BİTKİ KORUMA BÜLTENİ 2017, 57(3) : 305 - 315
ISSN 0406-3597
DOI 10.16955/bitkorb.297213

Stomach poison activity of some plant extracts on Colorado potato beetle (Coleoptera: Chrysomelidae)

Mustafa ALKAN[1] **Ayhan GÖKÇE[2]** **Kenan KARA[3]**

ABSTRACT

Bazı bitki ekstraktlarının Patates böceği (Coleoptera: Chrysomelidae) üzerindeki mide zehiri aktivitesi

Discovery of new eco-friendly methods for management of insect-pests is very important in integrated pest management. Toxicities of six different plant extracts (*Acanthus dioscoridis* L., *Achillea millefolium* L., *Bifora radians* Bieb., *Heracleum platytaenium* Boiss, *Humulus lupulus* L. and *Phlomoides tuberosa* (L.) Moench) were evaluated against Colorado potato beetle (CPB) larvae under laboratory conditions. Methanol extracts prepared from vegetative parts of plants were initially tested against 3rd instar larvae via feeding method. Maximum mortality was observed in the case of *H. platytaenium* and *H. lupulus* extracts after 48 hours when tested against 3rd stage larvae. So these extracts bearing the maximum biological activity were further evaluated using 2nd and 4th instar larvae. Second instar larvae were more susceptible to *H. platytaenium* and *H. lupulus* than 3rd instar larvae. However, 4th stage larvae were the most resistant to these extracts' activities. These results indicated that *H. platytaenium* could be potentially used in the management of CPB, especially as a stomach poison.

Keywords: *Leptinotarsa decemlineta*, bio-pesticides, stomach poison, *Heracleum platytaenium*, *Humulus lupulus*, plant extract, CPB

ÖZ

Entegre zararlı yönetiminde zararlı böceklerin yönetimi için yeni, çevre dostu metodların geliştirilmesi çok önemlidir. Altı farklı bitki ekstraktının (*Acanthus dioscoridis* L., *Achillea millefolium* L., *Bifora radians* Bieb., *Heracleum platytaenium* Boiss, *Humulus lupulus* L.

[1] Department of Entomology, Plant Protection Central Research Institute, Yenimahalle, Ankara, Turkey
[2] Department of Plant Production and Technologies, Faculty of Agricultural Sciences and Technologies, Ömer Halisdemir University, Niğde, Turkey
[3] Department of Plant Protection, Faculty of Agriculture, Gaziosmanpasa University, Tokat, Turkey
Corresponding author (Sorumlu yazar) e-mail: alkan0101@gmail.com - mustafa.alkan@tarim.gov.tr
Received (alınış): 10.03.2017, Accepted (kabul ediliş): 13.06.2017

ve *Phlomoides tuberosa* (L.) Moench) toksisiteleri Patates böceğine karşı laboratuvar koşullarında değerlendirilmiştir. Bitkilerin vejetatif aksamlarından hazırlanan methanol ekstraktları beslenme yoluyla 3.dönem larvalara karşı test edilmiştir. Maksimum ölüm oranı *H. platytaenium* ve *H. lupulus* ekstraktında 48 saat sonra gözlemlenmiştir. Bu yüzden bu ekstraktlar ile ileri biyolojik aktivite testleri 2. ve 4. dönem larvalar ile de yürütülmüştür. *H. platytaenium* ve *H. lupulus* ekstraktları için 2. dönem larvalar 3. dönem larvalardan daha hassas olmuşlardır. Test edilen dönemlerden 4. dönem larva bu ekstraktların aktiviteleri bakımından en direçli dönem olmuştur. Bu sonuçlar *H. platytaenium*'un Patates böceğinin yönetiminde özellikle mide zehiri olarak potansiyel olarak kullanılabileceğini ortaya koymuştur.

Anahtar kelimeler: *Leptinotarsa decemlineta*, biopestisit, mide zehiri, *Heracleum platytaenium, Humulus lupulus,* bitki ekstraktı

INTRODUCTION

Worldwide, potato is the fifth highest crop by acreage (Anonymous 2015a). The tubers are rich source of proteins, carbohydrates, minerals (K, Mn, Mg, Fe, Cu and P) and vitamins (C, B1, B3, B6, K, folate, pantothenic acid) (Alisdair et al. 2001). Turkey is among the important potato producing countries of the world. Area under potato cultivation was around 130000 hectares and total potato production was nearly 5 million tones in 2014 (Anonymous 2015b).

However per acre production of potato in Turkey is less as compared to many countries primarily due to attack of insect-pests. Around 270 insects and 17 mite species are reported on potato under field and storage conditions worldwide. In case of severe insect attack, yield losses may reach to 100% (Chalfant 1990). The CPB is considered as one of the most serious insect pest of potato crop (Weber and Ferro 1994) and is also a vector of bacterial ring rot disease (Christie et al. 1991). It is a multivoltine insect and uncontrolled populations can completely defoliate potato plants/crop during the growing season (Hsiao and Fraenkel 1968). Severe attack during the plant growth cycle may lead to 100% yield loss (Hare 1980). Chemical control is most commonly used strategy for management of insect-pests (Jaleel et al. 2014, Alkan et al. 2015). The CPB has an important role in creating the modern pesticide industry, with hundreds of chemicals tested against it (Alyokhin 2009).

Insecticides are heavily used to control insects by killing them or preventing them from eating in undesirable behaviors (Afzal et al. 2015). The frequent and uncheck use of insecticides can lead to phytotoxicity and resistance problem (Stewart et al. 1997, Mota-Sanchez et al. 2000). Ability to adapt to toxic substances together with high selection pressure eventually resulted in a large number of insecticide-resistant CPB populations. It has developed resistance to 52 different compounds belonging to all major classes of insecticides (Whalon et al. 2015). Resistance levels vary greatly in different populations and beetle life stages, but in some cases can be very high (up to 2,000-fold) (Alyokhin 2009). The frequent use of

insecticides can also affect the physiological make-up of the target pests by causing changes in reproduction parameters, growth and development (Afzal et al. 2015).

Since 1990, the bio-pesticides have been used commercially especially when the problems associated with the intensive use of pesticides against various pest species started (Nitao 1987, Pascual-Villalobos and Robledo 1999, Chiasson et al. 2004, Thacker 2002). Biopesticides (plant based) are generally made from the secondary metabolites that are synthesized after pest attacks or unfavorable conditions by the plants (Bourgaud et al. 2001). They are natural repellents, antifeedants and have toxic properties against many insect pest species (Mordue (Luntz) and Nisbet 2000). Although there are many reports regarding insecticidal properties of plant extracts, the practical application of them is still limited (Hassan and Gökçe 2014).

An alternative control strategy is needed against CPB to delay the resistance development against insecticides and also to manage the negative impact of insecticides on the environment and human health. As pointed out above, the plant extracts have various modes of action against insect pests and they might have important roles in eco-friendly approaches to manage the pests. In this study, the residual toxicity of six different plants extracts were tested on various development stages of CPB. Additionally, dose-response bioassay was carried out with *H. platytaenium* and *H. lupulus* extracts.

MATERIALS AND METHODS

The experiment was conducted at Laboratory of Entomology, Department of Plant Protection, Gaziosmanpasa University, Tokat, Turkey.

Collection of plant materials

Collected plants species, their material used in experiment and their location are listed in Table 1. Samples were collected in summer and spring during the year 2009 (Gökçe et al. 2005). Plant materials (seeds, leaves, stem and flowers) were washed with distilled water to remove dust particles and dried at room temperature for two weeks in the laboratory. Samples were ground to get fine powder using grinder (M-20 IKA Universal Mill, IKA Group, Wilmington, NC, USA) and then preserved in glass jar (25 cmL×12cmW) at 15±5˚C for further usage.

Table 1. Name of selected plants species, their analyzed part and location of collection

Botanical Name	Family	Analyzed part	Location
Humulus lupulus	Cannabaceae	Cone	Tokat, Turkey
Heracleum platytaenium	Apiaceae	Leaf, Stem	Trabzon, Turkey
Achillea millefolium	Asteraceae	Leaf, Stem	Tokat, Turkey
Acanthus dioscoridis	Acanthaceae	Leaf, Stem	Erzincan, Turkey
Phlomoides tuberosa	Lamiaceae	Leaf, Stem	Erzincan, Turkey
Bifora radians	Apiaceae	Leaf, Stem	Tokat, Turkey

Preparation of plant extracts

Plant extracts were prepared by using maceration method (Alkan and Gökçe 2012). The hexane, ethyl acetate and methanol were added according to their polarities into 200g plant sample using a glass jar. The samples were treated with hexane till 48 hours and suspension was filtered from plant material using filter paper. Later on, plant material was treated with ethyl acetate till 48 hours and suspension obtained from ethyl acetate extract was again filtered using filter paper. Finally, methanol was added to the plant materials, and same procedure was adopted for methanol extraction. The suspension was placed in an evaporator (RV 05 Basic 1-B, IKA® Werke GmbH & Co. KG, Germany) and excess solvent was evaporated. Methanol extracts obtained from *H. lupulus*, *B. radians* and *A. millefolium* were transferred into glass tubes and stored at 4°C in the refrigerator for further use.

Rearing of Colorado potato beetle

The CPB larvae were reared at Gaziosmanpasa University, Faculty of Agriculture, Department of Plant Protection (Gökçe et al. 2006). The CPB colony was continuously reared on potato plants (*Solanum tuborosum* L. cultivar Granola) which were planted at Gaziosmanpasa University Research Station in Tasliciftlik, Tokat, Turkey. Field was designated for organic potato production and there were no pesticide application for 3 years prior to the initiation of this project. Granola cultivar was planted in a 0.2 ha potato field. When the potato plants reached 3-5 leaves stage *L. decemlineata* adults were released into the field and all required stages for the studies were collected from that field.

Single dose efficacy trails

The extracts were diluted with acetone to obtain the concentration of 10.00% plant extract in acetone (w/v). Each side of potato leaves were sprayed with 20 µl of plant extracts suspension using a hand spray. Spinosad (Laser®, Dow Agro Sciences write E.C or percent formulation) was used as negative control at recommended dose of 0.1 ml/L for *L. decemlineata*. Spinosad suspension (20 µl) was applied to each side of potato leaves. Simple acetone (20 µl) was applied to potato leaves in control. During the first phase of the study, all plant extracts were screened against 3rd instar larvae of the CPB. In the second phase, *H. platytaenium*, *H. lupulus* extracts and spinosad were also tested on 2nd and 4th instar larvae. In each replicate, 15 *L. decemlineata* larvae were used in three different groups as 5 larvae were released on each treated potato leaf. The mortality was recorded after 24, 48 and 72 hours of treatment. The experimental trials were performed using completely randomized design (CRD) with eight treatments and three replications.

Dose-response bioassay

Based on the single dose screening test, further dose-response bioassays were carried out with *H. platytaenium* and *H. lupulus* extracts. Four different concentrations of these plants extracts i.e. 1%, 3%, 5% and 7.5% (w/v) were tested

on 3rd instar larvae. The treatment was carried out as described above. The larval mortality was recorded after 72 hours. Completely randomized design (CRD) was used for the experiment set up and each treatment was replicated thrice. Each replicate contained three groups of 5 larvae and total 45 larvae were used at each dose.

Statistical analysis

Percent mortality results were subjected to arcsine transformation. Data were subjected to analysis of variance and differences among treatment were analyzed by Tukey Multiple Comparison Test (α =0.05). All statistical analysis were done using MINITAB® version 16 software package (Minitab Inc. US2010).

RESULTS AND DISCUSSION

Single dose bio-assays

In single-dose screening test, methanol extracts of 6 plants were tested on 3rd instar larvae of CPB. All plant extracts, except *A. dioscoridis* and *P. tuberosa*, caused mortality of CPB larvae after 48 hours (Table 2). Highest percentage mortality (43.20%) of CPB larvae was recorded in *H. platytaenium* treatment. This was followed by *H. lupulus* extract with 27.20% mortality. Negative control (spinosad) was very toxic to 3rd instar larvae and caused 98.30% mortality at 48 h. The mortality rates recorded from these two extracts and spinosad were statistically different to a great extent from other treatments ($F=58.79$, $df=7.15$, $P<0.05$). These results are in conformity with the work of Çam et al. (2012) who tested different plant extracts against various developmental stages of CPB and reported that the plant extract, *H. lupulus* was effective against all the stages of CPB except adult stage. However, our results are different from some other studies (Ermel et al. 1983, Pavela et al. 2008, Sajfrtova et al. 2008), which could be result of difference in collection place and time of the plant species and difference in extraction methods that were used in the studies. Collecting time and place of plant species affect the specific amount of chemical molecules present in leaves and also the extraction methods are directly related to the presence of chemical compounds in the extracts e.g. α- Thujame, α - Pinene, Sabinene, 1,8- Cineole, camphor and ρ - Cyr man -8-ol. These factors play important roles for inducing the insecticidal properties of extracts (Karuppusamy and Muthuraj 2011). Promising results of *Heracleum* spp. extracts against insect-pests like *Sitophilus zeamais* and *Tribolium castaneum* adults are known (Chu et al. 2012). *Heracleum* spp. essential oils contain aliphatic esters, terpenes and these components are known to have antibacterial, antifungal, anti-dermatophytic activities (Torbati et al. 2013). The stomach poison activity of *H. platyaenium* observed in this study could be related with the presence of above chemical compounds in leaves of *H. platytaenium* selected for extraction.

Table 2. Stomach poison effects of plant extracts and spinosad to 3rd stage larvae of CPB after 48 hours

Treatment	Mortality% ± SD*
Control	0.00±0.00 e
Humulus lupulus	27.19±1.77bc
Heracleum platytaenium	43.19±2.05 b
Achillea millefolium	11.61±0.14 cd
Acanthus dioscoridis	0.00±0.00 e
Bifora radians	6.67±0.00 de
Phlomoides tuberosa	0.00±0.00 e
Spinosad	98.30±3.37 a

The mean follow-up of the column that different lowercase letters, shows that the average differs significantly (ANOVA P <0.05, Tukey test).
* SD: Standard Deviation

H. platytaenium and *H. lupulus* extracts alongside with spinosad as a chemical standard l were tested against the 2nd instar larval stage of CPB (Table 3). Plant extract of *H. lupulus* caused about 98.30% mortality that was statistically similar to the chemical standard, spinosad treatment. These two treatments were statistically significantly different from both *H. platytaenium* (46.70% mortality) and control (0.00% mortality) ($F=146.29$, $df=3.9$, $P<0.05$). First three stages of CPB are more susceptible to the plants extracts than bigger stage larval instars (Scott et al. 2003, 2004; Çam et al. 2012).

Table 3. The stomach poison effect of plant extracts and spinosad to 2nd stage larvae of CPB after 48 hours

Treatment	Mortality% ± SD*
Control	0.00±0.00 c
Heracleum platytaenium	46.65±0.90 b
Humulus lupulus	98.30±3.37 a
Spinosad	98.30±3.33 a

The mean follow-up of the column that different lowercase letters, shows that the average differs significantly (ANOVA P <0.05, Tukey test).
* SD: Standard Deviation

The plants extracts were also tested on the 4th instar larvae of CPB. There was a significant differences among the mortality rates of 4th instar larvae of CPB when they were fed on *H. lupulus*, *H. platytaenium* and spinosad treated potato leaves ($F=44.80$; $df=3.11$, $P<0.05$). Interestingly, *H. platytaenium* extracts produced 13.10% mortality after 48 hours and it was less than those were seen on 2nd and 3rd stage. *H. lupulus* extracts stomach poison activity fluctuated depending on the tested stage of CPB and it caused 46.50% mortality after 48 hours. Spinosad activity remained at the similar level and 93.30% mortality was observed in the insecticide treated replicates (Table 4). Similar results were also reported by other researchers that the plants extracts are effective on the immature stages of the CBP (Scott et al. 2004).

Table 4. Stomach poison effects of single concentration of plant extracts and spinosad to 4th stage larvae of CPB after 48 hours

Treatment	Mortality %± SD*
Control	0.00±0.00 d
Heracleum platytaenium	13.05±3.64 c
Humulus lupulus	46.54±3.62 b
Spinosad	93.33±0.00 a

The mean follow-up of the column that different lowercase letters, shows that the average statistical significantly different (ANOVA P <0.05, Tukey test).
* SD: Standard Deviation

Dose-response bioassay

Dose-response bioassay with the most promising plant extracts (*H. platytaenium* and *H. lupulus*) revealed that toxicity of plant extract was not directly proportional to tested dose of plant extracts in this study. Population treated with *H. platytaenium* extract showed 100% mortality at 5.00% and 7.50% tested concentrations. Whereas 95.80% mortality was recorded at 3.00% concentration followed by 92.20% mortality at 1.00% tested concentration (Table 5). These results suggested that 1.00% concentration of *H. platytaenium* could be the best option for testing stomach poison effect of this plant. This could be due to the larval preference of feeding on leaflets that were treated with low concentration of plant extract. As a result, the more active ingredient entered the insect's body and leading to the death of the CPB larvae. Similar results were reported by Wawrzyniak and Lamparski (2006) who tested different plant extracts effects on CBP feeding and development. Maximum mortality i.e. 58.91% was obtained at 3.00% concentration followed by 53.35% mortality at 7.50% concentration of *H. lupulus* extracts. Around 51.12% mortality was observed in case of leaves treated with 5.00% concentration. In control treatments mortality wasn't recorded. This low percent mortality observed in *H. lupulus* treatments comparing with *H. platytaenium* could be due to repellent effects of the plants extracts (Gökçe et al. 2012).

Table 5. Dose-response results of 3rd stage *Leptinotarsa decemlineata* larvae treated with various concentration *Heracleum platytaenium* and *Humulus lupulus* after 72 hours

Treatment	Tested Concentrations	Mortality%± SD*
Heracleum platytaenium	1.00%	92.24±7.46 b
	3.00%	95.81±5.85 ab
	5.00%	100.00±0.00 a
	7.50%	100.00±0.00 a
	Control	0.00±0.00c
Humulus lupulus	1.00%	0.00±0.00 b
	3.00%	58.91±8.72 a
	5.00%	51.12±3.42 a
	7.50%	53.35±1.01 a
	Control	0.00±0.00 b

The mean follow-up of the column that different lowercase letters, shows that the average statistical significantly different (ANOVA P <0.05, Tukey test).
* SD: Standard Deviation

In this study, the stomach poisoning effects of different plant extracts were tested against various larval stages of CPB. Greatest stomach poison activity was found in case of plant extracts of *H. platytaenium* and *H. lupulus*. Dose efficacy studies indicate that even 1.00% concentration of *H. platytaenium* could be effective in controlling the beetle larvae. However, further field studies are needed to evaluate the original potential of this plant extracts. Meantime, isolation and characterization of active compound(s) from this plant extract should be studied for development of this plant extract as a plant based bio-pesticide for controlling CPB.

ACKNOWLEDGMENTS

This study was supported by Turkish Republic Prime Minister State Planning Organization (27-DPT-01-07-01) and by the Ministry of Food, Agriculture and Livestock, General Directorate of Agricultural Research and Policy (TAGEM-BS-12 / 04-04 / 01- 04). The authors thank to Prof. Dr. Mark E. Whalon (Michigan State University) for his valuable time contribution to this study. This article has been drawn up from a certain part of the doctoral project of Mustafa Alkan and this study was presented as a poster presentation at the Plant Protection Congress V, Antalya, Turkey.

REFERENCES

Afzal M.B., Shad S.A., Abbas N., Ayyaz M. and Walker W.B. 2015. Cross-resistance, the stability of acetamiprid resistance and its effect on the biological parameters of cotton mealybug, *Phenacoccus solenopsis* (Homoptera: Pseudococcidae), in Pakistan. Pest Management Science, 71: 151-158.

Alisdair R., Fernie R. and Willmitzer L. 2001. Update on tuber formation, dormancy, and sprouting molecular and biochemical triggers of potato tuber development. Plant Physiology, 127: 1459-1465.

Alkan M. and Gökçe A. 2012. Toxic and behavioural effects of *Tanacetum abrotanifolium* L. Druce (Asteraceae) stem and flower extracts on *Sitophilus granarius* and *Sitophilus oryzae* (Col., Curculionidae). Turkish Journal of Entomology, 36 (3), 377-389.

Alkan M., Gökçe A. and Kara K. 2015. Antifeedant activity and growth inhibition effects of some plant extracts against larvae of Colorado potato beetle [*Leptinotarsa decemlineata* Say (Col: Chyrsomelidae)] under laboratory conditions. Turkish Journal of Entomology, 39 (4), 345-353.

Alyokhin A. 2009. Colorado potato beetle management on potatoes: Current challenges and future prospects. Fruit, Vegetable and Cereal Science and Biotechnology, 3 (Special Issue 1), 10-19.

Anonymous. 2015a. faostat.fao.org/site/339/default.aspx. (Accessed date: 05.07.2015)

Anonymous 2015b, http://rapory.tuik.gov.tr/09-03-2015-00:15:32-368530409144341289842646212 9.html. (Accessed date: 05.03 2015).

Bourgaud F., Gravot A., Milesi S. and Gontier E. 2001. Production of plant secondary metabolites; a historical perspective. Plant Science, 161: 839-851.

Çam H., Gökçe A., Kadıoğlu İ., Yanar Y., Gören N. and Whalon M.E. 2012. Residual toxicities and stomach poison effects of plant extracts to different stages of Colorado potato beetle [*Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae)]. Turkish Journal of Entomology, 36 (2), 249-254.

Chalfant R.B. 1990. Ecology and management of sweet potato insects. Annual Review of Entomology, 35: 157-80.

Chiasson H., Vincent C. and Bostanian B. 2004. Insecticidal properties of a *Chenopodium* based botanical. Journal of Economic Entomology, 97: 1378-1383.

Christie R.D., Sumalde A.C., Schutz J.T. and Gudmestad N.C. 1991. Insect transmission of the bacterial ring rot pathogen. Am. *Potato* Journal, 68: 363-372.

Chu S.S., Cao J., Liu Q.Z., Du S.S., Deng Z.W. and Liu Z.L. 2012. Chemical composition and insecticidal activity of *Heracleum moellendorffii* Hance essential oil. Chemija, 23: 108-12.

Ermel K., Pahlich E. and Schmuttrerer H. 1983. Comparison of the azadirachtin content of neem seed from ecotypes of asian an african origin. Schriftenreihe der Gesellschaft fur Technische Zusammenarbeit., 91-94.

Gökçe A., Isaacs R. and Whalon M.E. 2012. Dose-response relationships for the antifeedant effects of *Humulus lupulus* extracts against larvae and adults of the Colorado potato beetle. Pest Management Science, 68: 476-481.

Gökçe A., Stelenski L.L. and Whalon M.E. 2005. Behavioral and electrophysiological responses of leafroller moths to selected plant extracts. Environmental Entomology, 34: 1426-1432.

Gökçe A., Whalon M.E., Çam H., Yanar Y., Demirtaş İ. and Gören N. 2006. Plant extract contact toxicities to various developmental stages of Colorado potato beetles (Coleoptera: Chrysomelidae). Annals of Applied Biology, 149: 197-202.

Hare J.D. 1980. Impact of defoliation by the Colorado potato beetle on potato yields. Journal of Economic Entomology, 73: 369-73.

Hassan E. and Gökçe A. 2014. Production and consumption of biopesticides. In Advances in Plant Biopesticides (pp. 361-379). Editors: Dwijendra, Singh, Springer India.

Hsiao T.H. and Fraenkel G. 1968. The influence of nutrient chemicals on the feeding behavior of the Colorado potato beetle, *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae). Annals of the Entomological Society of America, 61: 44-54.

Jaleel W., Saeed S., Naqqash M.N. and Zaka S.M. 2014. Survey of Bt cotton in Punjab Pakistan related to the knowledge, perception and practices of farmers regarding insect pests. International Journal of Agriculture and Crop Sciences, **7** (1), 10.

Karuppusamy S. and Muthuraja G. 2011. Chemical composition and antioxidant activity of *Heracleum sprengelianum* (Wight and Arnott) essential oils growing wild in Peninsular India, Iranian Journal of Pharmaceutical Research, 10 (4), 769-775.

Mordue (Luntz) A.J. and Nisbet A.J. 2000. Azadirachtin from the neem tree *Azadirachta indica*: its action against insects. Anais da Sociedade Entomologica do Brasil, 29 (4), 615-632.

Mota-Sanchez D., Whalon M.E., Grafius E. and Hollingworth R. 2000. Resistance of Colorado potato beetle to imidacloprid resistance. Pest Management, 11: 31-34.

Nitao J.K. 1987. Test for toxicity of Coniine to a polyphagous herbivore, *Heliothis zea* (Lepidoptera:Noctuidae). Environmental Entomology, 16: 656-659.

Pascual-Villalobos M.J. and Robledo A. 1999. Anti-insect activity of plant extracts from the wild flora in southeastern Spain. Biochemical Systematics and Ecology, 27: 1-10.

Pavela R., Sajfrtova M., Sovova H. and Barnet M. 2008. The insecticidal activity of *Satureja hortensis* L. extracts obtained by supercritical fluid extraction and traditional extraction techniques. Applied Entomology and Zoology, 43 (3), 377-382.

Sajfrtova M., Sovova H. and Pavela R. 2008. Effect of extraction methods on botanical insecticide activity. 11th European Meeting on Supercritical Fluid Reaction, Materials and Natural Products Processing.

Scott I.M., Jensen H., Nicol L., Bradbuty R., Sanches-Vindas P., Poveda L., Arnason J.T. and Philogene B.J.R. 2004. Efficacy of Piper (Piperaceae) extracts for control of

common home and garden insect pests. Journal of Economic Entomology, 97: 1390-1403.

Scott I.M., Jensen H., Scott J.G., Isman M.B., Arnason J.T. and Philogene B.J.R. 2003. Botanical insecticides for controlling agricultural pests: Piperamides and the Colorado potato beetle, *Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae). Archives of Insect Biochemistry and Physiology, 54: 212-225.

Stewart J.G., Kennedy G.G. and Sturz A.V. 1997. Incidence of insecticides resistance in population of Colorado potato beetle, *Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae) on Prince Edward Island. Canadian Entomology, 129: 21-26.

Thacker J.M.R. 2002. An introduction to Arthropod pest control. Cambridge University Press, Cambridge, ISBN**:** 9780521567879, 343 pp.

Torbati M., Nazemiyeh H., Lotfipour F., Asnaashari S., Nemati M. and Fathiazad F. 2013. Composition and antibacterial activity of *Heracleum transcaucasicum* and *Heracleum anisactis* aerial parts essential oil. Advanced Pharmaceutical Bulletin, 3: 415-418.

Wawrzyniak M. and Lamparski R. 2006. Effect of Umbelliferae (Apiaceae) plant water extracts on Colorado potato beetle (*Leptinotarsa decemlineata* Say) feeding and development. Electronic Journal of Polish Agricultural Universities, 9 (4): 23.

Weber D.C. and Ferro D.N. 1994. Colorado potato beetle: Diverse life history poses challenge to management. In: Zehnder G.W., Powelson M.L., Jansson R.K. and Raman K.V. (eds) Advances in potato pest biology and management, pp. 54-70. APS Press, St Paul, MN.

Whalon M.E., Mota-Sanchez D., Hollingworth R. and Duynslager L. 2015. Arthropod pesticide resistance database. http://www.pesticideresistance.com (Accessed date: 12.05.2014).