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Effect of medicinal Plants Neem and Tulsi on the Serology of *Tenebrio molitor* (Tenebrionidae)

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Abstract

As cities are developing in measurement with a upward thrust in the population, the quantity of food requirement is increasing and becoming unmanageable. The fulfillment and serenity of food grains is a pressing want of our current and future. It has been proved that mealworms, the larvae of Tenebrio molitor Linnaeus, are found in the destruction of stored grain pest. Neem is one of the most broadly used pesticide and is effective in most of the insect pest. The Tulsi is also used as the control of pest. We have used the Neem(Azadirachta indica) and Tulsi(Ocimum sanctum) for the control of Tenebrio molitor. For the treatment of Tenebrio molitor. chemicals are used that are causing bioaccumulation and many diseases as reported by many researchers. Three firms of larvae have been fed chosen Neem and Tulsi as an only food, whilst a manipulate population used to be fed on oatmeal. The mass loss, dry depend on content and biochemical composition of mealworms have been assessed in the performed laboratory experiments. The protein attention in homogenates of the larvae was once determined by means of the Bradford method. To decide the degree of hydrolized carbohydrates we used anthrone method. The classical sulfo-phospho-vanillin assay (SPVA) used to be quantitate complete lipids in mealworms. These consequences allowed to examine the effect of neem and tulsi on the growth and development of Tenebrio molitor.

Keywords: *Tenebrio molitor,* Mealworm, neem & tulsi, serology.

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Introduction

Tenebrio molitor is the major pest of stored grain and commonly known as yellow mealworm. The mealworm is the largest of the insect species that attack stored grain and grain products. The use of synthetic pesticides was exceedingly popular in the pest management system employed all over the world.

Review of Literature

The indiscriminate and uncontrolled use of synthetic pesticides has led to problems (Dhaliwar et.al 2010). The Pest resistant to pesticides of the root extract of Decalepis hamiltonii against stored products (Rajashikar et.al 2010) suggest that we need to know the serological aspects of pest for making better pesticide. Neem and tulsi effects in reproductive organ development and reproduction of Tenebrio molitor by Hassan S.M.M. & Shaw P.K.(2021). Plant product as fumigants for stored product leave toxic Residue on food, water, air and soil (Rajendran & sriranjini 2008). Insect resistant is slow or less in storage management (Sharan, et.al 2014). There have no unknown environmental hazard in minor pest assuming measure status by cytochrome involvement in the interaction between plants and insects (Brattsten 1985). Presently, plant botanicals occupy a very small niche in the world of synthetic pesticides but the increasing environmental concern had led to surge in use of environmental sustainable and friendly “Green” alternatives. Reduction of progeny of some stored products in coleoptera by Schmidt, et.al (1991) suggest about the hormonal imbalance of the insect treated with insecticides. The effective insecticides on the growth of pests by fumigant toxicity against four major stored products (Shaaya et.al 1991) also shown an effective result. Due to all these various reasons, botanicals are considered as commercial valuable green pesticides and are gaining tremendous impetus(Aruna et.al 2015). Although there was some problems faced during commercialization and application [Maul et.al 2007] and [Yang, et.al 2015]. The ontogenic development of digestive system in mealworm Larva by Rodjaroen et.al(2020) & Hassan and Shaw(2020). The present article consists of extensive information on use and mode of action of different Botanical and their components against stored grain pest.

Materials and method

In natural conditions, beetles are specified in decaying and rotting bark of deciduous trees. The larvae and adult beetles feed on dead organisms and remain in warehouses cereal products, pantries, fowl farms and dovecotes. Experiment was carried out in the parallel 4 groups (20 specimens each, the parameter for selecting 20 specimens in each group was their similar mass). Larvae were weighted weekly. The variants used in tests were as follows: larvae eating oatmeal (control); larvae eating Tulsi; larvae eating Neem; starved larvae. After 3 weeks larvae were homogenized using cold mortar and pestle with ice-cold 0.1M phosphate buffer pH 7.4, 1mM EDTA (1:10 w/v). For carbohydrates assay aliquots of homogenates were frozen directly for further analysis. Required aliquots for lipid assay were treated with chloroform: methanol (1:2 v/v). The rest of homogenate were centrifuged at 11 000 x g, 5 min and frozen in aliquots at -20°C in 2ml Eppendorf vials for protein assay. Firstly, we determined the mass loss of studied larvae. Secondly, we studied biochemical composition of mealworms. Protein content was assessed with the Bradford assay [Nampoothiri, et.al 2010]. To determine the level of hydrolized carbohydrates we used anthrone reagent [Handel el.al,1985] and measure the absorbance of samples at 625 nm. The classical sulfo-phosphovanillin assay (SPVA) was used to quantitate the total lipids in mealworms [Handel, et.al, 1985]. Absorbance was measured spectrophotometrically at 525 nm. In all assays multipoint standard curves with high linear regression (R2: protein curve = 0.91; carbohydrate curve = 0.99; lipids curve = 0.99) were used. All statistical analyses were performed in Statistica software package. Differences in mass change of mealworms and mass loss for two types of materials with the duration of exposure were tested for each factor using analysis of variance (ANOVA).

Results and discussion

Mass loss content

We determined the percentage of mass changes of larvae and mass loss of Plant extracts after 7, 14 and 21 days from the beginning of the experiment. The results are presented in appropriate Tables (Tab. 1) and Figures (Fig. 1). The results show the biggest mass change (-16.9) for studied larvae fed Neem (Tab. 1& Fig. 1). The masses loss of studied materials were significantly different ($P < 0.05$). The significant increasing trend in mass change (25mg) was observed in the control sample (Tab. 1) which is not surprising. As it was expected, we observed the lowest mass change of starved larva (Table 1) (Fig. 1). The significant differences were observed between control mealworms eating oatmeal and larvae fed Neem ($P < 0.05$). Suprisingly, the mass of larvae is greater in starved condition compared to mass of larvae fed on Neem. This result indicates that mealworm invested more energy in detoxification process and hence loss their mass in terms of proteins, carbohydrate and lipids.

Protein content

The protein content in the tissues of mealworms not varied significantly depending on the food type. The results are shown in Fig. 2. As the proteins are the main building material for the body of insect they can be used in the hydrolyzed form as fuel for metabolism or converted to glucose through gluconeogenesis. As our results show protein levels protein content varied between 16–25 ng/mg and were not significantly affected by the food type. Some authors noticed that protein synthesis decreases during food deprivation [Renault 2003 & Bosquet 1977], leading to a decrease in protein levels as observed here in starved larvae and mealworms fed Tulsi and Neem. As it was suggested by Renault, et.al (2003) an insect's recovery from starvation may depend upon its mobility and readiness for searching food, thus a compromise between the demand for carbohydrates and the preservation of muscle proteins is needed. On the other hand, such materials like Neem and Tulsi are not a good source of energy at all which lead to the depletion of proteins in mealworms.

Carbohydrates content

Carbohydrates provide most of the energy during insect life. Some insects metabolize glycogen during the initial stages of starvation, then switch to lipid and protein metabolism when carbohydrates are gone. Other species, however, depend more heavily on lipid metabolism during exposition to stress factors [Marron et.al, 2003]. The lowest carbohydrates content was registered for mealworms fed Neem and Tulsi as these materials are not reliable food source for development and life cycle completion (Fig. 3). As it was shown in the experiment the carbohydrates content was not significantly depleted comparing to the control mealworms fed oatmeal.

Lipids content

The most significant changes during stress and starvation are usually observed in the levels of triglycerides, as the level of these compounds decreases significantly. The most common source of energy in insects during starvation period is the oxidation of fatty acids stored in the form of triglyceride, large amounts of lipids are accumulated in insect fat body, with fat content exceeding 50% of the total dry mass in selected studied insects [Marron et.al,2003]. Levels of stored fat significantly help to survive during starvation and environmental stress, the level of triglyceride is always connected with the duration of starvation period [Marron et.al,2003]. In our studies the amount of lipids is the highest in the control mealworms fed oatmeal which is in the line with the observations of other authors suggesting that starvation is connected with the rapid fat reserves burning [Marron et.al 2003] (Fig. 4). Larvae under control have the highest amount of lipids comparing to mealworms fed Neem and Tulsi which suggest that although they decompose these materials they do not ingest it and use the energy effectively for life activities.

Table 1. Mass changes of mealworms caused by waste eating and starvation with the comparison of control larvae.

	Mass loss 0 day (mg)	Mass loss 7 day (mg)	Mass loss after 14 days (mg)	Mass loss after 21 days (mg)
control	0	5	15	25
Tulsi	0	-3	-9	-15
Neem	0	-4	-12	-16.9
starved	0	-4	-10	-12

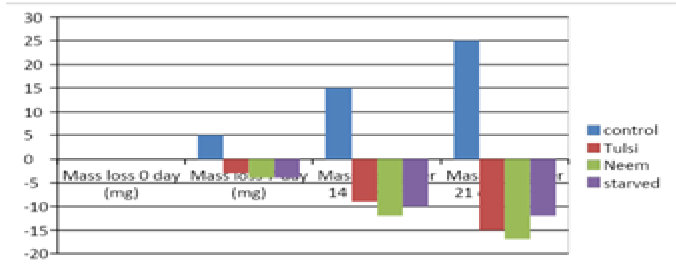

Fig. 1. The average mass of larvae after 7, 14 and 21 days of the experiment.

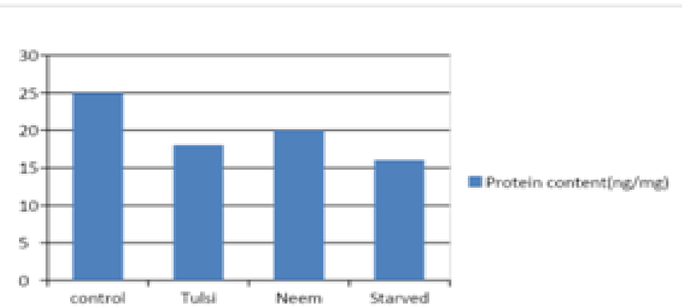

Fig. 2. The protein content in mealworms fed different types of food and starved (ng/mg).

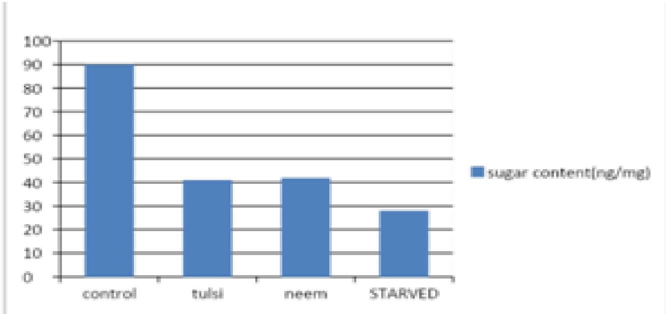

Fig. 3. The carbohydrates content in mealworms fed different types of food and starved (ng/mg).

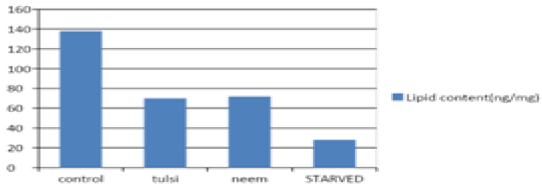

Fig. 4. The lipid content in mealworms fed different types of food and starved (ng/mg).

Objective of the Study

To assess the effect on serology of the stored grain pest Tenebrio molitor by using extracts of Neem (Azadirachta indica) and Tulsi (Ocimum sanctum). These extracts are being used to analyze the effects on lipid, protein and carbohydrate of Tenebrio molitor.

Conclusion

In our observations on processing commonly used medicinal plants such as Neem & Tusi with *T. molitor* larvae we recorded that all specimens survived three weeks of experiment period, although they required much more effort to maintain their vital functions feeding Tulsi, Neem or starving. The conducted biochemical assays confirmed that stored energy in the form of Proteins, lipids and carbohydrates are used for keeping the basic metabolic rates. The mass loss of all studied materials (Tulsi, Neem, STARVED) suggest that they are eaten by larvae and decomposed efficiently by microorganisms presented in the gut of mealworms. On the other hand, the mass changes of studied specimens indicate that all of these materials are not very efficient source of energy for mealworms, they only allow them to survive but not to deliver a sufficient energy as mealworms lost weight constantly. As it was expected. The interest of researcher in T. molitor larvae and their adult is mainly due to their nutritional potential and their adults are serious grain pest.

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