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# Evaluation of artificial diets and two species of natural prey as laboratory food for *Chilocorus* spp.

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**Abstract**

Promising diets were screened and the most successful modified with additives used in artificial diets for other entomophagous insects. Two suitable diets were obtained, one for adults and one for larvae of *Chilocorus nigritus* (Fabricius) (Coleoptera: Coccinellidae). They were still inferior to natural prey and not adequate as the sole food source for rearing consecutive generations. They are valuable as substitute food in the insectary during shortages of natural prey. Oleander scale *Aspidiotus nerii* Bouché and *Asterolecanium miliaris* (Boisduval) were evaluated as natural prey for *C. nigritus* and two other potential biocontrol agents in southern Africa, *C. bipustulatus* (Linnaeus) and *C. infernalis* Mulsant. *A. nerii* and *A. miliaris* were suitable for all life stages of *C. nigritus* and adults of *C. bipustulatus* and *C. infernalis*. *A. miliaris* was inadequate for larvae of *C. bipustulatus* and *C. infernalis*.

## Introduction

*Chilocorus nigritus* (Fabricius) controls scale insect pests on several crops in numerous countries (Samways, 1984). It is particularly valuable in controlling red scale, *Aonidiella aurantii* (Maskell) (Hemiptera: Diaspididae) on citrus in southern Africa (Samways, 1984, 1988).

Laboratory rearing of large numbers of *C. nigritus* on natural prey is logistically difficult as with most entomophagous insects (Waage *et al.*, 1985). Culture contamination, seasonal availability of food substrate for prey and seasonal population cycles make maintenance of a continuous supply of adequate quantities of natural prey particularly difficult. An artificial diet is required to facilitate rearing for further research and commercial rearing of large numbers.

No reports of an artificial diet suitable for *C. nigritus* were found in the literature. Smirnoff (1958) reared *C. bipustulatus* (Linnaeus) on an artificial diet suitable for numerous other coccinellids. Okada *et al.* (1972) developed a valuable diet for the aphidophagous coccinellid *Harmonia axyridis* Pallas, based on lyophilized brood of honeybee drones. M. G. Hill (pers. comm.) had partial success with rearing *C. bipustulatus* and *C. cacti* (Linnaeus) on a brood-based diet. Diets based on honeybee brood have been widely used for entomophagous coccinellids (Matsuka *et al.*, 1972; Niijima, 1979; Matsuka *et al.*, 1982). However, in the majority of cases, when rearing entomophagous insects on artificial diets, adult size is below average, larval survival is poor and fecundity suppressed (Waage *et al.*, 1985).

The promising diets mentioned above were fed to *C. nigritus* and the most successful used as a base for improvement. Diet additives which have

been valuable in developing artificial diets for other entomophagous insects, were selected from the literature. The base diet was altered and tested in steps.

An important prey of *C. nigritus* in the field in southern Africa, is *Asterolecanium miliaris* (Boisduval) (Hemiptera: Diaspididae) on giant bamboo *Dendrocalamus giganteus* Munro (Samways, 1984; Hattingh & Samways, 1991). *Aspidiotus nerii* Bouché (Hemiptera: Diaspididae) is a valuable insectary prey for *C. nigritus* (Samways & Tate, 1986). These two prey species were used as reference points for evaluating the suitability of artificial diets for *C. nigritus*. *C. bipustulatus* and *C. infernalis* Mulsant have been imported into southern Africa as potential biocontrol agents of diaspidid scales on citrus but have failed to establish. *A. miliaris* and *A. nerii* were comparatively evaluated as food for *C. nigritus*, *C. bipustulatus* and *C. infernalis*.

## Materials and methods

*Artificial diets.* Trials were conducted under controled environmental conditions of 25–26 °C, 60–70% RH and 14L:10D photoperiod. Arenas were half-petri dishes, 100 mm in diameter, 10 mm deep, with white filter paper floors. The arenas were each covered with fine nylon gauze clamped around the petri dishes with elastic bands. Each arena contained a glass vial, 30 mm long, 8 mm in diameter, containing water with a wick of cotton wool. Polyester fibre egg pads, 20 mm × 20 mm × 5 mm were provided and replaced every seven days.

Beetles were reared on *A. nerii* prior to feeding on the diets. Adults between 2 and 4 weeks after eclosion and first instar larvae less than two days old were used. One pair of adults was placed in each arena, five replicates per diet, and ten larvae per arena, four arenas per diet.

The freeze-dried diets B, C, and E to U, were stored in air-tight glass vials in a refrigerator at 5 °C. The diet was powdered and sprinkled around the arenas. The highly hygroscopic diets became moist and soft within an hour in the high-humidity environment. Food and filter-paper floors were replaced every seven days.

Composition of artificial diets tested is given in Table 1. *C. nigritus* were reared on diet A, diet B (M. G. Hill, pers. comm.), diet C and diet D (Smirnoff, 1958). The most successful of these, diet B, was then modified by adding potential phagostimulants, producing diets E to I. The two most successful of these were modified by adding vitamin and mineral mixes to one, producing diets J to M, and a series of miscellaneous supplements to the other, diets N to T. The most successful components were incorporated into diet U.

*Initial screening. C. nigritus* individuals were reared on *A. nerii* and on *A. miliaris*, and aspects of their biology measured for comparison with individuals reared on the artificial diets. A trial was conducted in which no food was provided but water was available, and another without food or water. Water was provided in vials with all other diets.

Diet A was presented as thin streaks on an inverted watch glass. The brood and royal jelly in diet B were blended together, the wheatgerm, yeast and vitamin C were combined, ground to a fine powder, thoroughly mixed with the wet constituents and the mixture freeze dried. The agar powder in diet C was mixed with hot water, cooled to 45 °C and the other constituents, mixed as in diet A, added. Diet C and D were allowed to set in plastic drinking straws, 6 mm in diameter, split down the length and 3 mm lengths of the diets were removed from the straws and placed in the arenas.

*First alterations.* A number of potential phagostimulants were added to diet B. In diet E (Table 1), the honey, royal jelly and brood were blended together and the dry constituents added. The sucrose and glucose of diets F and G respectively, were mixed with the dry constituents. The pulverised Oleander scale and Oleander scale + honey in diets H and I respectively were added to the wet constituents before mixing with the dry components.

*Table 1.* Composition of diets tested, percentages are proportions of the wet weights of the complete diets for each component

| Component | Proportion of diet (%) | | | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | A | B | C | D | E | F | G | H | I | J | K | L | M | N | O | P | Q | R | S | T | U |
| Honeybee brood | | 87.4 | 65.4 | | 83.5 | 83.5 | 83.5 | 86.1 | 82.1 | 59.6 | 82.0 | 73.8 | 81.0 | 83.4 | 78.5 | 78.5 | 78.5 | 82.5 | 82.5 | 82.5 | 54.8 |
| Wheatgerm | | 8.8 | 6.6 | | 8.4 | 8.4 | 8.4 | 8.6 | 8.3 | | 8.3 | 7.4 | 8.1 | 8.4 | 8.4 | 8.4 | 8.4 | 8.4 | 8.4 | 8.4 | 8.3 |
| Brewers yeast | | 0.2 | 0.2 | 0.4 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.1 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 | 0.2 |
| Ascorbic acid | | 1.8 | 1.4 | | 1.7 | 1.7 | 1.7 | 1.8 | 1.7 | 1.2 | 1.7 | 1.5 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 |
| Royal jelly | | 1.8 | 1.4 | 3.5 | 1.7 | 1.7 | 1.7 | 1.8 | 1.7 | 1.2 | 1.7 | 1.5 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 1.7 | 3.0 |
| Agar powder | | | 0.8 | 1 | | | | | | | | | | | | | | | | | |
| Additional water | 50 | | 24.2 | 76.7 | | | | | | | | | | | | | | | | | |
| Honey | 50 | | | 4.6 | 4.5 | | | | 4.5 | 3.3 | 4.5 | 4.1 | 4.4 | | | | | | | | 4.5 |
| Oleander scale | | | | 1.5 | | | | 1.5 | 1.5 | 1.1 | 1.5 | 1.3 | 1.5 | | | | | | | | 3.0 |
| Sucrose | | | | 12.3 | | 4.5 | | | | | | | | | | | | | | | 4.0 |
| Glucose | | | | | | | 4.5 | | | | | | | 4.5 | 4.5 | 4.5 | 4.5 | 4.5 | 4.5 | 4.5 | 9.0 |
| Wheatgerm oil | | | | | | | | | | 33.5 | | | | | | | | | | | 0.5 |
| Vitamin E | | | | | | | | | | | 0.1 | | | | | | | | | | |
| Supplement 1 (multi-vitamin & mineral) | | | | | | | | | | | | 10.2 | | | | | | | | | |
| Supplement 2 (multi-vitamin) | | | | | | | | | | | | | 1.4 | | | | | | | | |
| Cholesterol | | | | | | | | | | | | | | 0.1 | | | | | | | |
| Casein | | | | | | | | | | | | | | | 5.0 | | | | | | |
| Amino acid concentrate | | | | | | | | | | | | | | | | 5.0 | | | | | |
| Dessicated liver | | | | | | | | | | | | | | | | | 5.0 | | | | |
| Supplement 3 | | | | | | | | | | | | | | | | | | 1.0 | | | |
| Supplement 4 | | | | | | | | | | | | | | | | | | | 1.0 | | |
| Pollen | | | | | | | | | | | | | | | | | | | | 5.0 | |
| Fructose | | | | | | | | | | | | | | | | | | | | | 11.0 |

*Second alterations.* Vitamin and mineral mixes were added to diet I. The wheatgerm oil and vitamin E in diets J and K respectively, were added to the wet components before mixing with the dry matter. The vitamin-mixture powders in diets L and M (Table 2) were added to the dry constituents.

*Third alterations.* Miscellaneous supplements were added to diet G. The cholesterol in diet N, was dissolved in 500 mg ether and mixed with the wet components before adding the dry parts. The ether was evaporated off prior to freeze drying. An analysis of the amino acid concentrate in diet P is given in Table 2.

Supplement 3 in diet R consisted of the following components at concentrations of less than 0.01% of wet weight of complete diet, except where percentage composition appears in parentheses: Vitamin A, D, E (0.01%), C, $B_1$, $B_2$, $B_6$, $B_{12}$, nicotinamide (0.02%), biotin, folic acid, pantothenic acid, choline (0.02%), inositol (0.02%) ginseng (0.05%), lecithin, aspartic acid, lysine, arginine, leucine, isoleucine, phenylalanine, threonine, valine, glycine, proline, tryptophan, histidine, methionine, serine, tyrosine, alanine, cystine, glycerophosphates, Na, K, Mn, I, P, Ca, Mg, Zn, Se, S, Cu, Fe.

Supplement 4 in diet S, was similar to supplement 3 in diet R, but also contained mixed fatty acids, mixed sterols and bovine-organ and -gland extract. The pollen in diet T was removed from storage cells in a hive of bees foraging primarily on *Eucalyptus grandis* Hill ex Maiden. The most successful additives used in the study were added in combination to diet B to make diet U.

*Table 2.* Percentage composition of supplements 1 and 2 and amino acid concentrate in diets L, M and P respectively, proportion of wet weight of diet before freeze drying

| Component | % of diet | Component | % of diet | Component | % of diet |
|---|---|---|---|---|---|
| | | Diet L | | | |
| Vitamin A | 0.16 | Biotin | <0.01 | Fe | 0.09 |
| Vitamin D | <0.01 | Ca ascorbate | 1.23 | Mg | 0.15 |
| Thiamine | 0.39 | Choline | 0.39 | Cu | <0.01 |
| Riboflavin | 0.25 | Inositol | 0.20 | Zn | 0.17 |
| Nicotinamide | 0.59 | Benzoic acid | 0.12 | Mn | 0.07 |
| Pantothenic acid | 0.39 | Glutamic acid | 0.12 | K | 0.34 |
| Pyridoxine | 0.25 | Folic acid | <0.01 | Ca | 0.25 |
| Vitamin $B_{12}$ | <0.01 | Lecithin | 0.12 | | |
| Vitamin E | 0.54 | I | <0.01 | | |
| | | Diet M | | | |
| Vitamin $B_1$ | <0.01 | Vitamin C | 0.16 | Biotin | <0.01 |
| Vitamin $B_2$ | <0.01 | Nicotinamide | 0.04 | Valeric acid | 0.09 |
| Vitamin $B_6$ | <0.01 | Pantothenic acid | 0.02 | | |
| Vitamin $B_{12}$ | <0.01 | Folic acid | <0.01 | | |
| | | Diet P | | | |
| Phenylalanine | 0.33 | Lysine | 0.46 | Alanine | 0.42 |
| Leucine | 0.69 | Valine | 0.46 | Glycine | 0.25 |
| Isoleucine | 0.06 | Histidine | 0.18 | Aspartic acid | 0.63 |
| Threonine | 0.21 | Arginine | 0.20 | Serine | 0.23 |
| Methionine | 0.05 | Proline | 0.28 | Glutamic acid | 0.59 |

*Natural prey.* Time from egg hatch to adult eclosion for *C. bipustulatus*, *C. infernalis* and *C. nigritus* were determined when reared on *A. miliaris* on internode sections of bamboo, or on *A. nerii* on butternuts *Cucurbita moschata* cv. Waltham. The percentage survival of immature stages was recorded and live adults were weighed one day after eclosion.

The successful use of artificial substrates for the oviposition of *C. bipustulatus* and *C. nigritus*, made it possible to measure their fecundity. Individual pairs of *C. bipustulatus* were enclosed in rectangular cardboard arenas 106 mm × 30 mm × 30 mm, closed with fine nylon gauze clamped around the arenas with elastic bands. These arenas were attached to the surface of scale bearing bamboo and 20 mm × 20 mm egg pads of frayed linen provided. *C. nigritus* pairs were enclosed in circular plastic collars, 34 mm in diameter and 10 mm high, covered with fine nylon gauze clamped around the collars with elastic bands. These were attached to the surfaces of scale bearing butternuts and 10 mm × 20 mm × 5 mm egg pads of polyester fibre provided. Once a week for three weeks, the arenas were moved to new positions to avoid prey depletion and the egg pads replaced.

## Results

Adult survival, fecundity and immature development on artificial diets, natural prey or without food is given in Table 3 and the diet modifications evluated in Table 4. The mean longevity of *C. nigritus* without food and water was 5.7 days

*Table 3.* Survival, reproduction, fecundity (eggs/pair/7 days), weights and development rate of *C. nigritus* fed on artificial diets, *A. nerii*, *A. miliaris*, or starved with or without water

| Diet | Adult survival for 60 days (%) | Reproducing pairs (%) | Fecundity | Immature survival (%) | Adult weight (mg) | Development time (days) |
|---|---|---|---|---|---|---|
| No food & no water | 0 | 0 | 0 | 0 | – | – |
| No food, + water | 0 | 0 | 0 | 0 | – | – |
| *A. nerii* | 90 | 100 | 32.2 | 84 | 6.6 | 23 |
| *A. miliaris* | 90 | 90 | 31.5 | 85 | 6.8 | 25 |
| A | 70 | 0 | 0 | 0 | – | – |
| B | 90 | 80 | 2.2 | 22 | 4.7 | 29 |
| C | 100 | 20 | 0.4 | 0 | – | – |
| D | 90 | 40 | 1.7 | 0 | – | – |
| E | 90 | 60 | 3.3 | 57 | 4.6 | 28 |
| F | 90 | 60 | 2.4 | 62 | 4.7 | 28 |
| G | 100 | 80 | 6.5 | 55 | 5.0 | 28 |
| H | 90 | 80 | 1.5 | 53 | 4.8 | 28 |
| I | 100 | 100 | 3.7 | 42 | 5.0 | 28 |
| J | 0 | 0 | 0 | 0 | – | – |
| K | 100 | 60 | 3.8 | 44 | 4.6 | 28 |
| L | 20 | 0 | 0 | 0 | – | – |
| M | 90 | 100 | 1.5 | 26 | 5.2 | – |
| N | 100 | 60 | 0.9 | 9 | 3.5 | – |
| O | 100 | 80 | 2.3 | 8 | 4.0 | – |
| P | 90 | 40 | 0.4 | 10 | 3.2 | – |
| Q | 100 | 60 | 2.6 | 3 | 3.9 | – |
| R | 100 | 50 | 4.5 | 33 | 3.9 | – |
| S | 80 | 20 | 0.4 | 19 | 3.3 | – |
| T | 100 | 80 | 1.3 | 20 | 4.6 | – |
| U | 100 | 80 | 2.4 | 46 | 5.6 | 28 |

*Table 4.* Comparisons between artificial diets tested, mean counts for each measurement ranked and the mean rank for each diet calculated

| Measurements | Ranks of measurements | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Initial diets | | | | 1st modifications | | | | | |
| | A | B | C | D | E | F | G | H | I | B |
| Adult survival for 60 days | 4 | 2.5 | 1 | 2.5 | 4.5 | 4.5 | 1.5 | 4.5 | 1.5 | 4.5 |
| No. pairs reproducing | 4 | 1 | 3 | 2 | 5.5 | 5.5 | 3 | 3 | 1 | 3 |
| Fecundity | 4 | 1 | 3 | 2 | 3 | 4 | 1 | 6 | 2 | 5 |
| Immature survival | 3 | 1 | 3 | 3 | 2 | 1 | 3 | 4 | 5 | 6 |
| Adult weight | 3 | 1 | 3 | 3 | 6 | 4.5 | 1.5 | 3 | 1.5 | 4.5 |
| Development rate | 3 | 1 | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 6 |
| Mean | $3.0^{a}$ | $1.3^{b}$ | $2.7^{a}$ | $2.6^{a}$ | $4.0^{ab}$ | $3.8^{ab}$ | $2.2^{a}$ | $3.9^{ab}$ | $2.3^{a}$ | $4.8^{b}$ |
| | 2nd modification | | | | | | | | | |
| | J | | K | | L | | M | | I | B |
| Adult survival for 60 days | 5.5 | | 4.5 | | 5.5 | | 1.5 | | 1.5 | 3 |
| No. pairs reproducing | 5.5 | | 4 | | 5.5 | | 1.5 | | 1.5 | 3 |
| Fecundity | 5.5 | | 1 | | 5.5 | | 4 | | 2 | 3 |
| Immature survival | 5.5 | | 1 | | 5.5 | | 1 | | 2 | 3 |
| Adult weight | 5.5 | | 4 | | 5.5 | | 1 | | 2 | 3 |
| Development rate | 5.5 | | 2 | | 5.5 | | 2 | | 2 | 4 |
| Mean | $5.5^{c}$ | | $2.7^{ab}$ | | $5.5^{c}$ | | $2.2^{ab}$ | | $1.8^{a}$ | $3.3^{b}$ |
| | 3rd modification | | | | | | | | | |
| | N | O | P | Q | R | S | T | G | | |
| Adult survival for 60 days | 3.5 | 3.5 | 7 | 3.5 | 3.5 | 8 | 3.5 | 3.5 | | |
| No. pairs reproducing | 4.5 | 2 | 6.5 | 4.5 | 6.5 | 8 | 2 | 2 | | |
| Fecundity | 6 | 4 | 7.5 | 3 | 2 | 7.5 | 5 | 1 | | |
| Immature survival | 6 | 7 | 5 | 8 | 2 | 4 | 3 | 1 | | |
| Adult weight | 6 | 3 | 8 | 4.5 | 4.5 | 7 | 2 | 1 | | |
| Mean | $5.2^{bc}$ | $3.9^{ab}$ | $6.8^{c}$ | $4.6^{bc}$ | $3.7^{ab}$ | $6.9^{c}$ | $3.1^{ab}$ | $3.1^{ab}$ | | |
| | Most successful diets | | | | | | | | | |
| | B | G | I | U | | | | | | |
| Adult survival for 60 days | 3 | 2 | 2 | 2 | | | | | | |
| No. pairs reproducing | 3 | 3 | 1 | 3 | | | | | | |
| Fecundity | 4 | 1 | 2 | 3 | | | | | | |
| Immature survival | 4 | 1 | 3 | 2 | | | | | | |
| Adult weight | 4 | 2.5 | 2.5 | 1 | | | | | | |
| Development rate | 4 | 2 | 2 | 2 | | | | | | |
| Mean | $3.7^{a}$ | $1.9^{b}$ | $2.1^{b}$ | $2.2^{b}$ | | | | | | |

Absence of a common letter in the superscript indicates a significant difference, Friedman ANOVA, followed by a nonparametric multiple comparison (Siegel & Castellan, 1989), $\alpha = 0.05$.

and 9 days when water was provided. Females survived for 7.2 days and males for 4.4 days without food and water. With water provided, females survived for 10.6 days and males for 7.2 days.

Of diets A, B, C and D screened initialy, diet B gave the best results (Table 4). Diets G and I were the most successful phagostimulatory modifications to diet B. Diets K and M were the most useful of the diets based on diet I with vitamin and mineral additives, but were not superior to diet I itself. Diets O, R and T were the most successful modifications of diet G, but were still not superior to diet G. The highest rate of egg laying was obtained with diet G but was only 20% of the rate when fed on *A. nerii*. Diet U, which incorporated the most useful additives used in this study, was superior to diet B and similar in value to diets G & I. Diet U produced the heaviest adults although these were still 15–18% lighter than when reared on natural prey and immature survival to adult eclosion on this diet was 46% compared with 84% on *A. nerii*. Diet U was therefore the best artificial diet tested for larval rearing and diet G the best for adult maintenance.

Weights of adult *C. bipustulatus* and *C. infernalis* from larvae reared on *A. miliaris* were significantly lower than when reared on *A. nerii* (Table 5). Weights of *C. nigritus* were not significantly different when reared on these two prey species. Duration of immature development of the three *Chilocorus* spp. was not significantly different when reared on these prey species. The rate of oviposition by *C. bipustulatus* and *C. nigritus*, did not differ significantly on the two prey species.

## Discussion

Fecundity and longevity of insects is reduced in below average sized adults (Beddington *et al.*, 1976; Slansky & Rodriguez, 1987). Therefore the production of below average sized adults as a result of rearing on a sub-optimal diet can be equated to a reduction in fitness. Rearing of larvae on these artificial diets produced smaller adults than rearing on natural prey and are therefore still inferior to natural prey. These artificial diets can be valuable as supplements at times of shortage of natural prey, particularly when small quantities of prey are still available for larval rearing. The highest fecundity on an artificial diet was obtained with diet G, making it the most suitable diet for adult maintenance. Diet U supported the highest percentage survival of immatures and the heaviest subsequent adults, making it most suitable for larval rearing. *A. miliaris* and *A. nerii* are both adequate food sources for rearing *C. nigritus* larvae. *A. miliaris* is, however, inferior to *A. nerii* as prey for larvae of *C. bipustulatus* and *C. infernalis* because subsequent adults were smaller than those in a culture maintained on *A. nerii*. This

*Table 5.* Percentage survival of immature stages, weights of adults within 24 h of eclosion and time from egg hatch to eclosion, for *C. bipustulatus*, *C. infernalis* and *C. nigritus*, reared on *A. nerii* and *A. miliaris*

| Predator & prey spp. | % immature survival | Adult weight mean ± 1 SE (n) | Development time (days) mean ± 1 SE (n) | Eggs/pair/day mean ± 1 SE (n) |
|---|---|---|---|---|
| *C. bipustulatus* | | | | |
| *A. nerii* | 69 | $6.7^{a} \pm 0.2$ (31) | $29.0^{a} \pm 0.4$ (33) | $6.2^{a} \pm 0.6$ (12) |
| *A. miliaris* | 73 | $4.4^{b} \pm 0.1$ (29) | $28.7^{a} \pm 0.3$ (29) | $6.0^{a} \pm 0.9$ (12) |
| *C. infernalis* | | | | |
| *A. nerii* | 76 | $11.3^{a} \pm 0.3$ (36) | $26.5^{a} \pm 0.4$ (37) | – |
| *A. miliaris* | 60 | $5.7^{b} \pm 0.2$ (15) | $25.0^{a} \pm 0.5$ (15) | – |
| *C. nigritus* | | | | |
| *A. nerii* | 78 | $6.6^{a} \pm 0.1$ (37) | $22.3^{a} \pm 0.3$ (42) | $4.6^{a} \pm 0.4$ (15) |
| *A. miliaris* | 85 | $6.8^{a} \pm 0.2$ (28) | $23.6^{a} \pm 0.4$ (34) | $4.5^{a} \pm 0.8$ (15) |

Absence of a common letter in the superscript indicates a significant difference, $\alpha = 0.01$, Mann-Whitney U-test, each *Chilocorus* sp. tested separately.

may have important implications for the survival of *C. bipustulatus* and *C. infernalis* in the field in southern Africa, and may in part explain the lack of success in establishment of these species.

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