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GROWTH REGULATORY ACTIVITIES OF INDIGENOUS PLANT EXTRACTS AGAINST RED FLOUR BEETLE, *Tribolium castaneum* (HERBST) (COLEOPTERA: TENEBRIONIDAE)

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Growth regulatory effects of extracts of four medicinal plants which including *Salsola baryosma, Pegnum hermala*, *Saussurea costus* and *Nicotiana tabacum* were evaluated and data regarding pupation and adult emergence of *Tribolium castaneum* (Herbst) was observed. Applications of various concentrations viz, 2.5, 5.0, 7.5 and 10.0% of acetone extracts of test plants were made on the wheat flour, data regarding pupation and adult emergence was collected after each 5 days of treatment till the emergence of adults. The results showed that plant extracts had significant effect on percent pupation and percent adult emergence and with increase in concentration their emergence decreased. The acetone extract of *N. tabacum* at maximum concentration (10%) showed minimum percent pupation (40%) and percent adult emergence (28%) followed by the plant extracts of *S. baryosma, P. hermala* and *S. costus*. The acetone extract of *S. costus* was proved least effective; it showed maximum percent pupation (88%) and percent adult emergence (78%) at lower concentration (2.5%). From these results it was concluded that the extracts of indigenous medicinal plants could be useful as growth inhibitors for suppressing the population of *T. castaneum*.
Keywords: Adult emergence, growth inhibition, plant extracts, pupation, stored grain insect pests

INTRODUCTION

During storage, huge losses are reported by insect pests of stored cereals (Jillani, 1983) because they reduce the quantity and quality of stored grains (Madrid *et al*., 1990). Stored cereals, pulses and their products are attacked by more than 600 species of beetle pests . About 10-25% post-harvest losses have been estimated worldwide due to insect damage, microbial deterioration and other factors (Mathews, 1993). The red flour beetle, *Tribolium castaneum* Herbst (Coleoptera: Tenebrionidae) is a prevalent pest of stored grains that lives in and feeds on wheat flour (Lu *et al*., 2010). The occurrence of this pest is primarily controlled by fumigant insecticides (Klementz, 2008). However, resistance has been developed due to their consistent and repetitive use against this pest (Kumar *et al*., 2011). Khan *et al*. (2012) reported that higher quantities and consistent use of pesticides like pyrethroids may lead to serious problems of hematological, biochemical, reproductive and pathological changes in the human body after entering into the food chain. Desneux *et al*. (2007) described the side effect of pesticides on non-target organisms including beneficial arthropods.

Consequently, it is a priority to find alternative ways for control of insect pests which should be environmentally safe and easily biodegradable (Vinayachandra and Chandrashekar, 2011). The plant kingdom could be a rich source of a large number of chemicals which could be developed as control agents against stored grain pests successfully (Arnason *et al*., 1989). Repellent, toxicant and anti-feedants effect from several plant products have been reported against a number of coleopteran that attack stored products (Huang *et al*., 2000; Papachristos and Stamopoulos, 2002; Tapondjou *et al*., 2002; Tripathii *et al*., 2002; Hasan *et al.*, 2006; Ali *et al*., 2012).

Plant extracts in controlling stored grain insect pests, may have several desirable effects, which includes fumigant activity (Suthisut *et al*., 2011), contact toxicity to some insect (Kim *et al*., 2011), anti-feedant (Stefanazzi *et al*., 2011), repellency (Carroll *et al*., 2011), or change in some biological parameters such as growth rate, development and reproduction (Papachristos and Stamopoulos, 2002). The purpose of this study was to estimate the effects of plant extracts of *S. baryosma* (Khar boti), *P. hermala* (Hermal), *S. costus* (Kust-e-shireen) and *N. tabacum* (Tobacco) on pupation and adult emergence against *T. castaneum*, which is one of the most destructive insect pests of stored products.

MATERIALS AND METHODS

***Collection and rearing of T. castaneum*:** The population of different ages of red flour beetle, *T. castaneum* (Coleoptera: Tenebrionidae) was collected from different storage

structures located in Faisalabad district, Punjab, Pakistan during the year 2011, and reared in the laboratory of Grain Research, Training and Storage Management Cell, UAF to achieve a homogenous population.

T. castaneum were cultured in transparent sterilized glass jars (1.0 kg) in a controlled atmosphere having temperature 30±1°C and relative humidity 65±5% (Hussain *et al*., 2009). To avoid the escape of beetles the rearing jars were covered with plastic lids having netting on it for aeration. The culture medium was wheat flour which is the preferred food of *T. castaneum*. In each jar hundred adults of *T. castaneum* were released for the purpose of coupling and egg laying. After 5 days, the beetles were removed from these jars. The flour in which eggs were laid by the beetles was again put into the jars and kept under optimum rearing conditions in incubator (SANYO) to develop a homogenous population.

***Collection and preparation of plant extracts*:** The plants of *Salsola baryosma* and *Saussurea costus* were collected from the fields of district Layyah, while dried parts of plants *Pegnum hermala* and *Nicotiana tabacum* were purchased from market.

The leaves of test plants were cleaned by washing with sterilized water. After shade drying, the leaves were grounded in electrical grinder to convert them into crude powder. Extracts were obtained by mixing 50 g powder in 100 ml acetone for 24 hours while rotating at 220 rpm by using Rotary Shaker (IRMECO, OS-10). After filtration, the solvent from the filtrate was evaporated by placing the filtrate in the rotary evaporator (Hasan *et al.,* 2005; Sagheer *et al.,* 2013). After evaporation, the extracts obtained were considered as stock solution and were put in clean and air tight lid bottles and stored at 4.0°C in refrigerator. For different bioassay studies, four concentrations (2.5, 5.0, 7.5 and 10.0 %) of each plant extract were prepared from the stock solution using acetone as solvent.

***Bioassay studies*:** To check the effect of plant extracts on growth of red flour beetle, diet incorporation method of bioassay was used. For this purpose, four concentrations (2.5, 5.0, 7.5 and 10.0%) of each plant extract were applied on wheat flour. The treated flour was used as diet of the insect Where as for control treatment, the wheat flour was only treated with acetone. Each treatment was replicated thrice. Acetone was allowed to evaporate for one hour from the treated flour. 60.0 gm of treated flour was put into each plastic vial (100.0 ml). Twenty 3rd instar larvae of *T. castaneum* were released on the treated flour in each vial. These treatment vials were placed in incubator at 30±2°C and 65±5% R.H. Data regarding pupation and adult emergence were collected after regular intervals till the complete adult emergence.

Analyses of variance of the collected data were carried out using statistica-6 software. Means of treatments were separated by using Tuckey-HSD test at 5% significant level.

RESULTS

To determine the impact of plant extracts such as *Salsola baryosma, Pegnum hermala, Saussurea costus* and *Nicotiana tabacum* on the percent pupation and adult emergence, growth regulatory assays were performed against larvae of *T. castaneum* (Herbst). All larvae which were exposed to different concentrations of various plant extracts at controlled conditions showed statistically different results as compared to control treatments. The result discloses that the effects of various concentrations of acetone extracts of test plants are highly significant on percent pupation and adult emergence. As shown in Fig. 1, minimum pupation rate (40%) was observed in treatments having 10% concentration of acetone extract of the *N. tabacum*, followed by 50% pupation in response to the plant extract of *S. baryosma* at a maximum concentration. The pupation percentage ranged from 40 to 88% in all the treatments excluding control in which percent pupation was an average of 94%. The highest pupation (88%) was observed at the minimum concentration of the plant extracts of *S. costus*.

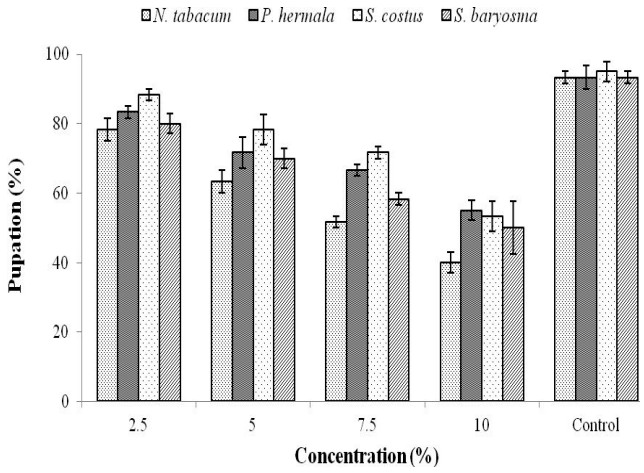

Figure 1. Concentration based response of plant extracts on the rate of pupation compared with the control treatment against *Tribolium castaneum* (Herbst).

N. tabacum extract induced 40, 52, 63 and 78% pupation at different concentration levels of 10, 7.5, 5 and 2.5%, respectively. In case of *P. hermala* the observed pupation was 55, 67, 72 and 83% at various dilutions of 10, 7.5, 5 and 2.5%, respectively and 53, 72, 78 and 88% pupation was noted in the case of *S. costus* extract. Whereas in case of *S. baryosma* the trend of percent pupation was in the following order: 50% <58% <70% <80% at concentration level of 10, 7.5, 5.0 and 2.5%, respectively.

The results also revealed that acetone extracts of test plants affected the development of *T. castaneum* as it is evident from highly decreased adult emergence rate (28%) exhibited by *N. tabaccum* extracts at 10% dilution level. Overall adult emergence ranged from 28 to 78%, excluding the control treatment. In which, *S. costus* again proved least effective as it showed relatively higher rate of adult emergence (78%) at a concentration level of 2.5%. Progressive increase in concentration was found to be inversely related to the adult emergence (Fig. 2).

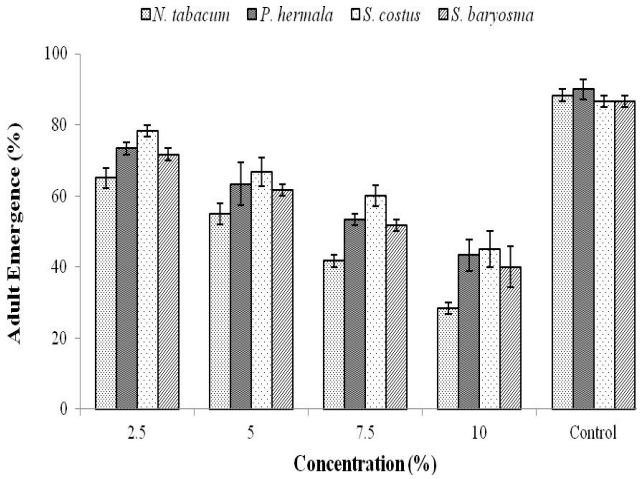

Figure 2. Concentration based response of plant extracts on the rate of adult emergence compared with the control treatment against *Tribolium castaneum* (Herbst).

N. tabacum extract resulted in 28, 42, 55 and 65% of adult emergence at serial dilutions of 10, 7.5, 5 and 2.5%, respectively. Similarly, in case of *P. hermala* the observed adult emergence was 43, 53, 64 and 73% at various dilutions of 10, 7.5, 5 and 2.5%, respectively. While 45, 60, 66 and 78% adult emergence was observed in the case of *S. costus*. On the other hand, in case of *S. baryosma,* adult emergence remained 40, 52, 62 and 72% at dose level of 10, 7.5, 5 and 2.5%, respectively.

Figure 3 indicates the overall effect of plant extracts on percent pupation and percent adult emergence against *T. castaneum.* From all test plants, the plant extract of *N. tabacum* was proved most effective as it showed minimum pupation (65%) as well as minimum adult emergence (49%). The trend of effectiveness of plant extracts was in the following order *N. tabacum>S. baryosma>P. hermala>S. Costus*. The response of the plant extracts varied statistically on pupation and adult emergence. Plant extracts of *P. hermala* and *S. costus* were proved least effective as they showed high rate of pupation (74%, 77%) and adult emergence (72%, 71%) respectively. Whereas, plant extract of *S. baryosma* also evidenced highly effective to lower down the pupation (70%) and adult emergence (59%) rate of *T. castaneum*. All these treatments with test plant extracts were proved significantly different from control treatments where 94% pupation and 88% adult emergence was observed.

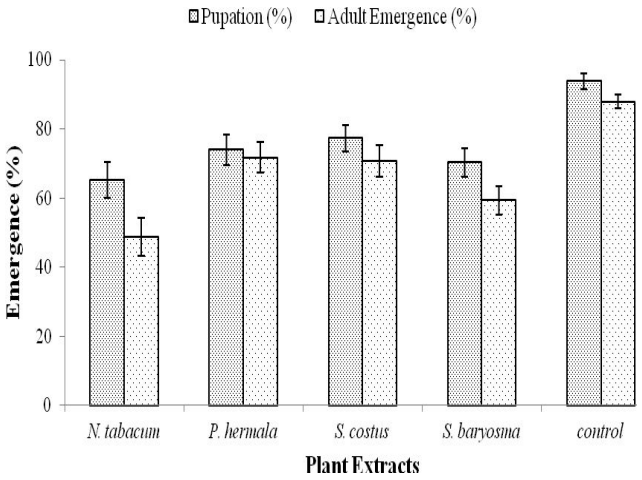

Figure 3. Average mean comparisons of the data regarding percent pupation and percent adult emergence of *Tribolium castaneum* (Herbst) in response to various plant extracts.

DISCUSSION

The present research work was carried out to investigate the impact of different concentrations of crude extracts of four plants (*S. baryosma, P. hermala, S. costus* and *N. tabacum*) on the growth and development of F_1 progeny of *T. castaneum.* The main theme of current study was to find out some safe alternative to highly toxic pesticides being used at present for the control of insect pests of stored grains. Four concentrations of each botanical extracts viz. 2.5, 5, 7.5 and 10% were used against stored grain insect, *T. castaneum* for their growth inhibition. The results disclosed that various concentrations of *S. baryosma, P. hermala, S. costus* and *N. tabacum* varied greatly against larvae of *T. castaneum* with regard to the rate of pupation and adult emergence. When the pupation and adult emergence rates of extracts were compared (Fig. 1 & 2), the highest rates of reduction of pupal and adult emergence were observed in bioassays with the acetone extract of *N. tabacum*.

Presently, chemical insecticides are serving as a tool for the control of stored grain pests (Epidi and Odili, 2009) but due to non-judicial use, these chemicals have resulted in the certain biological modifications in innocuous species and development of insecticide resistance (Rahman *et al*., 2009), consequently there is a need to replace these conventional harmful insecticides with biologically safer biocides (Talukdar *et al.*, 2004; Hassan *et al*., 2006; Epidi *et al*., 2008,

Epidi and Odili, 2009). Biocides of plant origin could be incorporated into the integrated pest management schemes because these are selective towards the insect pests, have less harmful effects on environment as well as other non-target fauna, can be applied easily and are locally available (Arnason *et al*., 1989; Schmutterer, 1992; Hedin and Hollingworth, 1997). Overall, ovicidal, repellent, deterrent and growth inhibitory activities are reported against a variety of insect pest attacking the stored commodities (Hill and Schoonhoven, 1981; Desmarchelier, 1994; Shaaya *et al*., 1997). High mortality, reduced growth rate and subsequent population reduction has also been reported due to the crude leaf powders against *T. castaneum* (Gandhi *et al.*, 2010). Percent emergence of larvae, pupae and adults was decreased with the increase in concentration of extracts of *Amaranthus hybridus*, *Calotropis procera*, *Salsola baryosma* and *Cuminum cyminum* (Sagheer *et al.,* 2011). Growth of larvae of *T. castaneum* inhibited significantly when they were fed on diet in which plant extracts of *Pegnum hermala, Ajuga iva, Aristolochia baetica* and *Raphanusra phanistrum* were incorporated (Jbilou and Sayah, 2006)**.**

From the results described above, it is concluded that plant extracts of *S. baryosma, P. hermala*, *S. costus* and *N. tabacum* reduced the growth regulatory activity of the test insect, *T. castaneum* at various serial dilutions. Results signify the potential of plant extracts to replace the conventional synthetic insecticides to control the population of *T. castaneum* in stored commodities. In the light of this study it is suggested that the commercial formulation of these plant extracts should be prepared for the control of stored grain insect pest and to conserve the environment.

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