients and handling times for fifth instar R. vanduzei nymphs and late first/early second instar C. rufilabris larvae when challenged with azalea lace bugs as prey on caged azalea stems.

<table>
<thead>
<tr>
<th>Predator</th>
<th>Prey instar</th>
<th>Attack coefficient + SE</th>
<th>95% confidence interval</th>
<th>Handling time (hrs) + SE</th>
<th>95% confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>R. vanduzei</td>
<td>4</td>
<td>0.06 ± 0.02a</td>
<td>0.02-0.10</td>
<td>2.77 ± 0.30a</td>
<td>1.77-3.76</td>
</tr>
<tr>
<td>C. rufilabris</td>
<td>4</td>
<td>0.08 ± 0.03a</td>
<td>0.02-0.15</td>
<td>2.37 ± 0.28a</td>
<td>1.62-2.92</td>
</tr>
<tr>
<td>R. vanduzei</td>
<td>5</td>
<td>0.03 ± 0.01a</td>
<td>0.01-0.04</td>
<td>3.83 ± 0.66ab</td>
<td>2.61-5.14</td>
</tr>
<tr>
<td>C. rufilabris</td>
<td>5</td>
<td>0.04 ± 0.01a</td>
<td>0.02-0.06</td>
<td>2.37 ± 0.35a</td>
<td>1.67-3.06</td>
</tr>
<tr>
<td>R. vanduzei</td>
<td>4.5</td>
<td>0.05 ± 0.01a</td>
<td>0.02-0.08</td>
<td>3.96 ± 0.40b</td>
<td>3.17-4.74</td>
</tr>
<tr>
<td>C. rufilabris</td>
<td>4.5</td>
<td>0.06 ± 0.01a</td>
<td>0.03-0.08</td>
<td>2.41 ± 0.22a</td>
<td>1.97-2.86</td>
</tr>
</tbody>
</table>

Means in each column followed by the same letter are not significantly different (P > 0.05).
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