SCIENTIFIC NOTE

Potential Use of Antibiotic to Improve Performance of Laboratory-Reared *Nezara viridula* (L.) (Heteroptera: Pentatomidae)

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Potencial de Uso de Antibiótico para Melhorar a Performance de *Nezara viridula* (L.) (Heteroptera: Pentatomidae) Criada em Laboratório

RESUMO - O antibiótico estreptomicina adicionado à água de beber na concentração de 125 mg/l acelerou o desenvolvimento ninfal de *Nezara viridula* (L.) (Heteroptera: Pentatomidae), aumentou a sobrevivência e duplicou a longevidade dos adultos, sem afetar a sobrevivência ninfal e o peso dos adultos, quando comparado aos insetos testemunhas. A estreptomicina apresenta potencial para ser utilizada em sistemas de criação do inseto, em especial no tratamento de insetos coletados no campo, reduzindo a introdução de bactérias potencialmente patogênicas na colônia e melhorando a qualidade geral da criação.

PALAVRAS-CHAVE: Percevejo, criação, antimicrobiano

ABSTRACT - The antibiotic streptomycin added to the drinking water at a concentration of 125 mg/l during nymphal development of *Nezara viridula* (L.) (Heteroptera: Pentatomidae)accelerated the development in ca. 2 days, increased survivorship, and doubled adult longevity; nymph survivorship and adult body weight were not affected when compared to control insects. Streptomycin has potential in rearing *N. viridula*, especially in improving quality of field-collected adults, by mitigating the introduction of pathogenic bacteria, and improving the quality of the population.

KEY WORDS: Stink bug, rearing, antimicrobial

The southern green stink bug, *Nezara viridula* (L.) (Heteroptera: Pentatomidae), is a polyphagous pest on several crops of economic importance and is distributed throughout the warmer regions of the world (Todd 1989, Panizzi *et al.* 2000). Because of its importance and wide distribution, colonies of *N. viridula* are kept in several laboratories throughout the world. Usually bugs are fed with combinations of natural diets, such as fresh green bean pods, *Phaseolus vulgaris* L., and raw shelled peanuts, *Arachis hypogaea* (L.) (Harris & Todd 1981, Jones 1985). Mature seeds of soybean, *Glycine max* (L.) Merrill, and sunflower, *Helianthus annuus* (L.), and fruits (berries) of privet, *Ligustrum lucidum* Ait., are also commonly used to rear *N. viridula* (e.g., Corrêa-Ferreira & Panizzi 1999, Vandekerkhove & De Clercq 2004).

Laboratory colonies of *N. viridula* are often revitalized with the introduction of field-collected adults. However, these feral insects, besides carrying larvae of tachinid flies (Corrêa-Ferreira 1984) and trypanosomatids (Sosa-Gómez *et al*. 2005), are also infected with microorganisms, such as bacteria (Hirose *et al*. 2006). Elimination of these microorganisms using antibiotics, added to the water or food, or by injection (Wilkinson 1998), could improve the quality of laboratory-reared populations. Therefore, in this study we added the antibiotic streptomycin (Streptomycin sulfate – 750 units/mg) to the drinking water, and evaluated its effect on nymphal development and survivorship, adult survivorship and longevity, and fresh body weight of *N. viridula*.

Egg masses of *N. viridula* were obtained from a laboratory colony established in cages (50 x 50 x 70 cm) with potted soybean plants, mature soybean seeds, and shelled peanuts. Egg masses were collected on the day of oviposition, placed in petri dishes (9 cm diameter) lined with filter paper, and kept in an environmental chamber (25 ± 1°C, 60 ± 10% RH, photoperiod 14 hL : 10 hD). As 1st instars ecloded, the filter paper was moistened with an antibiotic solution (streptomycin 125 mg/l); 1st instars are known to drink water during their development (Lockwood & Story 1986). On the first day of the 2nd instar, 120 nymphs were removed and

Table 1. Mean (±SE) nymph developmental time (days) of *N. viridula* exposed to a solution of antibiotic (streptomycin - 125 mg/l) or water (control), fed with soybean pod in the laboratory (number of nymphs in parentheses).

Treatments	Instar				Female	Male
	2nd	3rd	4th	5th		
Streptomycin	4.7 ± 0.07 b (51)	3.8 ± 0.12 b (49)	4.9 ± 0.14 b (48)	7.6 ± 0.21 a (47)	21.5 ± 0.50 b (25)	20.3 ± 0.60 a (22)
Control	5.0 ± 0.10 a (53)	4.5 ± 0.15 a (53)	5.4 ± 0.13 a (52)	8.2 ± 0.23 a (46)	23.8 ± 0.65 a (27)	21.8 ± 0.47 a (19)

Means followed by the same letter in each column do not differ significantly using the Tukey test ($P < 0.05$).

individually placed in petri dishes and fed with an immature soybean pod cv. BR 37. Sixty nymphs were exposed to moistened cotton with the antibiotic solution, added to a plastic container (0.3 x 2.8 cm), and 60 nymphs were exposed to cotton with distilled water only (control). *N. viridula* nymphs are known to take up water in the absence or presence of food (Vandekerkhove & De Clercq 2004). Nymphs were placed at random in an environmental chamber and daily observations were made on molting and mortality; soybean pods and wet cotton were replaced every two days.

On the day of emergence, pairs of adults (n = 12) from each treatment were placed in plastic box (11 x 11 x 3.5 cm) lined with filter paper and covered with a lid. In each box was added a soybean pod plus wet cotton with or without antibiotic, as described above. The boxes were distributed at random in an environmental chamber, and daily observations were made. Data on percentage nymph mortality and developmental time, fresh body weight at adult emergence, and percentage adult survivorship and longevity were calculated. Data were submitted to the analysis of variance (ANOVA), and means compared by the Tukey test ($P < 0.05$), using the data analysis program Statistica version 6.0 (StatSoft 2001).

N. viridula survivorship of nymphs (offered streptomycin or not) was similar (ca. 78%). However, developmental time from 1st to 4th instars was faster for nymphs exposed to streptomycin than for those exposed to water only, and no difference was observed during 5th instar development (Table 1). Treated female nymphs took 2.3 days less than control nymphs to complete development, while male nymphs took 1.5 days less than control nymphs (Table 1).

Fresh body weight at adult emergence (female and male) was not significantly different for streptomycin-treated (174.2 and 147.8 mg, respectively) and untreated (control) nymphs (165.0 and 140.7 mg), despite the tendency of greater body weight for the first compared to the later (F values = 1.858 and 1.877).

Females *N. viridula* survivorship up to day 30 was similar for those offered streptomycin or not. After this period, those treated with streptomycin showed 80% survivorship up to day 70, while control females survivorship dropped to 35% at day 35, and down to 0% at day 70 (Fig. 1). A similar trend was observed for males, with streptomycin-treated males showing greater survivorship than control males from day 10 on; all untreated males died at day 55, while 50% of treated males were still alive at this time. Treated-females and males lived almost two fold longer then the untreated ones (Fig. 1).

Our results showing that streptomycin speeds nymphal development and increases adult longevity suggest that this

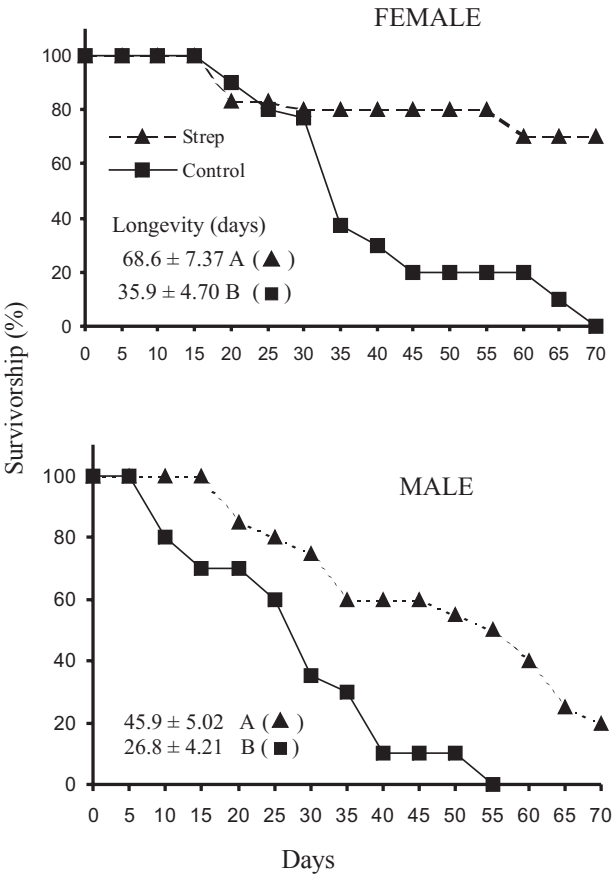

Figure 1. Survivorship (%) and mean (± SE) longevity of *N. viridula* adults, exposed to a solution of antibiotic (streptomycin - 125 mg/l) or water (control), fed with soybean pod in the laboratory. Means followed by the same letter do not differ significantly using the Tukey test ($P < 0.05$).

antibiotic is preventing the occurrence of potentially pathogenic bacteria, without harming the insect. Therefore, streptomycin could be used in laboratory colonies or in mass rearing facilities of *N. viridula*, in particular in treating the colony when feral insects are added. This will avoid the introduction of potentially pathogenic bacteria, and prevent development of resistant populations to the antibiotic.

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