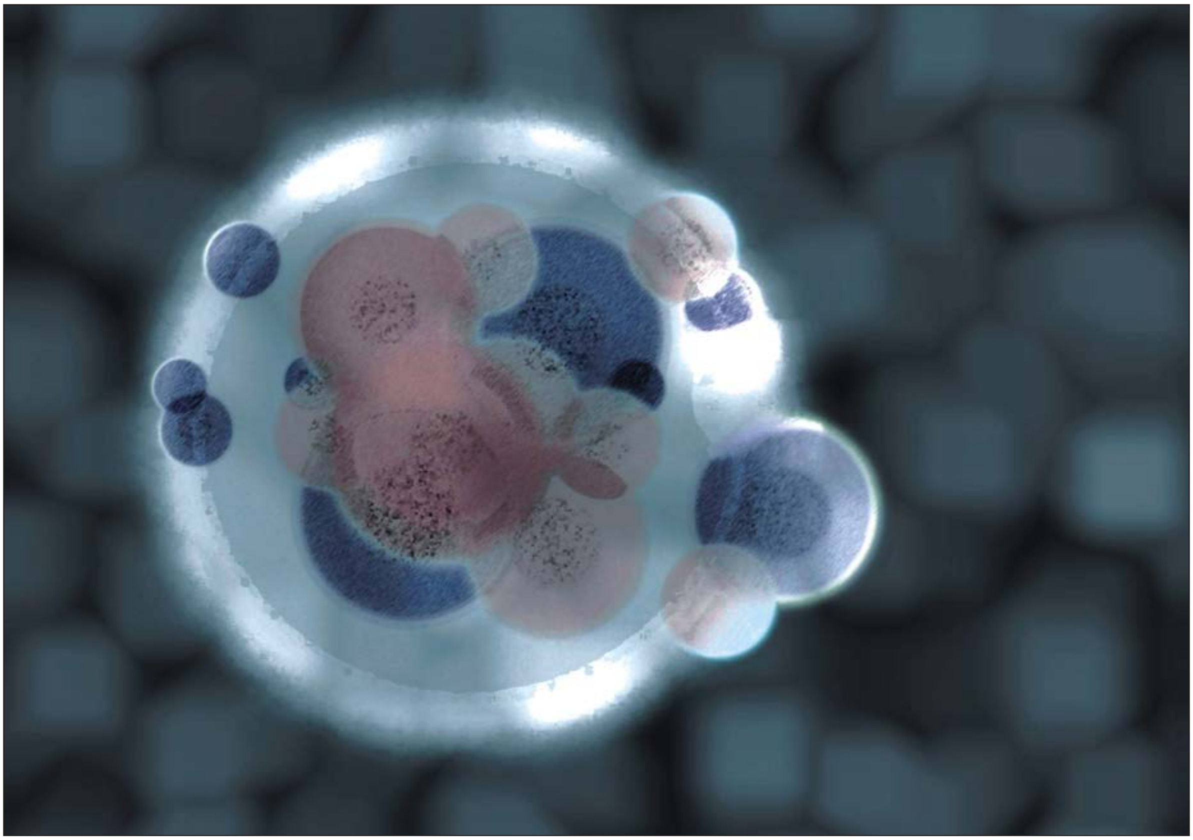

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Insecticidal Effects of Ethanolic Extract from *Verbascum cheiranthifolium* Boiss. Against Two Stored-Product Insect Pests Species

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Abstract: Ethanolic extract from flowers of *Verbascum cheiranthifolium* Boiss. (Scrophulariaceae) was examined for its effect on mortality and progeny production against adults of *Sitophilus oryzae* (L.) and *Tribolium castaneum* (Herbst.). The insecticide was applied at five dose rates, which 0.25, 0.5, 1.0, 2.0 and 3% (w/v). Adults of the above-mentioned species were exposed to the treated wheat at 25°C and 65% RH and mortality was assessed after 24 and 48 h, 7, 14 and 21 day of exposure. Then all adults were removed and the treated substrate remained at the same conditions for an additional 45 day after this interval, the commodity was checked for progeny production. For two species mortality increased with the increase of dose and exposure interval. Results indicate that *S. oryzae* was more sensitive than *T. castaneum*. In case of *S. oryzae* mortality was 100% after 21 day of exposure at the highest dose rate. For two species, complete suppression of the progeny production was observed in the treated wheat than in the untreated wheat even in the lowest dose rate. Therefore, this botanical material can be used for protection of stored Wheat from infestations of stored-product insect pests.

Key words: Insect pests, progeny production, mortality, *Verbascum cheiranthifolium*

INTRODUCTION

Recently, there has been a growing interest in research concerning the possible use of plant extracts as alternatives to synthetic insecticides. Different types of plant preparations such as powders, solvent extracts, essential oils and whole plants are being investigated for their insecticidal activity including their action as fumigants, repellents, anti-feedants, anti-ovipositions and insect growth regulators (Rembold, 1994; Schmutterer, 1995; Prakash and Rao, 1997; Isman, 2000; Weaver and Subramanyam, 2000).

Higher plants are a rich source of novel natural substances that can be used to develop environmental safe methods for insect control (Arnason *et al*., 1989). Yang and Tang (1988) reviewed the plants used for pest insect control and found that there is a strong connection between medicinal and pesticidal plants.

Tribolium castaneum (Herbst.) and *Sitophilus oryzae* (L.) are considered as major pests of stored grain (Howe, 1965). Control of these insects relies heavily on the use of synthetic insecticides and fumigants. But their widespread use has led to some serious problems including development of insect strains resistant to insecticides (Zettler and Cuperus, 1990; White, 1995; Riberio *et al*., 2003). Toxic residues on stored grains, toxicity to consumers and increasing costs of application.

Convenient to use and environmentally friendly. Considerable efforts have been focused on plant derived materials, potentially useful as commercial insecticides. Toxic effects of plant products on some pests have been studied by Stevenson *et al*. (1993), Koul *et al*. (1996) Sahayaraj and Sekr (1996), Senthilkumar *et al*. (1997) and Sahayaraj and Paulraj (1998). Roy *et al*. (2005) established leaf extracts of *Shiyalmutra* (*Blumea lacera*) as botanical insecticides against lesser grain bore and rice weevil. Kabaru and Gichia (2001) revealed that the insecticidal and anti-feedant activity of extracts derived from different parts of the mangrove tree *rhizophora mucronata* (Rhizophoraceae).

Verbascum cheiranthifolium boiss. (Scrophulariaceae) is locally used to kill fishes in northern and northwest regions of Iran. However, flowers of this plant have not been studied yet for insecticidal activity.

The aim of this study was to produce crude extract from *V. cheiranthfolium* for to determine insecticidal activity and effect on progeny production against of two stored-product beetle species.

MATERIALS AND METHODS

Plant extract and test insects: Crude extract of botanical was used. *Verbascum cheiranthifolium* boiss.

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(Scrophulariaceae) collected at flowering stage from Urmia, Iran, in July, 2006. The identification of this plant was carried out according to flora of iranica (Rechinger, 1982). Flowers of this plant were separated and dried naturally on Laboratory benches at room temperature (23-24°C) for 10 days. The plant materials were powdered using an electric grinder. Hundred gram of the dried powders extracted with 70% ethanol.

The extract was concentrated using a rotary evaporator at a maximum temperature of 45°C and were then further dried in an oven at 40°C for 48 h and powdered again. The dried extract was then dissolved in distilled water to prepare solutions of different concentrations (0.25, 0.5, 1, 2 and 3% w/v).

Adults of *S. oryzae* and *T. castaneum* were used in the test. The *S. oryzae* adults used were taken from a culture that was kept in the Laboratory on whole wheat at 27±1°C, 65±5% RH and continuous darkness.

The *T. castaneum* adults were taken from a culture kept on wheat flour plus 5% brewers yeast (by weight) at 28±1°C and 65±5% RH. All individuals used in the test 7-10 days old. Also, commodity was untreated, clean winter wheat (variety *Zarrin*) that obtained from Agricultural Research Center of west Azerbaijan, Urmia, Iran, was used in the tests.

Bioassay: All tests were conducted at 25°C, 65% RH and continuous darkness. Fixed quantity (1 kg) of diet (wheat) was then sprayed with 100 mL of each solution, 0.25, 0.5, 1, 2 and 3% (w/v). Also, there was 1 kg of wheat which was sprayed with water alone and served as control. From each combination, four samples, of 50 g each, were taken. Each sample was placed in a small glass pots (7cm diameter and 8.5 cm height). Twenty five *S. oryzae* adults were introduced into each glass pots and then covered with nylon mesh secured with rubber bands. The pots were placed in incubators, at the conditions described above. Dead adults were counted 24 and 48 h, 7, 14 and 21 days later. The same procedure was repeated for *T. castaneum* and for two species repeated four times (Athanassiou *et al*., 2005).

Progeny production count: After the 21 day mortality count, all adults (dead and alive) were removed and the glass pots were left in the incubators at the same conditions for an additional period of 45 day. Then, the glass pots were opened and the emerged individuals were counted. All the emerged *S. oryzae* individuals were adults, because the larvae of this species develop inside the grain kernels, whereas for *T. castaneum* apart from adults, immature also were recorded, given that this species is an external feeder (Athanassiou *et al*., 2005).

Data analysis: Control mortality was corrected by using the formula Abbott (1925). The data were arcsine transformed before analysis. The mortality counts were analyzed separately for each species, by using the GLM procedure (SAS, 1996), with insect mortality as the response variable and dose rate and exposure interval as main effects. The dose required to kill 50% of the insect (LD_{50}) was estimated using probit analysis (SPSS, 1999). The Percentage of reduction in progeny production was determined by the [(No. Progeny in control-NO. Progeny in treatment)/No. Progeny in control×100] formula (Aldryhim, 1995).

RESULTS

Mortality of *S. oryzae* adults: All main effects as well as associated interactions were significant at the p = 0.000 level (dose: F = 38.6; df = 4; exposure: F = 398.8; df = 4; dose × exposure: F = 8.37; df = 16).

Mortality of the exposed *S. oryzae* adults increased with the increase of the exposure interval and dose rate (Fig. 1). On wheat treated with the lowest botanical dose rate almost 53% of the exposed adults were still alive at the 21 day exposure interval (Fig. 1), whereas after 21 day of exposure, adult mortality on diet treated with 3% (w/v) botanical dose rate reached 100% (Fig. 1).

Mortality of *T. castaneum* Adults. All main effects as well as associated interactions were significant at the p = 0.000 level (dose: F = 83.5; df = 4; exposure: F = 136.5; df = 4; dose × exposure: F = 31.5; df = 16).

As noted for *S. oryzae*, mortality increased with the increase of the exposure interval and dose rate (Fig. 1). In contrast with other species, mortality of *T. castaneum* adults at the low dose rates was negligible (Fig. 1), moreover, mortality at the highest dose rate was almost 64% after 21-d exposure time (Fig. 1).

Moreover, LD_{50} value for *S. orysae* lower than required value for *T. castaneum* (*S. oryzae*. LD_{50}: 3.79, df: 3, chi square: 1.5 and *T. castaneum*. LD_{50}: 6.39, df: 3, chi square: 0.62) therefore, in is clear from the results that the, botanical extract on *S. oryzae* was effective.

Progeny production: The application of this plant material significantly reduced progeny production. No progeny was found in wheat treated, therefore, for two species, complete suppression of the progeny production was observed in the treated commodity in comparison with the control, even in the lowest dose rate (100% reduction in progeny production for *S. oryzae* and *T. castaneum* at all dose rates).

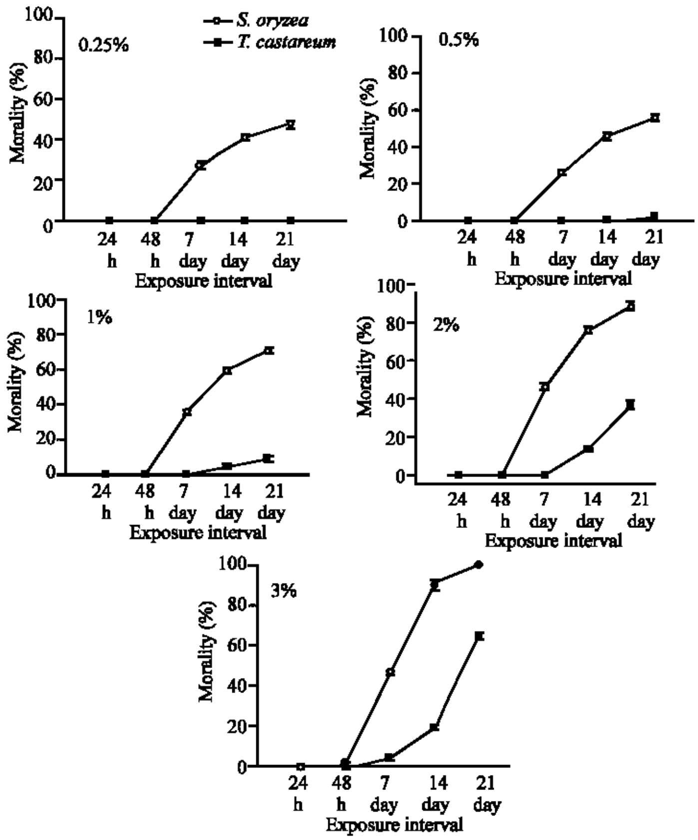

Fig. 1: Mean mortality (±SE) of *S. oryzae* and *T. castaneum* adults exposed for 24 and 48 h, 7, 14 and 21 day in wheat treated with 0.25, 0.5, 1, 2 and 3% (w/v) botanical extract

DISCUSSION

The result of the current study suggest that this botanical extract is effective against *S. oryzae* and *T. actinium* on wheat, but its effectiveness is highly determined by the characteristics of the dose rate and the exposure interval. Also, the same dose rate in not equally effective between the two species examined, except for effect on progeny production. Botanical extract is effective against *S. oryzae* than *T. castaneum*. Moreover, results show that for this species application rates and/or longer exposure intervals are needed to obtain a satisfactory level of mortality.

However, present results indicated that higher concentrations of this botanical extract for a relatively short period are much more effective than lower concentrations for a long period. In contrast is its ability to reduce progeny production in the treated grain. In our tests, progeny production of all species was inhibited completely in all the dose rates, indicating that even if oviposition occurred before death, the activity of botanical extract during the first molt of larvae was satisfactory.

Females of *S. oryzae* lay their eggs in the external part of the kernel (Brich, 1945; Golebiowska, 1969) and it is likely that newly hatched larvae are exposed to botanical before entering the kernel. But, in *T. castaneum* generally, F_1 numbers were extremely low, even in the control commodity; this finding could be due to the fact that *T. castaneum* cannot be developed easily in sound seeds (Aitken, 1975).

The finding of our study agree to earlier reports that indicated that most plant extracts though to have insecticidal properties and can control pests through affecting other biological activities (Berenbaum, 1989; Jacobson, 1989; Ascher, 1993; Schmutterer, 1995; Mostafa *et al*., 1996; Musabyimana *et al*., 2001).

From the progeny production of these insects, emergence of adults insects from all control samples indicated that tested insects were capable of effective oviposition and that prevention of progeny emergence was exclusively due to treatment. Thus extract of *V. cheiranthifolium* either suppressed oviposition or killed the larvae hatching from eggs laid in the medium culture. These results suggest that there may be different compounds in extracts possessing different bioactivities.

Similar observations on other plant extracts effect on several insects have been studied. For example, Sadek (2003) showed that the time of pupation of *Spodoptera littoralis* (Boisduval) of larvae increased by the extract of *Adhatoda vasica* (Nees). Jeyabalan *et al.* (2003) have reported that extract of *Pelargonium citrosa* (Van leenii), prolonged the duration of larval instars and the total developmental time of *Anopheles stephensi* (liston). Zhong *et al.* (2001) have also highlighted that extract from *Rhododendron molle* (G. Dorn) flowers extend the duration of duration of developmental of *Pieris rapae* L.

Present results have shown that *V. cheiranthifolium* Boiss. Posses high insecticidal activity on *S. oryzae* and *T. castaneum*. Abbassi *et al.* (2003) have found that same effect on desert locust *Schistocerca gregaria* (Forskal).

We can conclude that this study suggest that ethanolic extract of *V. cheiranthifolium* possesses toxic principles with significant insecticidal effect and could be a potential grain protectant against *S. oryzae* and *T. castaneum*.

Today, the environmental safety of an insecticide is considered to be of paramount importance. The world flora has a variety of plant species and in order to increase the number of plants used for pest control, more studies should be carried out. Thus, a variety of effective substance found in different plant species could be discovered. Consequently, substances alternative to many chemical pesticides, with pollute our natural sources and threaten our future, can be found. In addition, cheaper pesticides can be obtained and environmental pollution will gradually decrease.

REFERENCES

Abbassi, K., Z. Atay-Kadiri and S. Ghaout, 2003. Biological effects of alkaloids extracted from three plants of Moroccan arid areas on the desert locust. Physiol. Entomol., 26: 232-236.

Abbott, W.S., 1925. A method of computing the effectiveness of an insecticide. J. Econ. Entomol., 18: 266-267.

Aitken, A.D., 1975. Insect Travelers, I. Coleoptera, Technical Bulletin 31. HMSO, London, United Kingdom.

Aldryhim, Y.N., 1995. Efficacy of the amorphous silica dust, Dryacide, against *Tribolium canfusum* Du Val and *Sitophilus oryzae* (L.) (Coleoptera: Tenebrionidae and Curculionidae). J. Stored. Prod. Res., 26: 207-210.

Arnason, J.T., B.J.R. Philogene and P. Morand, 1989. Insecticides of plants origin. Am. Chem. Soc. Symposium Series, Washington, pp: 387.

Ascher, K.R.S., 1993. Noncoventional insecticidal effects of pesticides available from the neem tree, *Azadirachta indica*. Arch. Insect. Biochem. Phys., 22: 433-449.

Athanassiou, C.G., D.C. Kontodimas, N.G. Kavallieratos and M.A. Veroniki, 2005. Insecticidal effect of NeemAzal against three stored-product beetle species on rye and oats. J. Econ. Entomol., 98: 1733-1738.

Berenbaum, R.M., 1989. North America Ethnobotanicals as Sources of Novel Plant Based Insecticides. In: Insecticides of Plant Origin. Arnason, J.J., B.R. Philogene and P. Morand (Eds.), ACS Sympos. Ser., 387: 11-24.

Birch, L.C., 1945. The influence of temperature on the development of the different stages of *Calandra oryzae* L. (Coleoptera: Curculionidae) and *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae). Aust. J. Exp. Biol. Med. Sci., 23: 29-35.

Golebiowska, Z., 1969. The feeding and fecundity of *Sitophilus granaries* (L.), *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae) and *Rhyzoperta dominica* (F.) (Coleoptera: Bostrychidae) in wheat grain. J. Stored Prod. Res., 5: 143-155.

Howe, R.W., 1965. Losses caused by insects and mites in stored foods and foodstuffs. Nutr. Abstr. Rev., 35: 285-302.

Isman, M.B., 2000. Plant essential oils for pest and disease management. Crop Prot., 19: 603-608.

Jacobson, M., 1989. Botanical Pesticides, Past, Present and Future. In: Insecticides of Plant Origin. Arnason, J.J., B.R. Philogene and P. Morand (Eds.), ACS Symposium Ser., 387: 1-10.

Jeyabalan, D., N. Arul and P. Thangamathi, 2003. Studies on effects of *Plargonium citrosa* leaf extracts on malarial vector, *Anopheles stephensi* Liston. Bioresour. Technol., 89: 185-189.

Kabaru, J.M. and L. Gichia, 2001. Insecticidal activity of extracts derived from different parts of the mangrove tree *Rhizophora mucronata* (rhizophoraceae) Lam. against three arthropods. Afr. J. Sci. Tech. (AJST), Sci. Eng. Ser., 2: 44-49.

Koul, O., J.S. Shankar and R.S. Kapil, 1996. The effect of neem allelo-chemicals on nutritional physiology of larval *Spodoptera litura*. Entomol. Exp. Applic., 79: 43-50.

Mostafa, T.Y., S.M. Mahgoub and M.S. Ahmed, 1996. The efficiency of certain plant powders against cowpea weevil *Callosobruchus maculates* (F.) (Coleoptera: Bruchidae). Egypt. J. Agric. Res., 74: 307-319.

Musabyimana, T., R.C. Saxena, E.W. Kairu, C.P.K.O. Ogol and Z.R. Khan, 2001. Effects of neem seed derivatives on behavioral and physiological responses of the *Cosmopolites sordidus* (Coleoptera: Curculioni-dae). Hortic. Entomol., 94: 449-454.

Prakash, A. and J. Rao, 1997. Botanical Pesticicide in Agriculture. CRC Press, New York.

Rechinger, K.H., 1982. Flora Iranica. No. 150, Graz: Akademische Druck-u. Velagsanstalt, pp: 462.

Rembold, H., 1994. Advances in Invertebrate Reproduction. Elsevier Sci. Publ., 3: 481-491.

Roy, B., R. Amin and M.N. Uddin, 2005. Leaf extracts of Shiyalmutra (*Blumea lacera*) as botanical insecticides against lesser grain borrer and rice weevil. J. Biol. Sci., 5: 201-204.

Sadek, M.M., 2003. Antifeedant and toxic activity of *Adhatoda vasica* leaf extract against *Spodoptera littoralis* (Lep., Noctuidae). J. Applied Entomol., 127: 396-404.

Sahayaraj, K. and R. Sekar, 1996. Efficacy of plant extracts against tobacco caterpillar larvae. Int. Arach. Newslett., 16: 38.

Sahayaraj, K. and M.G. Paulraj, 1998. Screening the relative toxicity of some plant extracts to *Spodoptera litura* Fab. (Insecta: Lepidoptera: Noktuidae) of groundnut. Fresenius Environ. Bull., 7: 557-560.

SAS, 1996. SAS/STAT User's Guide. Release 6.12 Edn., SAS Institute, Cary, NC.

Schmutterer, H., 1995. The neem tree *Azadirachta indica* A. Juss and other Maliaceous plant sources of unique natural products of integrated pest management medicine, industry and other purposes. VCH, Weinheim, Federal Republic of Germany.

Senthilkumar, N., D. Jeyabalan and K. Murugan, 1997. Antifeeding and growth inhibiting effect of neem leaf exudates on *Spodeptora litura*. Indian J. Entomol., 59: 151-154.

SPSS, 1999. SPSS for Windows User's Guide, Release 10. SPSS Inc., Chicago.

Stevenson, P.C., W.M. Blaney and M.S.J. Simmonds, 1993. Developmental inhibition of *Spodoptera litura* (Fab.) larvae by a novel caffeoylquinic acid from the wild groundnut, *Arachis paraguariensis* (Chod, et Hassl). J. Chem. Ecol., 19: 2917-2933.

Weaver, D.K. and B. Subramanyam, 2000. Botanicals. In: Alternative to Pesticides in Stored-Product IPM. Subramanyam, B.H. and D.W. Hagstrum (Eds.), Kluwer Academic Publishers, MA, 303-320.

White, N.D.G., 1995. Insects, Mites and Insecticides in Stored Grain Ecosystems. In: Stored Grain Ecosystem. Jayas, D.S., N.D. White and W.E. Muir (Eds.), Marcel Dekker, New York, USA., pp: 123-168.

Yang, R.Z. and C.S. Tangs, 1988. Plants used for pest control in China: A literature review. Econ. Bot., 42: 376-406.

Zettler, J.L. and G.W. Cuperus, 1990. Pesticide resistance in *Tribolium castaneum* (Coleopteran: Tenebrionidae) and *Rhyzopertha dominica* (Coleoptera: Bostrichidae) in wheat. J. Econ. Entomol., 83: 1677-1681.

Zhong, G.H., M.Y. Hu, Q.F. Weng, A.Q. Ma and W.S. Xu, 2001. Laboratory and field evaluations of extracts from *Rhododendron molle* flowers as insect growth regulator to imported cabbage worm, *Pieris rapae* L. (Lepidoptera: Pieridae). J. Applied Entomol., 125: 563-596.