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Research Article

Sublethal Effects of *Solanum tuberosum* and *Lycopersicon esculentum* Leaf Extracts on *Tenebrio molitor* and *Harmonia axyridis*

Solanum tuberosum ve Lycopersicon esculentum Yaprak Özütlerinin Tenebrio molitor ve Harmonia axyridis Üzerine Subletal Etkisi

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Abstract

Repellent or attractant activity of extracts obtained from *Solanum tuberosum* and *Lycopersicon esculentum* leaves were tested against *Tenebrio molitor* (Coleoptera: Tenebrionidae) and *Harmonia axyridis* (Coleoptera: Coccinellidae). Different developmental stages of the insects (larvae of different age and imagoes) were exposed to the extracts which is present in the substratum. Extracts were used in different concentrations of 1, 10, 100 or 1000 ppm. Distilled water was used as control. In this study, we observed migration of the insects towards the exposed area especially with lower concentrations of extracts which is taken as attractant activity. In other cases, insects showed migration towards control areas which is taken repellent activity. The reaction differed between both insect species, as well as between younger, older larvae and adults of *T. molitor*. Therefore we infer from this study that both plants may be a source of substances which will be used in insect pest management for plant protection.

Keywords: Botanical insecticides, *Solanum tuberosum, Lycopersicon esculentum,* Leaf extract, *Tenebrio molitor, Harmonia axyridis*

Öz

Solanum tuberosum and *Lycopersicon esculentum* bitkilerinin yapraklarından elde edilen özütlerin *Tenebrio molitor* (Coleoptera: Tenebrionidae) ve *Harmonia axyridis* (Coleoptera: Coccinellidae) böceklerine karşı uzaklaştırıcı ve çekici aktiviteleri denendi. Böceklerin farklı gelişme evrelerindeki (farklı evrelerdeki larvalar ve erginler) bireyler bir petri kutusu içerisinde bulunan filter kağıdına emdirilmiş özütlere maruz bırakıldı. Elde edilen özütler 1, 10, 100 ve 1000 ppm konsantrasyonlarda denendi. Saf suya emdirilmiş filtre kağıtları ise kontrol grubu olarak kullanıldı. Çalışmada özellikle düşük konsantrasyonlardaki özütlerin uygulandığı alanlara doğru böceklerin yöneldiği görüldü (çekici etki). Bazı zamanlarda ise böceklerin özütlerden ayrılarak kontrol alanına doğru göç ettiği (uzaklaştırıcı etki) belirlendi. Bu tepkiler *T. molitor*'un gelişme evrelerine göre farklılık göstermekle beraber her iki böcek türüne göre de değişiklik gösterdi. Bu çalışma her iki bitkiden elde edilen özütlerdeki maddelerin zararlı böceklere karşı bitkileri korumak amacıyla kullanılabileceğini göstermektedir.

Anahtar Kelimeler: Bitkisel insektisitler, *Solanum tuberosum, Lycopersicon esculentum* Yaprak özütü, *Tenebrio molitor, Harmonia axyridis*

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1. Introduction

Crop protection demands various strategies, which complement each other. Modern crop protection strategies involve usage of natural enemies of pests, rotation of crops, mechanical control of pests and usage of new formulations, including plant and animal toxins (Chowanski et al. 2014). Such managements leads to decreased usage of toxic and unspecific synthetic pesticides. Nowadays, natural substances are more and more frequently used against pests. These compounds, including secondary plant metabolites, show antimicrobial (Aygan et al. 2011, Mesiani et al. 2015), antifungal (Cantelli et al. 2014) and zoocidal activity (Ntalli et al. 2014, Adamski et al. 2014). Moreover, they also deter herbivores from eating plant parts. Therefore, high concentrations of these substances may act as repellent. On the contrary, their low concentrations may attract herbivores. Hence, no simple, concentration–dependent reaction can be observed. Furthermore, bioaccumulation may also influence insects' reaction.

In the series of the previous reports we showed that Solanaceae plants produce substances which can be used in crop protection (Adamski et al. 2014, Ventrella et al. 2015). Although they may not cause acute lethality of pests, several sublethal effects were observed, for example decreased fertility, lower hatching success or various morphological malformations (Büyükgüzel et al. 2013, Ntalli et al. 2014). These effects decrease number of individuals of pests inhabiting agrocenoses. In consequence, lower number of pests may be overcome by predators, mechanically destroyed or killed with the use of lower amount of insecticides introduced into environment. Hence, usage of natural substances with repellent/attractant activity is amongst the most important strategies of Integrated Pest Management. Such substances may cause intensive emigration of pests from agrocenoses. Therefore, they increase crop yield without causing serious pollution.

Repellent or attractant activity of natural plant products is extremely interesting in case of stored products, where chemical insecticides frequently cannot be used. In such a situation repellency or attractant activity, combined with traps against insects, may successfully limit presence of pests and crop losses. Also extracts or pure alkaloids obtained from plants may be used against pests which do not feed on a given species (Marciniak et al. 2010). Since they had not been exposed to these substances previously, they had no chance to develop resistance, neither on the physiological level, nor due to the genetic selection.

Natural substances may also play important role in management of invasive insect species. Contrary to those species, which inhabit the environment for many decades, invasive species may not evolve resistance against substances obtained from plants present in their new environment. Hence, natural substances may be a very important tool in plant protection against such species.

In the present paper we show the results of our preliminary research on the possible repellent or attractant activity of extracts obtained from leaves of two economically important and widely harvested plants, namely potato (*Solanum tuberosum*) and tomato (*Lycopersicon esculentum*). Leaves of both species are off the agronomic value, they are not used as a food. Therefore, they are cheap source of substances, which can be used as natural pesticides. We examined the effect of leaf extracts against two economically important insect species: omnivorous yellow mealworm (*Tenebrio molitor* L.) and predatory harlequin ladybird (*Harmonia axyridis* (Pallas)). The first one is a common pest of stored products, while the second one is regarded as one of the most important invasive species, worldwide. As an aggressive predator, it may alter the ecological balance within ecosystems. It is also known as an economic pest of fruit production and processing, especially of wine grapes (Koch 2003, Hutchison et al. 2010, Brown et al. 2011).

2. Material and Methods

2.1 Insects

Larvae and adults of *T. molitor* were obtained from a culture maintained in the Department of Animal Physiology and Development, as described previously (Rosinski et al.1979). These insects do not have fixed number of larval instars, the duration of larval stages may vary. Therefore, young (40mg) and older (150mg) larvae and 2 day-old adults were selected for the experiment. Adults of *H. axyridis* were collected in the North of Poznań in the end of September 2013. Dozens of individuals from natural environment were aggregating on buildings' walls, searching for overwintering locations.

2.2. Experimental procedures

Freshly collected potato and tomato leaves were collected and prepared as previously described (Cataldi et al. 2005, Adamski et al. 2014). The obtained extracts were diluted in distilled water to obtain final concentrations of 1, 10, 100 or 1000 ppm. Bottoms of the petri dishes (10cm in diameter) were inlayed with two semicircles made of a filter paper. One of them was soaked with 0.5ml of the tested extract concentration. The second half was soaked with water. After that, 10 larvae or adults of the tested species were placed in the middle of the petri dish. To observe the short time and long time effects, the location of specimens on both halves was noted after an hour and after 24 hours. Each experiment was repeated four times. The attractant/repellent activity was evaluated by calculating the number of individuals inhabiting both parts of experimental fields (extract-soaked and control). Then equations of effect-concentration lines, correlation between number of insects on extract-soaked substratum, EC_{50} (concentrations causing effect for 50% of population) values were determined using probit analysis (Finney, 1971):

Probit of mortality = logA+ Bxlog (concentration);

where:

A - control mortality (i.e. the value on the Y axis, where the line of mortality crosses it);

B - the slope of the probit line, (this value shows how fast toxicity raises, if toxicity raises significantly with a low increase of concentration, B has high values).

To calculate significance of observed results, chi-square test was used, at a significance level of 0.05.

3. Results

Effects caused by both tested extracts varied according to concentration, time of exposure and exposed species/stage. After 1h of exposure of *S. tuberosum* leaf extract to young *T. molitor* larvae and adults, both tested species showed concentration-dependent trend of distribution and high correlation between extract concentration and location of insects (Table 1). While significantly higher number of individuals grouped on the extract-soaked part of the experimental area at the lowest used concentration, the highest concentration was inhabited by less than 50% of individuals. Although there was no significant difference

Table 1. Effect of tested extracts on *T. molitor* and *H. axyridis*

ppm	*T. molitor* 40 mg larvae	*T. molitor* 150 mg larvae	*T. molitor* adults	*H. axyridis* adults
S. tuberosum extract, 1h				
Correlation coefficient	-0.97	0.56	-0.77	-0.97
Equation	Y = 4.208 + 0.357 x log conc	Y = 4.998 – 0.115 x log conc	Y = 3.898 + 0.381 x log conc	Y = 3.445 + 0.773 x log conc
IC_{50}/95 % confidence interval/	0.164 x 10^3 /0.64 x 10^2 – 0.42 x 10^3/	0.964 /0.15 x 10^{-1} – 0.63 x 10^2/	0.784 x 10^3 /0.24 x 10^3 – 0.26 x 10^4/	0.103 x 10^3 /0.66 x 10^2 – 0.16 x 10^3/
S. tuberosum extract. 24h				
Correlation coefficient	-0.98	-0.29	-0.98	0.94
Equation	Y = 4.428 + 0.443 x log conc	Y = 4.777 + 0.094 x log conc	Y = 4.675 + 0.368 x log conc	Y = 6.265 - 0.180 x log conc
IC_{50}	0.185 x 10^2 /0.99 x 10^1 – 0.38 x 10^2/	0.239 x 10^3 /0.49 x 10^1 – 0.12 x 10^5/	0.768 x 10^1 /0.32 x 10^1 – 0.19 x 10^2/	0.129 x 10^2 /0.90 x 10^1 – 0.18 x 10^2/
L. esculentum extract. 1h				
Correlation coefficient	-0.98	0.95	-0.90	0.74
Equation	Y = 4.448 + 0.457 x log conc	Y = 5.65 + 0.279 x log conc	Y = 4.120 + 0.696 x log conc	Y = 5.238 - 0.091 x log conc
IC_{50}	0.162 x 10^2 /0.83 x 10^1 – 0.32 x 10^2/	0.215 x 10^3 /0.60 x 10^2 – 0.78 x 10^2/	0.184 x 10^2 /0.11 x 10^2 – 0.30 x 10^2/	0.410 x 10^3 /0.51 x 10^1 – 0.33 x 10^5/
L. esculentum extract. 24h				
Correlation coefficient	-0.87	-0.87	-0.94	-0.97
Equation	Y = 5.008 + 0.122 x log conc	Y = 4.044 + 0.602 x log conc	Y = 2.070 + 1.816 x log conc	Y = 4.246 + 0.890 x log conc
IC_{50}	0.867 /0.16 x 10^{-1} – 0.47 x 10^2/	0.387 x 10^2 /0.23 x 10^2 – 0.65 x 10^2/	0.411 x 10^2 /0.33 x 10^2 – 0.51 x 10^2/	0.704 x 10^1 /0.47 x 10^1 – 0.11 x 10^4/

between exposed and control areas within groups exposed to leaf extracts in concentrations of 10 and 100ppm, the insects migrated toward areas soaked with low concentrations of extracts. Statistically significant lower number of individuals was noted for *H. axyridis* individuals at 1000ppm, too. On the other hand, there was no clear correlation between extract concentration and location of older larvae (Figure 1). Longer exposure caused significant increases of individuals of *H. axyridis* adults on extract-soaked areas, at 100 and 1000ppm. On the other hand, the significantly lower number of ladybirds inhabited substratum soaked with the lowest concentration. That resulted in very high positive correlation between extract concentration and number of *H. axyridis* (Table 1). Reverse effects (negative correlation) were observed in case of young mealworms' larvae and their adults, although differences were not statistically significant. 150mg larvae did not show any correlation (Table 1, Figure 2).

Short exposure of tomato extracts resulted in clear negative correlation what was observed for young *T. molitor* larvae and adult insects (Table 1). Opposite trend was observed for bigger mealworms and ladybirds (Figure 3). On the other hand, longer exposure resulted in high negative correlation for all tested groups, although sometimes we did not observe statistically significant changes. Interestingly, for both exposure time to 1 ppm significantly attracted *T. molitor* adults, whereas the highest concentration caused significantly lower number insects inhabiting this part of the experimental area (Table 1, Figure 4).

We did not observe acute toxicity against any of the tested groups. Probit analysis for repellent or attractant activity revealed B values lower than 1.0 in almost all cases. Only for *T. molitor adults,* treated *L. esculentum* extracts for 24h the value was higher (1.816).

4. Discussion

Although natural insecticides, especially plant extracts, may show lower acute toxicity than the synthetic ones, they are potentially crucial for plant protection. First of all, they are cheaper to obtain, than the commercial pesticides (Amoabeng et al., 2014). Next, due to their lower lethality, non-target species will not suffer so much of them. Third, they have lower persistence in the environment, than synthetic substances (Buss and Park Brown, 2014; Chowanski et al., 2014). Lastly, they often act as repellents or attractants. Therefore, they may be used in traps (attractants) or they may force pests to migrate from the exposed area, without changing the number of individuals within the population (repellents), what may significantly affect other trophic levels.

The acute toxicity of tested extracts was observed. B values lower than 1.0 show also, that the toxicity rises slowly. In

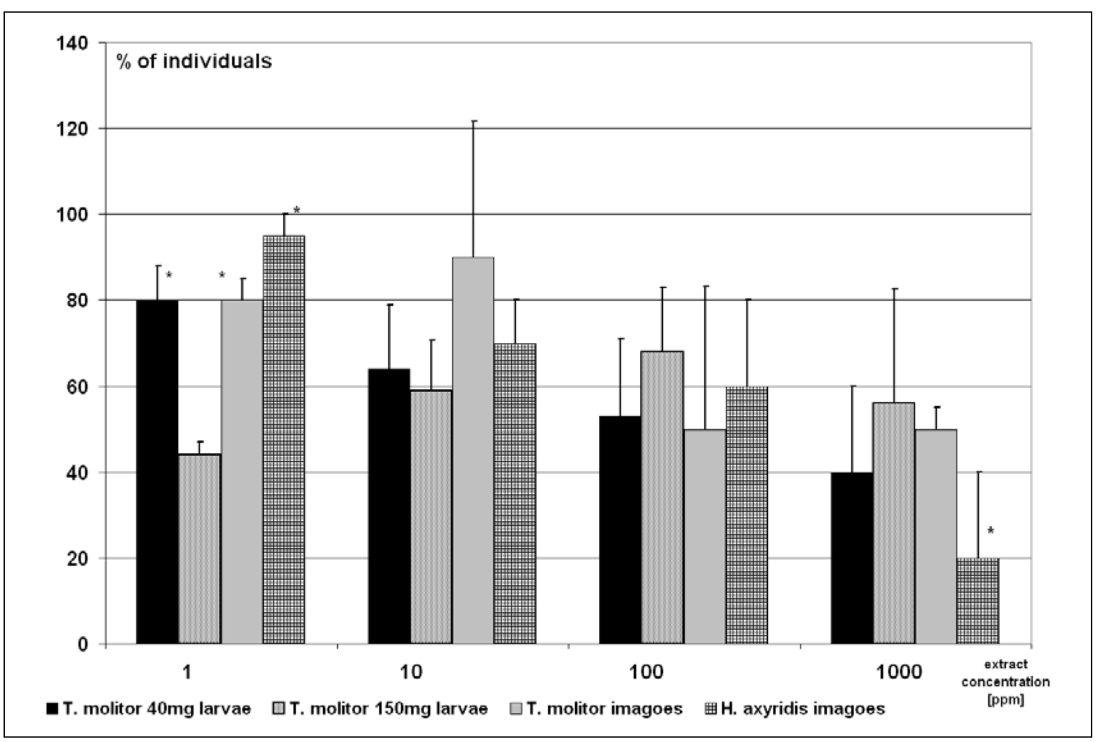

Figure 1. Percentage of insects present at the surface exposed to *S. tuberosum* leaf extract, after 1h of exposure. Data are means ± SD. * statisticall significant results, at a significance level of 0.05.

the other words, it seems to be difficult to obtain such a concentration of extracts, which would be lethally toxic to tested insects. Nevertheless, our study shows that extracts obtained from Solanaceae plants are a promising source of substances of sublethal, repellent and attractant activity. The response varied due to species, life stage of the insect, time of exposure as well as used concentration of extracts. Interestingly, the response of young *T. molitor* larvae and adults during 1 h exposure of *S. esculentum* extract was similar. They both showed tendency to prefer the areas soaked with

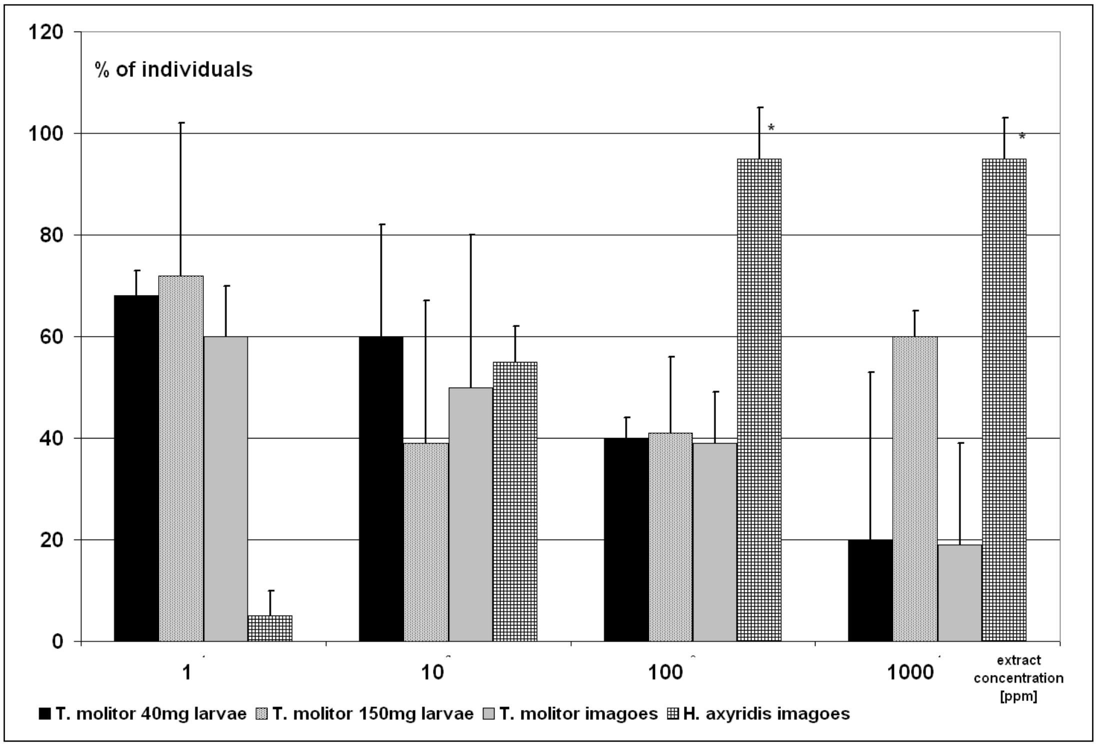

Figure 2. Percentage of insects present at the surface exposed to *S. tuberosum* leaf extract, after 24h of exposure. Data are means ± SD. * statisticall significant results, at a significance level of 0.05.

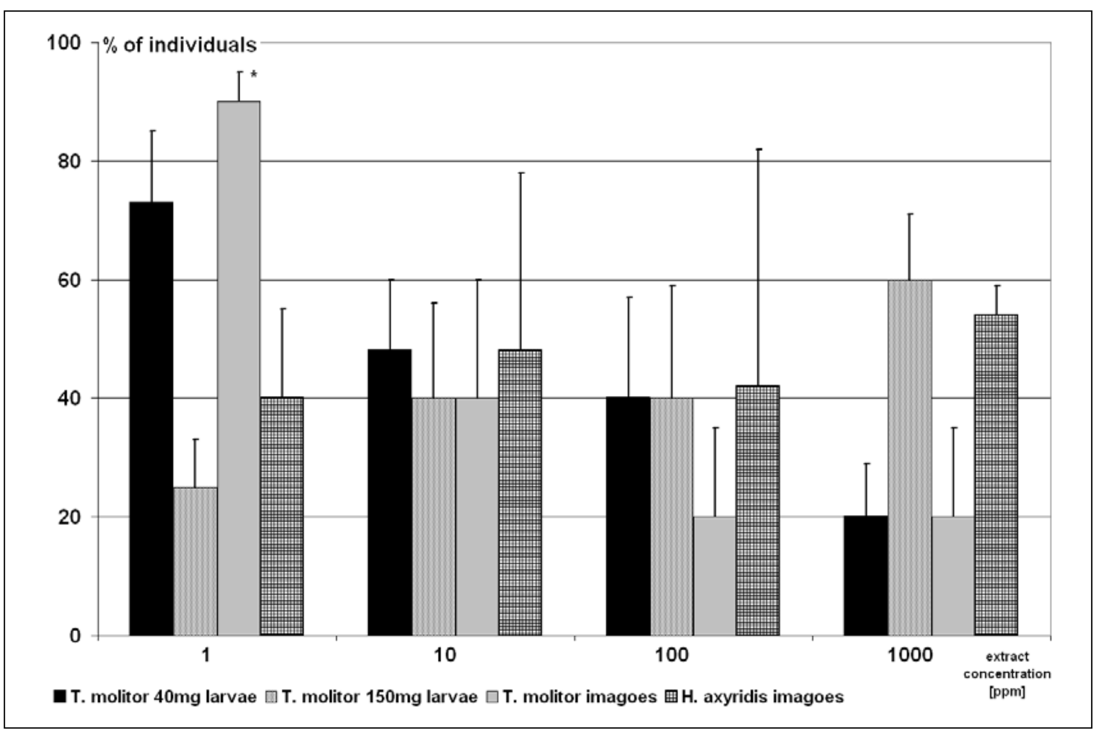

Figure 3. Percentage of insects present at the surface exposed to *L. esculentum* leaf extract, after 1h of exposure. Data are means ± SD. * statisticall significant results, at a significance level of 0.05.

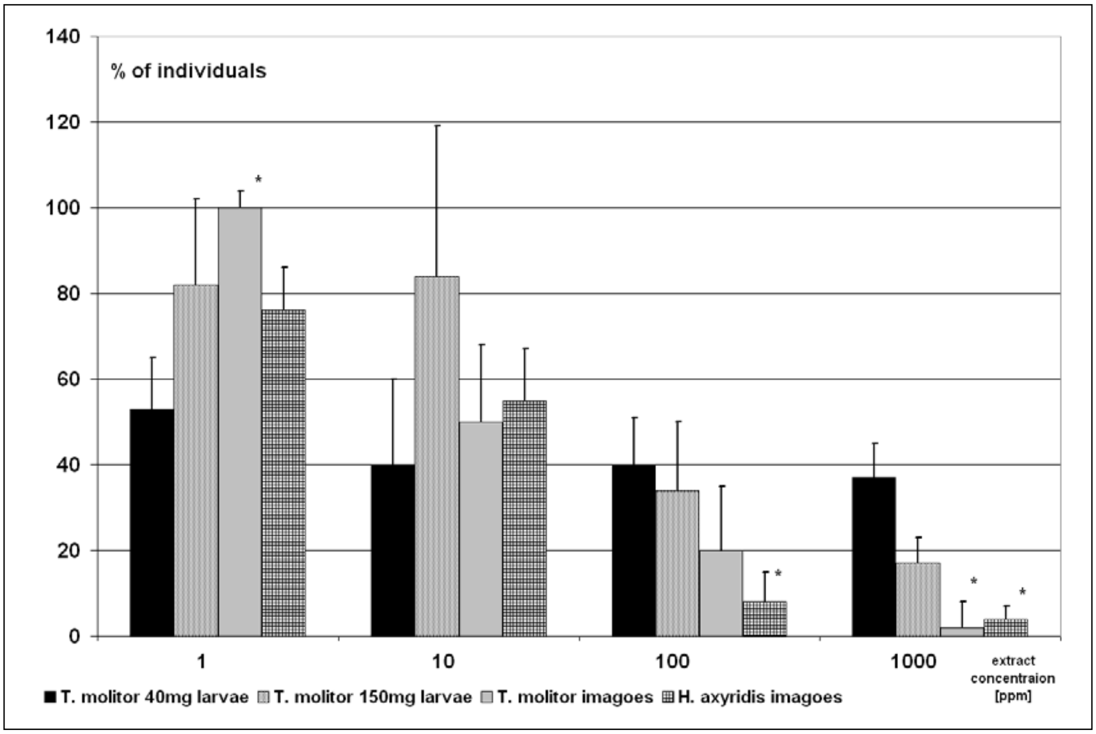

Figure 4. Percentage of insects present at the surface exposed to *L. esculentum* leaf extract, after 24h of exposure. Data are means ± SD. * statisticall significant results, at a significance level of 0.05.

the lowest concentration of the extract rather than the control ones. The same phenomenon was observed within *H. axyridis* adults. However, the high concentrations acted rather as a repellent agent for the above mentioned groups insects. This phenomenon is probably linked with changes of the concentrations of glycoalkaloids in maturing plant organs which makes them edible for herbivores (Friedman, 2002 and 2006). In consequence, mature fruits but not the undeveloped ones, are edible for herbivores. Hence only mature seeds can be distributed. Antifeedant effects of various *Solanum* spp. extracts were reported (Castillo et al., 2009; Chopa et al., 2009; Devanand and Rani, 2011; Jeyasankar et al. 2012). Ladybirds might migrate towards these areas, since low concentration of glycoalkaloids may be a signal for the presence of herbivores, which are the prey of these usually predatory insects. Older yellow mealworm larvae seemed not to have any preference towards or against the extract. In total, this group appeared to be least susceptible for both tested extracts. We did not observe statistically significant changes in distribution of these larvae for any of the tested concentrations. High correlation was noted only for *L. esculentum* extracts, after 1 h of exposure. Perhaps, this is connected with high body mass and developed resistance to various stress factors in older larvae.

Long term exposure of potato extracts resulted in similar trend in young mealworm larvae and adults. Both groups revealed increasing preferences towards control environment. That proves that longer exposure indicates inedible food. The opposite effect was observed within *H. axyridis* adults. They avoided exposed area with the lowest concentration of the extract, but gathered at exposed areas with higher concentrations. These results are in contrary to those observed during the first hour of exposure and need further investigation. Perhaps, the reason is that ladybugs can store large amounts of toxic substances. Therefore, they are resistant to them and, after the first detection of toxic substance, they may tolerate its high concentration, as not harmful. What is more, they feed on insects, which can be often found on Solanaceae plants. Hence, increased level of plant secondary metabolites in the environment may act as a stimulus which "suggests" that herbivore insects can be present at that area and attract predators (Wilmer et al. 2005).

Tomato extract did not cause such intensive effects for both exposure time intervals. Although there was a high correlation between extract concentration and number of insects, the majority of the results were statistically insignificant. However, *T. molitor* adults showed a significant tendency towards low concentrations of the extract in both exposure times and avoided the highest concentration during longer (i.e. 24 h) exposure time. For this group we obtained the highest value of B (1.816) what indicate high slope of the probit line and fast increasing effect. Similarly to *T. molitor*, *H. axyridis* avoided area of higher concentrations during 24 h exposure, although the slope was much less steep (B=0.89) but still one of the highest in the whole experiment. Therefore we think that *L. esculentum* can be a source of substances that have significant repellent activity. Indeed, high toxicity of tomato alkaloids and extracts, including acute toxicity and significantly disturbed larval growth, was reported (Weissenberg et al., 1998; Campbell and Duffey, 1981; Mulatu et al., 2006; Devanand and Rani, 2011). Therefore, insects preffer control areas, to avoid intoxication by high concentrations of poisonous substances present in *L. esculentum* tissues, which serve as food. In such situation, predatory insects are not attract by chemicals in the environment, due to low number of prey. Besides, they may be affected by harmful alkaloids present in tomato plant juices.

In conlusion, we want to emphasize, that extracts of repellent and attractant activity is extremely important for the food storage protection. They limit number of pests inhabiting stored crops and destroying them. However, unlike after usage of chemical insecticides, pests are not killed by repellents and attractants. In consequence, dead insect bodies do not pollute crops and do not decrease taste and dietetic values of products. Therefore, we consider usage of substances and extracts obtained from Solanaceae plants an important point of Integrated Pest Management strategies in a future. They may limit number of highly toxic, non-specific substances introduced to the environment and agriculture in the future.

5. References

Adamski, Z., Marciniak, P., Ziemnicki, K., Buyukguzel, E., Erdem, M., Buyukguzel, K., Ventrella, E., Falabella, P., Cristallo, M., Salvia, R., Bufo, SA., Scrano, L. 2014. Potato leaf extract and its component, alpha-solanine, exert similar impacts on development and oxidative stress in *Galleria mellonella* L. *Arch. Insect Biochem. Physiol.,* 87: 26-39.

Amoabeng, BW., Gurr, GM., Gitau, CW., Stevenson, PC. 2014. Cost:benefit analysis of botanical insecticide use in cabbage: Implications for smallholder farmers in developing countries. *Crop Prot.,* 57: 71-76.

Aygan, A., Karcıoğlu, L., Çömlekcioğlu, N., Keser, S., Battaloğlu, G., Tanış H. 2011. Antimicrobial Activity Determination of Dried Pericarp of *Solanum melongena* (Eggplant) Fruit. *Karaelmas Fen Müh. Derg.,* 1: 40-44.

Brown, PMJ., Thomas, CE., Lombaert, E., Jeffries, DL., Estoup, A., Lawson Handley, L-J. 2011. The global spread of *Harmonia axyridis* (Coleoptera: Coccinellidae): distribution, dispersal and routes of invasion. *BioControl*, 56: 623-641.

Buss, EA., Park Brown, SG. 2014. Natural Products for Managing Landscape and Garden Pests in Florida. *UF/IFAS Extension, ENY-350 (IN197)*, http://edis.ifas.ufl.edu,

Büyükgüzel, E., Büyükgüzel, K., Erdem, M., Adamski, Z., Marciniak, P., Ziemnicki K., Ventrella, E., Scrano, L., Bufo, SA. 2013. The influence of dietary α-solanine on the waxmoth *Galleria mellonella* L. *Arch. Insect Biochem. Physiol.,* 83: 15-24.

Cantelli, BAM., Barbosa1, FR., Bitencourt, TA., Miranda MA., Bastos JK., Marins, M., Fachin, AL., De Abreu, MH., Crivelenti, JD., Mesquita, TB. 2014. Evaluation of antifungal activity of glycoalkaloids from the Solanum lycocarpum St. Hil (lobeira) in the cell membrane of dermatophyte of Trichophyton rubrum. *BMC Proceedings*, 8(Suppl 4): P11.

Castillo, L., Gonzalez-Coloma, A., Gonzalez, A., Diaz, M., Santos, E., Alonso-Paz, E., Bassagoda, MJ., Rossini, C. 2009. Screening of Uruguayan plants for deterrent activity against insects. *Ind. Crops Prod.,* 29: 235-240.

Cataldi, TRI., Lelario, F., Bufo, SA. 2005. Analysis of tomato glycoalkaloids by liquid chromatography coupled with electrospray ionization tandem mass spectrometry. *Rapid Commun Mass Spectrom.,* 19: 3103–3110.

Campbell, BC., Duffey, SS. 1981. Alleviation of α-tomatine-induced toxicity to the parasitoid, *Hyposoter exiguae*, by phytosterols in the diet of the host, *Heliothis zea*. *J. Chem. Ecol.,* 7: 927-946.

Chowanski, S., Kudlewska, M., Marciniak, P., Rosinski, G. 2014. Synthetic insecticides – is there an alternative? *Polish J Environ. Studies,* 23: 291-302.

Chopa, CS., Benzi, V., Alzogaray, R., Ferrero, AA. 2009. Repellent activity of hexanic and ethanolic extracts from fruits of *Solanum eleagnifolium* (Solanaceae) against *Blattella germanica* (Insecta, Dictyoptera, Blattellidae) adults. *Bol. Latinoam. Caribe Plant. Med. Aroma.,* 8: 172-175.

Devanand, P., Rani, PU. 2011. Insect growth regulatory activity of the crude and purified fractions from *Solanum melongena* L., *Lycopersicum esculentum* Mill, and *Capsicum annuum* L. *J Biopestic.,* 4: 118-130.

Finney, DJ. 1971. Probit Analysis. London/New York. Cambridge University Press.

Friedman, M. 2002. Tomato glycoalkaloids: role in the plant and in the diet. *J Agric Food Chem.,* 50: 5751-5780.

Friedman, M. 2006. Potato glycoalkaloids and metabolites: roles in the plant and in the diet. *J. Agric. Food Chem.,* 54: 8655-8681.

Hutchison, WD., Galvan, TL., Burkness, EC., Koch, RL. 2010. *Harmonia axyridis* as an economic pest of wine grapes in the U.S.: Progress in developing an IPM program and potential impact in Europe. *Benefits and Risks of Exotic Biological Control Agents, IOBC/wprs Bulletin,* 58: 47-52.

Jeyasankar, A., Premalatha, S., Elumalai, K., Krishnappa, K. 2012. Biological activities of *Solanum pseudocapsicum* (*Solanaceae*) against cotton bollworm, *Helicoverpa armigera* Hubner and armyworm, *Spodoptera litura* Fabricius (Lepidotera: Noctuidae). *Asian Pac. J Trop. Biomed.,* 2, 981-986.

Koch, RL. 2003. The multicolored Asian lady beetle, *Harmonia axyridis*: A review of its biology, uses in biological control, and non-target impacts. *J Insect Sci.,* 3: 1-16.

Marciniak, P., Adamski, Z., Bednarz, P., Slocinska, M., Ziemnicki K., Lelario, F., Scrano, L., Bufo, SA. 2010. Cardioinhibitory properties of potato glycoalkaloids in beetles. *Bull. Environ. Contam. Toxicol.,* 84:153–156.

Mesiani, S., Oomah, B., Zaidi, F., Simon-Levert, A., Bertrand, C., Zaidi-Yahiaoui, R. 2015. Antibacterial activity of carob (*Ceratonia siliqua* L.) extracts against phytopathogenic bacteria *Pectobacterium atrosepticum*. *Microb. Pathog.,* 78: 95-102.

Mulatu, B., Applebaum, S.W., Kerem, Z., Coll, M. 2006. Tomato fruit size, maturity and alpha-tomatine content influence the performance of larvae of potato tuber moth *Phthorimaea operculella* (Lepidoptera: Gelechiidae). *Bull. Entomol. Res.,* 96: 173-178.

Ntalli, N., Kopiczko, A., Radtke, K., Marciniak, P., Rosiński, G., Adamski, Z. 2014. Biological activity of Melia azedarach extracts against Spodoptera exigua. *Biologia (Bratislava),* 69: 1606-1614.

Rosinski, G., Wrzeszcz, A., Obuchowicz, L. 1979. Differences in trehalase activity in the intestine of fed and starved larvae of *Tenebrio molitor* L. *Insect Biochem.,* 9: 485–488.

Ventrella, E., Marciniak, P., Adamski, Z., Rosiński G., Chowański S., Falabella, P., Scrano, L., Bufo, SA. 2015. Cardioactive properties of Solanaceae plant extracts and pure glycoalkaloids on *Zophobas atratus*. *Insect Sci*., 22: 251-262.

Weissenberg, M., Levy, A., Svoboda, JA., Ishaaya, I. 1998. The effect of some Solanum steroidal alkaloids and glycoalkaloids on larvae of the red flour beetle, *Tribolium castaneum*, and the tobacco hornworm, *Manduca sexta*. *Phytochemistry,* 47, 203-9.

Willmer P., Stone G., Johnston I. 2005. Environmental Physiology of Animals. Second Edition. Blackwell Publ.