ISSN print: 1224-5984
ISSN online: 2248-3942

Rom Biotechnol Lett. 2020; 25(4): 1877-1885
doi: 10.25083/rbl/25.4/1877.1885

Received for publication, September, 1, 2018
Accepted, December, 19, 2019

Original paper

Analysis of different host plants and identifying non-polluting bioinsecticide strategies to control the invasive pest *Nezara viridula*

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Abstract *Nezara viridula* is one of the newest and most aggressive pests affecting crops and ornamental plants in Romania. The broad polyphagism and invasive character of *N. viridula* necessitates periodic updates of its activities. To assess the status of the host plants, and indirectly, the size of the populations present on them, we analysed several areas at different locations in western Romania. Because large populations have been observed (up to 1264 ind/point) on many plants (the most common hosts being vegetables, corn and ornamental plants), attempts to find solutions to reduce the numbers of these pests have been attempted in a context of environmental friendliness. To find the best pest control solution, a biological bacterial product, *Saccharopolyspora spinosa*, was tested. The results have shown considerable damage to *N. viridula* (up to 98.5%) for many of the analysed sites. The bioinsecticide efficacy was tested, and the percentage of active insects was reduced to less than 30% within 1-14 days. As *N. viridula* has the potential to expand into new areas in the west of the country and also to other areas, and it has the potential to adapt to new host plants; therefore, it is necessary to use a control strategy that involves 1-2 treatments with the biological product tested here, at a concentration of 0.05%.

Keywords *Nezara viridula*, pest, damage, plants, bio-control.

To cite this article: Marcu V, Grozea I. Analysis of different host plants and identifying non-polluting bioinsecticide strategies to control the invasive pest *Nezara viridula*. *Rom Biotechnol Lett*. 2020; 25(4): 1877-1885. DOI: 10.25083/rbl/25.4/1877.1885

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Introduction

Known as the green stink bug, *Nezara viridula* (Hemiptera: Heteroptera: Pentatomidae) is one of the most aggressive species in the genus Nezara (BUSCHMAN, 1980 [1]). Of the 20 species in this genus, it is reported in Romania in the *smaragdulus* form (Fabricius, 1775) with a present status of “present/signalled/area” (MARCU & GROZEA, 2017 [2]; CHIMIȘLIU & DERJANSCHI, 2017 [3]. The geographic distribution of the species at the continental level is noted in Clarke and Walter (1993). This distribution has been monitored at the country/zone level since 1993 (CLARKE & WALTER, 1993 [4]). Since then, centralization and monitoring of the species has been achieved through automated reporting and updating (CABI compendium Invasive Species). The CABI/EPPO reports permanently centralise data regarding the distribution and spread of *N. viridula* in Europe and around the world (CABI/EPPO, 2019 [5]).

Being a tropical species with origins in warm and very hot countries (Ethiopia-Africa) (PANIZZI, 2008 [6]) *N. viridula* has expanded, especially in warmer areas (DAVIS, 1967 [7]; COKL & VIRANT-DOBERLET, 2010 [8]; GUPTA & al, 1993 [9]; TOUGOU & MUSOLIN, 2009 [10]) and also in temperate areas (such as the Eastern European countries) (SQUITIER, 2010 [11]; TILLMAN, 2006 [12]; COLAZZA & 1985 [13]; PROTIC & ROGANOVIC, 2007 [14]; RÉDEI & TORMA, 2003 [15], BARCLAY, 2004 [16]).

The variety of host species for *N. viridula* is extremely large. Smaniotto and Panizzi, in 2015 (SMANIOTTO & PANIZZI, 2015 [17] have centralised botanical families and identified the species on numerous plant species. They found that there is a wide variety of host plants found worldwide, and, among them, legumes from the Fabaceae family are preferred. Plants of the Asteraceae family and those of the Brassicaceae family are also been host, either accidentally or even with reproductive capacity for the insect (VELASCO & WALTER, 1992 [18]. The immature and mature individuals prefer immature seeds and fruits. They are extremely damaging to new leaves and buds. Major damage is caused to soy plants, beans, peas and cotton (DREES & RICE, 1990 [19]. However, they have also been observed feeding on almost all aerial organs of plants (foliage, stems, shoots, buds, flowers, fruits, seeds). Following feeding with floral organs, stink bugs cause abortion and damage to potential seeds (ATTIAH, 1974 [20].

It is a phytophagous species, and feed exclusively on plant tissues and are also important pests for many agricultural, horticultural and landscape crops (MIZELL, 2005 [21].

These insect pests are considered polyphagous because they feed on cultivated and also on wild plants. Although they do not cause damage by consuming wild plants, wild host plants play an important role in increasing the population level of the species. Nutritional resources for nymph development and reproduction of adults are important (PANIZZI, 1997 [22]; CAPINERA, 2001 [23]).

Among the most common control methods is the use of chemical pathways. Given the need for protection of the environment and human health, these classic methods should be replaced by other non-polluting methods. There are some recommendations from companies producing insecticides based on deltamethrin, thiacloprid, and thiamethoxam (individual or mixed) and these formulations are considered to be very effective for controlling both the adult and immature stages. Although these show safe results in control efforts, some researchers recommend alternating the use of chemicals with a non-polluting method (such as the use of parasites) (BROWN & al, 2012 [24]). In control strategies, an important role is played by the plant resistance approach to pest attacks (Buschman, 1980 [1]), but this can be difficult to achieve if we consider the extremely high diversity of hosts and permanent plants. Other non-polluting pathways are related to trap cultures, the use of natural enemies (TILLMAN, 2013 [25]) and bioinsecticides. (SPARKS & al, 2001 [26]; SALGADO, 1998 [27]).

Many methods are effective for fighting the stink bug, *N. viridula*, however the use of biological products (bioinsecticides) may be the best alternative to chemicals.

This insect produces significant economic losses in the agro-horticultural sector and, being newly introduced in our country, it does not have local natural enemies (which makes it more difficult to control). Another aspect that places it in the category of dangerous pests is its accentuated polyphagism and is even considered highly polyphagous (CAPINERA, 2001 [23]), which includes both monocotyledonous and dicotyledonous plants from various anthropic ecosystems. In addition, a national control strategy is missing. That is why, through this work, we want to draw the attention of specialists, farmers and administrators of green spaces and parks, so they are aware of the present level of populations and damages caused by this invasive species, so that we can attempt to control it with environmentally friendly methods.

Materials and Methods

Motivation for choosing the observation area

Considering that in every area analysed so far, the insect has adapted to new host plants, we found it appropriate to approach the area of research and started in the southwestern area of Romania, more precisely in Caras Severin County. It is located in the immediate vicinity of Timis county (where the pest was first reported) (GROZEA & al, 2012, [23]). Thus, this location ensures the existence of basic working material consisting of sufficient pest population levels to carry out research on damage from and testing of control products.

Selection of observation points

The landscape to be analysed includes a wide variety of relief forms, ranging from plains to mountainous areas, and different places were chosen to represent both low and high locations. The selection also considered human population distributions and the existence of vegetable gardens and green spaces. Therefore, the observation locations are the following: Anina, Bocsa, Baile Herculane, Caransebes, Moldova Noua, Oravita, Otelu Rosu and Resita with the following identification data:

Zone 1-RS (PO1): 45°29′30″ N, 21°91′79″ E; (PO2): 45°28′90″ N, 21°93′39″ E; (PO3): 45°29′06″ N, 21°89′04″ E; (PO4): 45°31′17″ N, 21°87′96″ E; (PO5): 45°32′74″ N, 21°87′12″ E.

Zone 2-CS (PO1): 45°42′35″ N, 22°22′30″ E; (PO2): 45°38′94″ N, 22°22′83″ E; (PO3): 45°41′12″ N, 22°21′82″ E; (PO4): 45°41′36″ N, 22°21′49″ E; (PO5): 45°41′54″ N, 22°20′70″ E.

Zone 3-BS (PO1): 45°37′ 89″ N, 21°78′ 42″ E; (PO2): 45°37′53″ N, 21°76′63″ E; (PO3): 45° 37′98″ N, 21°76 ′70″ E; (PO4): 45°37′ 91″ N, 21° 73′ 49″ E; (PO5): 45°36′ 87″ N, 21°70′ 52″ E.

Zone 4-ORA (PO1): 45°02′90″ N, 21°68′57″ E; (PO2): 45°02′90″ N, 21°68′57″ E; (PO3): 45°03′13″ N, 21°69′69″ E; (PO4): 45°03′20″ N, 21°70′59″ E; (PO5): 45°03′65″ N, 21°71′23″ E.

Zone 5-MN (PO1): 44°71′88″ N, 21°62′63″ E; (PO2): 44°73′29″ N, 21°66′16″ E; (PO3): 44°73′59″ N, 21°66′38″ E; (PO4): 44°73′68″ N, 21°66′97″ E; (PO5): 44°72′82″ N, 21°68′38″ E.

Zone 6-OR (PO1): 45°52′59′ N, 22°38′96″ E; (PO2): 45°51′82″ N, 22°35′67″ E; (PO3): 45°51′55″ N, 22°35′24″ E; (PO4): 45°50′90″ N, 22°34′70″ E; (PO5): 45°50′84″ N, 22°34′13″ E.

Zone 7-AN (PO1): 44°88′85″ N, 22°42′41″ E; (PO2): 44°87′88″ N, 22°41′42″ E; (PO3): 44°87′58″ N, 22°41′22″ E; (PO4): 44°86′80″ N, 22°40′65″ E; (PO5): 44°86′06″ N, 22°39′21″ E.

Zone 8-BH (PO1): 44°88′85″ N, 22°42′41″ E; (PO2): 44°87′88″ N, 22°41′42″ E; (PO3): 44°87′58″ N, 22°41′22″ E; (PO4): 44°86′80″ N, 22°40′65″ E; (PO5): 44°86′06″ N, 22°39′21″ E.

Organizing activities

The activities were divided into three categories: assessment of pest population levels (by quantifying the specimens from traps) in the period from 2015-2017; from direct observations on damaged plants (in 2016) (Figure 1) and determining the efficacy of the biological insecticide in the period 2017-2018. Insect monitoring studies and damage assessments were conducted in open fields during plant vegetation periods, with 5 observation points for each of the 8 analysed areas (detailed above). The evaluation of the damages was based on the frequency of the plants attacked; the total plants analysed (20 herbaceous plants were analysed and 1-5 woody plants were also analysed) depended on availability and size (the ornamental trees were of large size and variable number). Research into the efficacy of the biological product was carried out in protected conditions (in the sun and in the laboratory).

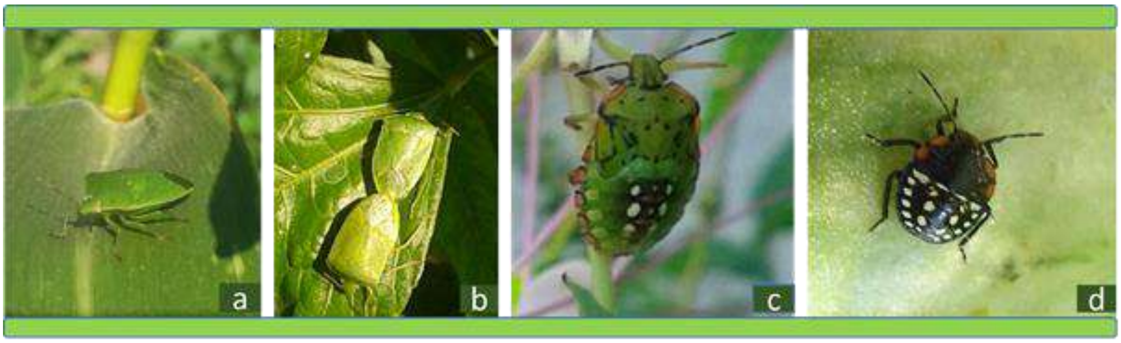

Figure 1. Mature and immature forms of *N. viridula* present in the analysed areas: **a**) a female on a corn leaf (in a private garden); **b**) female and male, completely mature, during the copulative act (on ornamental plants, in green space); **c**) nymph in stage II on an ornamental plant in the park; **d**) larvae in stage III on a tomato fruit in a private garden. In determining the population levels, only the active stages that produce damages (larvae of I-III instar, nymphs of I-II instar and adults as females and males).

The method used for biological control

The tests were conducted on a combination of 3 vegetable species (tomatoes, cucumbers, and beans) from PO4 zone 3, where the highest pest population levels were previously observed. The experimental lot was divided into 3 experimental sections: untreated culture; a treated culture using an available chemical insecticide (Actara 25WG-0.02%) and mentioned by BROWN & al, 2012, [24] as being useful and effective; and a bioinsecticide-treated culture (Laser 240 SC-0.05%). Each variant underwent 4 repetitions (2R in laboratory and 2R in the sun). A total of 20 insects (10 adults + 10 nymphs) under laboratory conditions, in a terrarium (/4T) and 50 insects (25 adults + 25 nymphs) in each sunny location (/4S) were tested yearly. Post-treatment evaluations were performed at 1, 3, 7, 10 and 14 days. The presence of active insects at different intervals from the treatment times was analysed by basic descriptive statistics (using box diagrams). To determine the effectiveness, Abbott's formula was used:

$$P = \frac{Mo - Mk}{100 - Mk} \cdot 100$$

where P is % efficiency (%), Mo is the mean mortality in the treated variants (%), and Mk is the mean untreated mortality (%).

Statistical analysis

The gross data from observations related to population levels, plant damage of and to the efficacy of the biological product relative to the classic product (insecticide) and an untreated culture were statistically interpreted by a biostatistics program. Basic descriptive statistics were as follows: average, minimum, maximum, lower quartile, upper quartile, variance, standard deviation and asymmetry coefficient. For each variable, descriptive statistics/year were made. Additionally, for assessing approximate probabilities, Duncan's multiple range test for the variables studied was used. The Ward method (by a Euclidean diagram) was also used as a criterion applied in the hierarchical analysis and for the differences between variables (MANLY, 2005 [29]).

Results and Discussion

Population levels of the pest during 2015-2017

The results showed that *N. viridula* was present in the monitored county (Caras Severin) at different levels. For some observation areas, no insects were reported, such as in zone 1/Resita, zone 6/Otelu Rosu, zone 5/Moldova Noua and zone 7/Anina, where a value of zero was observed (mean/min./max./=0.0 ind). The situation was different for the other areas studied (Figure 2).

The largest populations (including adults, nymphs and larvae) were observed in 2017 in Zone 3/Bocsa in PO4, with results averaging 118.0 with a maximum of 394.0 ind. and with asymmetry and flattening coefficients of 1.23 and 0.2, respectively. A significance analysis of the observation points showed that there were statistically significant differences between the observation points (PO4/zone 8, PO5/zone 8, PO2/zone 2, PO3/zone 4, PO4/zone 3) and the other observation points analysed, where $p < 0.05$.

In contrast, the smallest insect populations were reported in Zone 1/Resita, where the average value was 9.6 ind., the maximum was 23.0 ind., and the asymmetry/ flattening coefficients were 0.3.

An overall review of the number of insects recorded between 2015-2017 reflects a progressive evolution, and the number of individuals present on plants or crops increased from 557 to 1,264 ind. All the active stages of the insect were observed during the monitoring activities. At times only adults were observed, and at other times, only larvae and nymphs were observed.

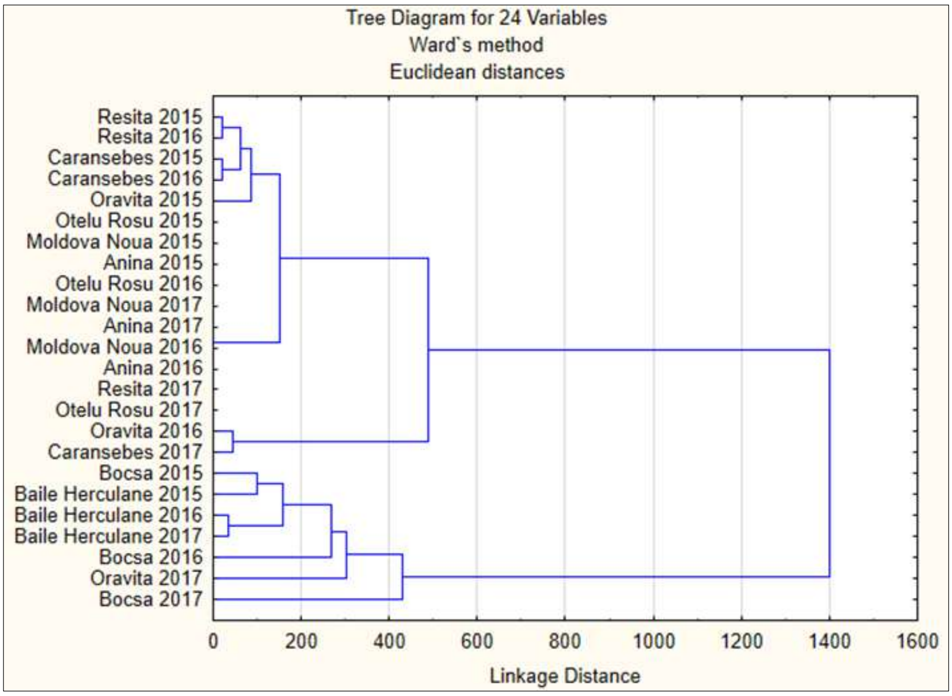

Figure 2. Diagram of the analysis of the population levels of *N. viridula* for the period 2015-2017 in 8 zones in Caras Severin County, western Romania. The cluster-type statistical analysis was performed by Ward's method, which identified from the initial data, similarities and dissimilarities of the variables (number of individuals/traps, zones/localities, year of study). As far as the technique used is concerned, the distances (Euclidean) for pairs of variables were evaluated.

Areas where the insect has not been reported are mountainous at altitudes over 1,000 m; those in which *N. viridula* was reported were hilly and piedmont areas, lowland valleys and plains. High populations were present only in the plains.

Because of the high values (expressed by large populations of Nezara) in Zone 3, it was considered to be the best location to carry out studies to evaluate the efficacy of the biological product.

Identification of host plants and evaluation of attacks on monitored plants

The range of plants monitored during 2016-2017 consisted of plants in the Poaceae family, Leguminosae, Cucurbitaceae, Solanaceae, Rosaceae, Moraceae, Oleaceae and Vitaceae. Their selection was made considering the availability and frequency of the species in the area surveyed. The plant species subjected to damage observations were beans, tomatoes, lilacs, cucumbers, raspberries, lobster claw plants, honeysuckle and vines (in mixed crops of 2, 3 plants or as individual crops). Observations made of the aforementioned plants have shown that all species monitored were found to be damaged, except for the lilac plants of the Oleaceae family.

To assess the damage caused by the green stink bug, an assessment was needed by associating the population level with the set of host plants referring to a point of observation at a given time. Where only 1-5 specimens were present, the damage was slightly visible (small points of discoloration) without noticeably affecting the organs of the attacked plants. By contrast, in the PO where the average pest numbers reached high values (>30 ind.), the damage was evident, and ranged from discoloration to vegetative tissue destruction. The severity of the damage varied from slight to pronounced, depending on the population levels, and on the phenophase of the plants and the month in which the observation was performed (VI-X). The results for 2016 showed that attacked plants were reported in the following observation locations (Figure 3): zone 1 (PO1, x=0.9 ind.), (PO2, x=0.62), (PO3, x= 0.1) and (PO4, x= 0.2); zone 2 (PO2, x=0.8); zone 3 (PO3, x=3.48), (PO4, x=6.86), (PO5, x=0.3); zone 4 (PO1, x=1.58), (PO2, x=4.24), (PO5, x=0.2); zone 8 (PO2, x=0.44), (PO3, x=2.28) and (PO4, x=7.76). Upon analysing the data, it was found that there were statistically significant differences between zone 3 (PO3), (PO4), zone 4 (PO2) and zone 8 (PO4) and all other points with positive values analysed ($p < 0.05$) in terms of the frequency of plants attacked by *Nezara viridula* (Figure 4).

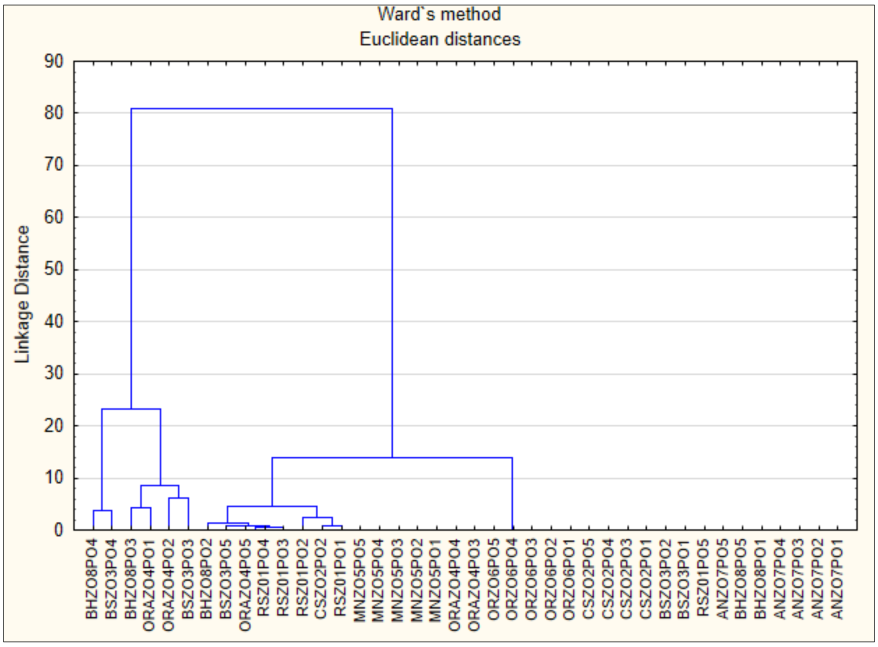

Figure 3. Diagram of analysis of the frequency of plants damaged by *N. viridula* in 2016 in the observation zones. The cluster-type statistical analysis was performed using Ward's method, which identified, similarities and disparities of the variables (total number of plants affected, total number of plants analysed, and zones/localities) from the initial data. As far as the technique used is concerned, the distances (Euclidean) for pairs of variables were evaluated. All zones (8) and observation points (40) were represented, regardless of positive or negative values.

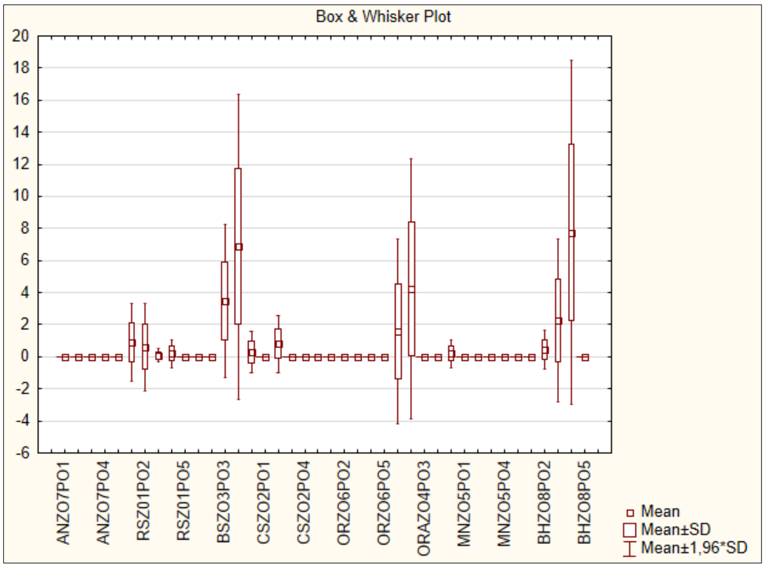

Figure 4. Boxplot diagram that graphically reflects summation of level population and plant frequency values (in zones where the values were positive): minimum, first quartile (or lower quartile), median, third quartile (or upper quartile), and maximum. The chart also shows aberrant values or values located far beyond the distribution. Values were recorded in 2016.

Based on the results of observations made during 2017, the results showed that plants were attacked in the following areas (Figure 5): zone 2 (PO2, x=8.04 ind.), (PO4, x=0.2); zone 3 (PO2, x=5.02), (PO3, x=1.4), (PO4, x=12.46), (PO5, x=12.0); zone 4 (PO3, x=3.36; PO2, x=11.36; PO3, x=3.4, PO4, x=1.88, PO5, x=0.92); zone 6 (PO4, x=20.2); and zone 8 (PO3, x=1.68), (PO4, x=8.38). It was observed that statistically significant differences existed between zone 2 (PO2), zone 3 with PO4 and PO5, zone 4 (PO2), zone 6 (PO4) and zone 8 (PO4) and all other sites (Figure 6) and showed positive values, where p <0.05.

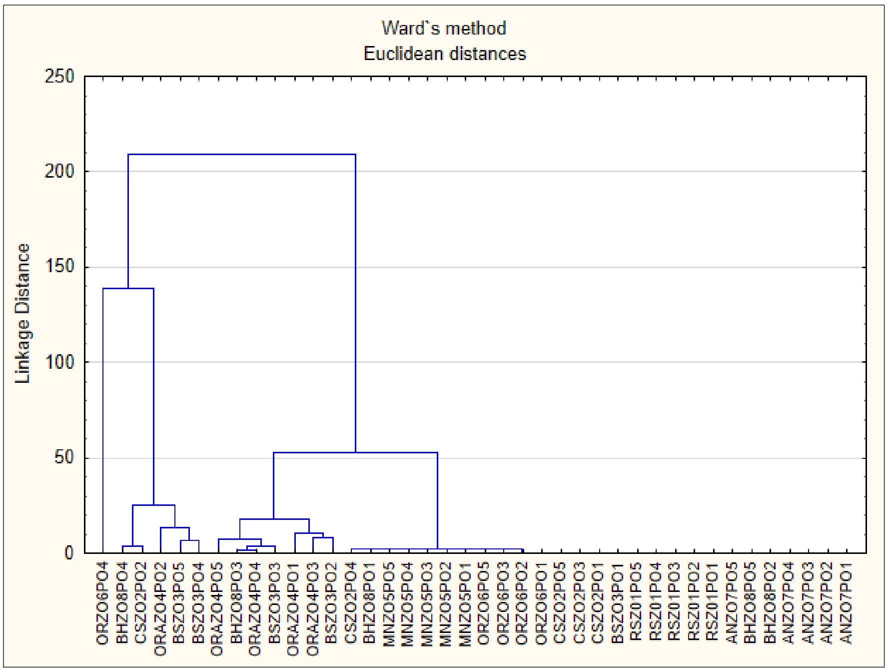

Figure 5. Diagram of the analysis of the frequency of plants damaged by *N. viridula* in 2017 in the observation zones. The cluster-type statistical analysis was performed by Ward's method, which identified from the initial data, similarities and disparities of the variables (total number of plants affected, total number of plants analysed, and zones/localities). As far as the technique used is concerned, the distances (Euclidean) for pairs of variables were evaluated. All zones (8) and observation points (40) were represented, regardless of positive or negative values.

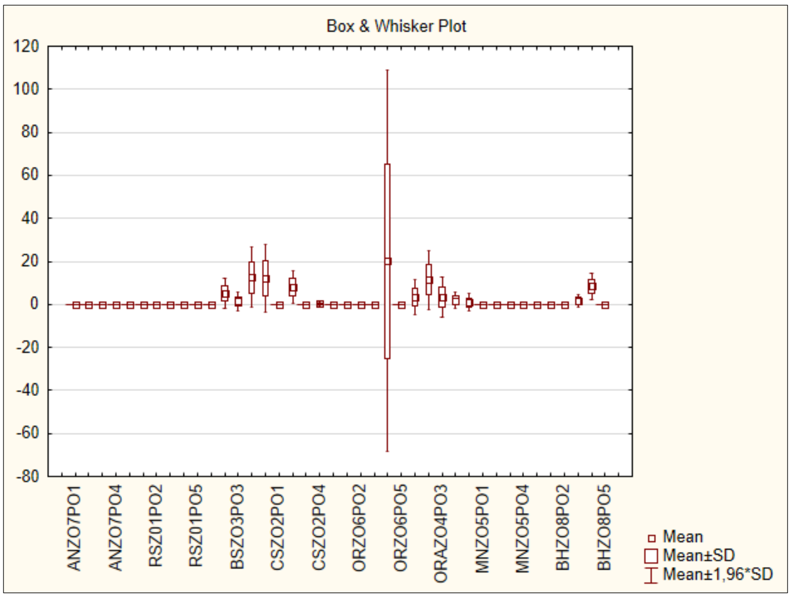

Figure 6. Boxplot diagram that graphically reflects summation of level population and plant frequency values (in zones where the values were positive): minimum, first quartz (or lower quartz), median, third quartz (or upper quartz), and maximum. The chart also shows the aberrant values or values located far beyond the distribution. Values were recorded in 2017.

The most-attacked plant species were tomatoes, cucumbers, beans and corn, when associated with 2 or 3 plant species, and comprised up to 98.5% of the total plants analysed. In zone 3 (PO4) and zone 6 (PO2), the average number of plants attacked among the 20 plants analysed was very high and was close to the maximum. High concentrations of attacked plants were recorded for plant combinations (beans, raspberries and tomatoes) in zone 2 (PO2) and also for individual plants (tomatoes) in zone 4 (PO3) and in zone 8 (PO4).

Average values of approximately 30-50% were recorded for tomato plants and tomatoes, corn and maize combinations, respectively, in zone 4 (PO1) and in zone 3 (PO2). The lowest values were recorded in zone 2 (PO4) in mulberry trees and in zone 4 (PO5) in vines.

Biological control through comparative testing of a bioinsecticide and a classic product

Statistical results (reported in 2017) of control tests, expressed as number of active insects, showed that between the biological treatment variant (Laser 240 SC) 1 day after treatment (BI 1) and the chemical treatment (Actara 25WG) (CI 14) 14 days after treatment, compared with the untreated variant C(U), there were significant differences ($p < 0.05$). Among all the other variants analysed there were visible but statistically insignificant differences, where $p > 0.05$ (Figure 7).

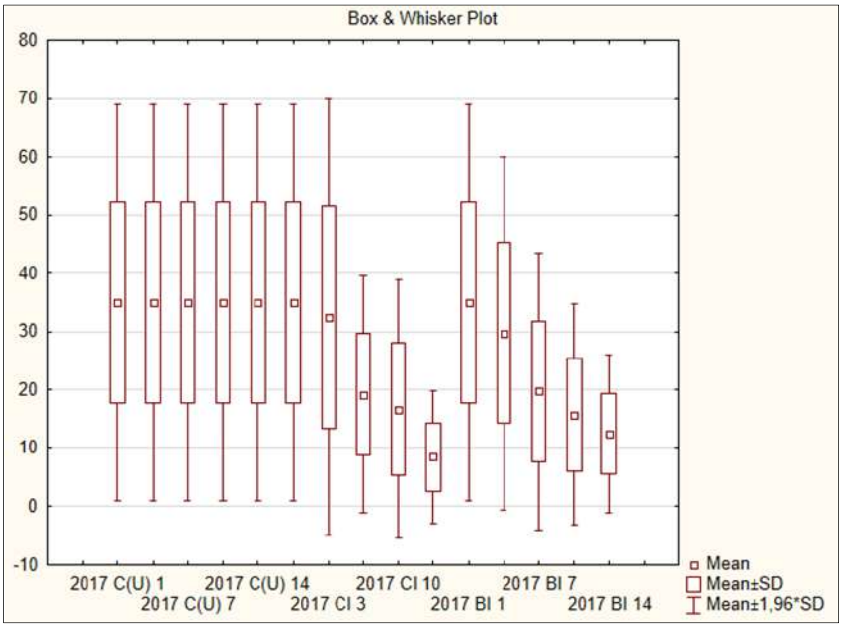

Figure 7. Boxplot diagram that graphically reflects a summary of the control tests (expressed as the number of active insects) after treatments (at intervals of 1, 3, 7, 10 and 14 days), where C(U) is untreated variant, CI is chemical insecticide and BI is bioinsecticide (or biological insecticide). The values analysed reflect the following: minimum, first quartile (or lower quartile), median, third quartile (or upper quartile), and maximum. Values were recorded in 2017.

The efficacy results (percent mortality-%/variance according to Abbott's formula) of the treatments (in 2017) varied depending on the time elapsed from the application of the products used and a mortality rate of 65.25% was attained after application of the biological product (Laser 240 SC) and 77.75% after application of the chemical (Actara 25WG), per the following:

$$P(bioinsecticide) = \frac{65.25 - 0{,}0}{100 - 0.0} x100 = 65.25\%$$

$$P(insecticide) = \frac{77.75 - 0{,}0}{100 - 0.0} x100 = 77.75\%$$

Statistically significant values were also recorded between the 14-day treatment (BI 14), the untreated variant (CU), the chemically treated treatment at 1 day of treatment (CI 1) and the biological variant at 1 day after product application (BI 1) ($p < 0.05$); but there were no statistically significant differences ($p > 0.05$) between the other variants analysed; however, in a raw data analysis, they did show variations (Figure 8).

The mortality/variant of percentage (carried over to 2018) showed an efficacy of 81.75% after application of the biological product (Laser 240 SC) and an efficacy of 70.75% after application of the chemical (Actara 25WG), as follows:

$$P(bioinsecticide) = \frac{70.75 - 0{,}0}{100 - 0.0} x100 = 70.75\%$$

$$P\ (insecticide) = \frac{81.75 - 0{,}0}{100 - 0.0} x100 = 81.75\%$$

Both treatment variants showed values higher than 64% (up to 85%) for a single treatment in August, which confirms the effectiveness of both products for population reduction of the species, especially if performing at least 2 treatments at 3-week intervals is considered.

The higher efficacy expressed for green stink bug mortality and, indirectly, the presence of active stages was recorded in the chemical variant when compared to the biological product variant.

The difference between the two treatments, is primarily balanced by the potential for harm; so, the wisdom of choosing between a polluting treatment (with an efficacy of 85%) and a non-polluting treatment with an efficacy of up to 70% is questionable.

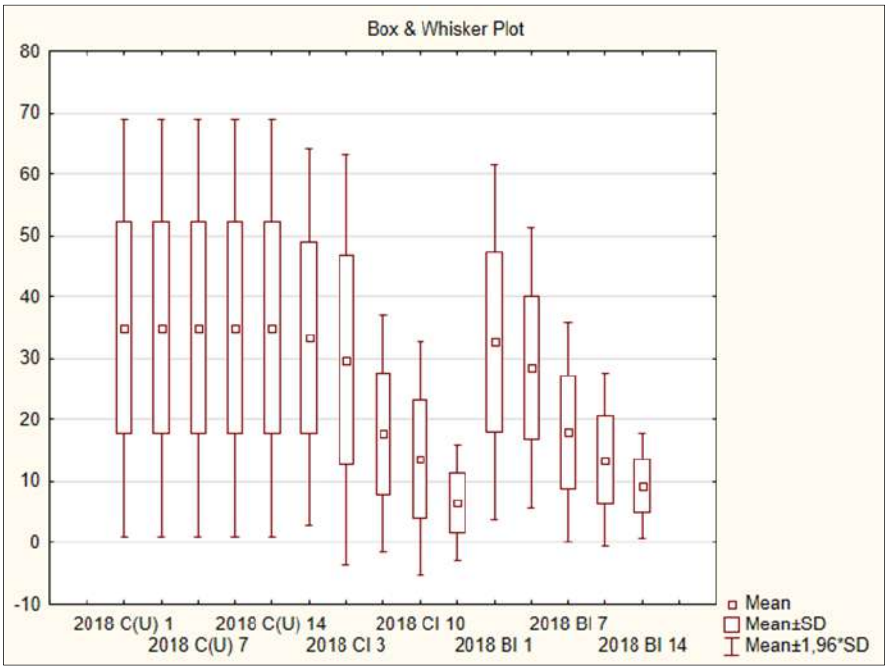

Figure 8. Boxplot diagram that graphically reflects a summary of the control tests (expressed as number of active insects) after treatments (at intervals of 1, 3, 7, 10 and 14 days), where C(U) is untreated variant, CI is chemical insecticide and BI is bioinsecticide (or biological insecticide). The values analysed reflected the following: minimum, first quartile (or lower quartile), median, third quartile (or upper quartile), and maximum. Values were recorded in 2018.

Conclusions

Based on the above, it can be concluded that the species *N. viridula* is present in a variety of areas, both in vegetable gardens and in green spaces, and it usually occurs in high populations if there are mixtures of plants (3 species of plants or more).

The maximum number of insects were observed on tomatoes that were in areas where other species of vegetables, ornamental plants or other crop plants (such as corn) were present. There is no definitive interpretation of the cause of the attractiveness of a particular plant, but we can create a hierarchy of preferences for a given time, in a certain area, and for a range of available plants (as was the case with the present research work). Thus, we note the attractiveness of solanaceae, cucurbitaceae, legumes, poaceae and rosaceae.

The damage inflicted by *N. viridula* may compromise some vegetable crops or ornamental plant species. As Harris and Todd (1980) [30] have mentioned, this damage is severely harmed, especially the new shoots and fruits of the plants; from the present observations it has also been shown that *N. viridula* also affects leaves and reproductive organs.

To limit its extent and to avoid damage, treatments with high-performance products are necessary because this pest is a species with great potential for adapting to new conditions.

It is obvious at present that it is necessary to reorient pest control strategies to favour bioinsecticides to reduce environmental pollution and to protect human health. Through our results, we have demonstrated that the tested bioinsecticide is effective for significantly reducing the population of active insects.

Abbreviations

PO means observation point, C(U) is untreated variant, CI is chemical insecticide and BI means bioinsecticide.

Acknowledgments

We would like to express our gratitude to all of the City Halls of the localities where observations were made, and garden owners and green space administrators for giving us access. Parts of the research in this paper are the results of the first author's doctoral studies in collaboration with the PhD coordinator (corresponding author).

Conflict of interest disclosure

There are no known conflicts of interest in the publication of this article. The manuscript was read and approved by all authors.

References

1. BUSCHMAN LL. Parasites of *Nezara viridula* (Hemiptera: Pentatomidae) and other Hemiptera in Florida. *Flor. Entomol*. 1980; 63: 154-162.
2. MARCU VC, GROZEA I. Assesment of population level of the invazive species *Nezara viridula* in diferent crops from south west of Romania. *Lucr. St. Ser-Agro*. 2017; 60: 175-178.
3. CHIMIȘLIU C, DERJANSCHI V. New data regarding the species *Nezara Viridula* (Linnaeus, 1758) (Heteroptera, Pentatomidae) in Oltenia Fauna (Romania). *Bul Stiint Rev Et, St Nat Muz.* 2017; 26 (39): 12-14.
4. CLARKE AR, WALTER GH. Biological control of the green vegetable bug *Nezara viridula* (L.) in eastern Australia: Current status and perspectives. In: Corey SA, Dall DJ, Milne WM, eds. Pest control and sustainable agriculture. Melbourne, Australia, CSIRO. 1993; 223-225.
5. CABI/EPPO. Invasive Species Compendium. Detailed coverage of invasive species threatening livelihoods and the environment worldwide; *Nezara viridula* (green stink bug); 2019 (https://www.cabi.org/isc/datasheet/36282).
6. PANIZZI AR. Southern green stink bug, *Nezara viridula* (L.) (Hemiptera: Heteroptera: Pentatomidae); *Encycl Entomol*. 2008; 3471-3471.
7. DAVIS CJ. Progress in the Biological Control of the Southern Green Stink Bug, *Nezara viridula* variety smaragdula (Fabricus) in Hawaii (Heteroptera: Pentatomidae). *Mushi*. 1967; 39: 9-16.
8. COKL A, VIRANT-DOBERLET M. Communication Through the Substrate as a Solution for Small Plant-dwelling Insects. *2nd Pan-American/Iberian Meeting on Acoustics*, Cancun, Mexico. 2010.
9. GUPTA SP, PRAKASH A, RAO J, GUPTA A. Qualitative losses of paddy grains due to bugs in the farmers fields of Coastal Orissa. *Ind. J. of Entomol*. 1993; 55(3): 229/236.
10. TOUGOU D, MUSOLIN DL. Rapid climate change promotes changes in distribution ranges of *Nezara viridula* and *Nezara antennata* in Japan. *Entomol. Experim. Appl.* 2009; 130(3):249-258 Doi: 10.1111/j.1570-7458.2008.00818.x.
11. SQUITIER JM. Featured creature fact sheets: southern green stink bug, Nezara viridula (Linnaeus) (Insecta: Hemiptera: Pentatomidae). *EENY-016*, 2010; University of Florida.
12. TILLMAN GPG. Sorghum as a trap crop for Nezara viridula L. (Heteroptera: Pentatomidae) in cotton in the Southern United States. *Environ. Entomol.*, 2006; 35: 771-783.
13. COLAZZA S, CIRICIOFOLO E, BIN F. First observations on damage by Nezara viridula (L.) (Het., Pentatomidae) to soyabean in Central Italy. Atti XIV Congresso Nazionale Italiano di Entomologia sotto gli auspici dell'Accademia Nazionale Italiana di Entomologia, 28 mag.-1 giu.1985 Palermo, Italy; *Accad. Naz. Ital. Entomol*. 1985; 371/ 378.
14. PROTIC L, ROGANOVIC D. Heteroptera on Cupressus sempervirens (Linnaeus) in Montenegro (State community of Serbia and Montenegro). *Acta Entomol. Serb*. 2007; (1/2):17-27.
15. RÉDEI D, TORMA A. Occurrence of the Southern green stink bug, Nezara viridula (Heteroptera: Pentatomidae) in Hungary. *Acta Phytopath. Entomol. Hung*. 2003; 38(3/4):365-367.
16. BARCLAY MVL. The green vegetable bug Nezara viridula (L.,1758) (Hem.:Pentatomidae) new to Britain. *Entomologist's Record*, 2004; 116: 55-58.
17. SMANIOTTO LF, PANIZZI AR. Interactions of Selected Species of Stink Bugs (Hemiptera: Heteroptera: Pentatomidae) from Leguminous Crops with Plants in the Neotropics. *Florida Entomol*. 2015; 98(1):7-17. Doi: http://dx.doi.org/10.1653/024.098.0103.
18. VELASCO LRI, WALTER GH. Availability of different host plant species and hanging abundance of the polyphagous bug *Nezara viridula* (Hemiptera: Pentatomidae). *Environm. Entomol.* 1992; 21(4): 751- 759.
19. DREES BM, RICE ME. Population dynamics and seasonal occurrence of soybean insect pests in southeastern Texas. *Southw. Entomol.* 1990; 15(1):49-56.
20. ATTIAH HH, ISA AL, ROFAIL F. An outbreak of Nezara viridula on citrus trees in Egypt. *Agricultural Research Review*, 1974; 52: 81.
21. MIZELL RF. Stink bugs and leaffooted bugs are important fruit, nut, seed and vegetable pests. *EDIS. UF/IFAS. ENY-718*, 2005.
22. PANIZZI AR. Wild Hosts Of Pentatomids:Ecological Significance and Role in Their Pest Status on Crops. *Ann. Rev. Ento.*, 1997; 42:99-122.
23. CAPINERA JL. Handbook of Vegetable Pests. Acad. Press, San Diego. 2001; 729 p.
24. BROWN SA, DAVIS JA, RICHTER AP. Efficacy of foliar insecticides on eggs of Nezara viridula (Hemiptera: Pentatomidae). *Flor. Entomol.*, 2012; 95: 1182-1186.
25. TILLMAN GPG. Stink Bugs (Heteroptera: Pentatomidae) and their Natural Enemies in Alfalfa in South Georgia. *J Entomol Sci.*, 2013; 48(1):1-8. Doi: http://dx.doi.org/10.18474/0749-8004-48.1.1.
26. SPARKS T, GARY DC, DURST G. Natural products as insecticides: the biology, biochemistry and quantitative structure-activity relationships of spinosyns and spinosoids. *Pest Manag Sci.*, 2001:57.
27. SALGADO VL. Studies on the mode of action of spinosad: Insect symptoms and physiological correlates. *Pest. Biochem. Physiol.*, 1998; 60: 91-102.
28. GROZEA I, ŞTEF R, VÎRTEIU AM, CĂRĂBEŢ A, MOLNAR L. Southern green stink bugs (Nezara viridula L.) a new pest of tomato crops in western Romania. *Res. J. Agric. Sci.* 2012; 44 (2): 24-28.
29. MANLY BFJ. Multivariate Statistical Methods: A primer. Third edition, Chapman and Hall. 2005.
30. HARRIS VE, TODD JW. Imature stages of the green stink bug, *Nezara Viridula* (L.). *J. G. Entomol. Soc.*, 1980b; 15(2): 109-114.