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ORIGINAL ARTICLE

Effect of Herbal Plant Extracts on Total Protein in *Tribolium castaneum* (Herbst)

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ABSTRACT
The red flour beetle, Tribolium castaneum (Herbst) is a common insect pest and is the most attacking pest that infests stored flour, cereals and other food grains. The herbal plant extracts of Eugenia jambolana (Jamun) seed and Aloe vera (Grihta Kumari) gum were tested against adult T. castaneum. Both the extracts exhibited time dependent reduction in total protein. Reduction in protein content had lead to the mortality of the insect pest T. castaneum. The mean percent mortality rate was higher in Aloe vera (Grihta Kumari) gum extract indicated its effectiveness against T. castaneum.
Key words: T. castaneum, Plant extracts, E. jambolana, Aloe vera, Total Protein.

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INTRODUCTOION

The Granaries, Mills, Ware Houses etc. are the most common places where red flour beetle, *Tribolium castaneum* (Herbst) is easily found. It's a major pest of flour and cereals that severely damages stored wheat, grams and other food grains in quality and quantity as well (1). It causes higher weight losses in wheat than other cereals. To control this pest is a challenge. Generally synthetic chemicals like methoprene, permethrin, cypermethrin, deltamethrin and fenvalerate etc. were tested and applied to control *T. castaneum* (2). However, these synthetic chemicals are leading to dangerous side effects. Few such issues are like pest resurgence, damages to non-target organisms, environmental damages, uneconomical, development of resistance in pests to such chemicals, toxic effects to users, etc. (3, 4).

To encounter such dangerous side effects, alternative pest control methods were tried. Researchers have tested some alternatives (5, 6) against *T. castaneum.* Some plant extracts, oils, etc. were tested as alternative products (7, 8 and 9). The biological activities of plant products were identified for the control of stored-grain pests. Few of them have shown remarkable results in controlling *T. castaneum.* It may be due to their rich insecticidal potential. Such plant products are advantageous over synthetic agents, as their application is comparatively environment and user friendly.

Families of these plant products exhibited repellent, insecticidal, anti-feedant, etc. properties against insect pests of stored commodities (6, 10). Few Citrus plant species are sources of botanical insecticides and few contain secondary metabolites that showed insecticidal activity against such insects (2). The herbal insecticides are safe and eco-friendly (11, 12 and 13). Primarily the toxicity of such herbal compounds is responsible for the potentiality of these plant extracts against pests (14).

This preliminary investigation was undertaken to study the efficacy of locally available medicinal herbal plant extracts of *Eugenia jambolana* (Jamun) seed and *Aloe vera* (Grihta Kumari) gum. These were tested against the adult stored grain insect pest *Tribolium castaneum*. It was aimed to investigate the effectiveness of these extracts on total protein of adult *Tribolium castaneum*. The mortality of these insects was also observed.

MATERIAL AND METHOD

Wheat flour was sterilized at 60^{0}C for 24 hours in an oven. A mixture of wheat flour with powdered dry yeast in a ratio of 19:1 was used as a food medium throughout the experimental period to maintain the culture. *T. castaneum* culture was obtained from National Chemical Laboratory, Pune. These cultures were kept under ambient conditions (28± 2°C and 65% R.H).

The culture protocol of beetles was established to get a regular supply of the newly formed adults for the experiment. *E. jambolana* (Jamun) seed and *Aloe vera* (Grihta Kumari) gum extracts in powdered form were procured from M/s Amsar Laboratories, Indore (MP, India). The trial experiments were conducted using these extracts. The experiment was designed at 8 gm concentration for both the individual extracts.

The insecticidal activity of *E. jambolana* (Jamun) seed and *Aloe vera* (Grihta Kumari) gum extracts were tested against adult insects of *T. castaneum*.

The Jamun and *Aloe* vera plant extracts were weighed (8 gm) and were poured in 100 ml food grade PVC bottles. Then twenty adult insects of *T. castaneum* were released in the bottles and the bottles were covered with muslin cloth for aeration and to avoid any insect to escape from the bottles. Three replicates were taken for each sample. The experiments were set up for 24, 48 and 72 hours. After every exposure period the bottles were opened and the insect survival and mortality was observed. The control insects were fed with normal wheat floor.

For Protein estimation live treated insects were homogenized with 1%, 200 µl SDS. After incubating at 55^o C for 10 minutes, it was centrifuged for 10 minutes at 3,000 RPM. In the supernatant 50%, 1 ml TCA was added and then centrifuged at 12,000 RPM for 20 minutes at 4^o C. The pallet was dissolved in 1 N, 200 µl NaOH and centrifuged for 10 minutes at 5000 RPM. 100 µl supernatant was added to the dilutions and was processed for Lowry's method (15). A standard graph was plotted between observances at 750 nm by spectrophotometer (Shimadzu, UV 1800) against the range of BSA. The experiment was repeated in triplicates.

The mean of mortality was calculated and further corrected by using the Abbot's formula (16).

RESULTS AND DISCUSSIONS

The results of the total protein in the adults of *T. castaneum* after treatment with *E. jambolana* (Jamun) seed and *Aloe vera* (Grihta Kumari) gum extracts in powdered form were shown in Table 1.0 and Fig. 1.0. It was observed that the mean total protein contents in all exposures against the controls were significantly reduced. The decrease in total protein was time dependent. With increase in time the percentage of total protein depletion increased. Total protein depletion was higher in *Aloevera* than in Jamun.

Decrease in total protein content may be due to the treatment of the insects with toxic components is a common phenomenon (17). Reduction in total protein content may be due to degradation of amino acids. The depletion in free amino acids affect adversely on total protein contents. The protein depletion further delays the metamorphosis (18).

This depletion of amino acids forces the insects to enter into the TCA cycle as a keto acid (14). This means that the insect releases the stored energy to overcome the toxic effects of herbal compounds. This additional energy is used to compensate the lower energy caused by stresses (14, 19). It revealed that total protein content depleted in order to compensate the energy lost due to the stress developed by toxicity of the extracts (20). The loss in total protein content is the root cause of the increased mortality of insect *T. castaneum* (21, 22).

The mortality increased with the exposure time. The mean percent mortality was higher in Aloe vera gum extract than the Jamun seed extract. The higher mortality rate of *T. castaneum* correlated the higher depletion of protein when treated with Aloe vera. The present findings were in accordance with previous studies.

Table 1.0 Total Protein Content (mg/ml)

TIME → / EXTRACT ↓	24 Hrs	48 Hrs	72 Hrs
CONTROL	1.2	1.19	1.21
JAMUN	1.06	0.86	0.55
ALOE VERA	0.9	0.55	0.35

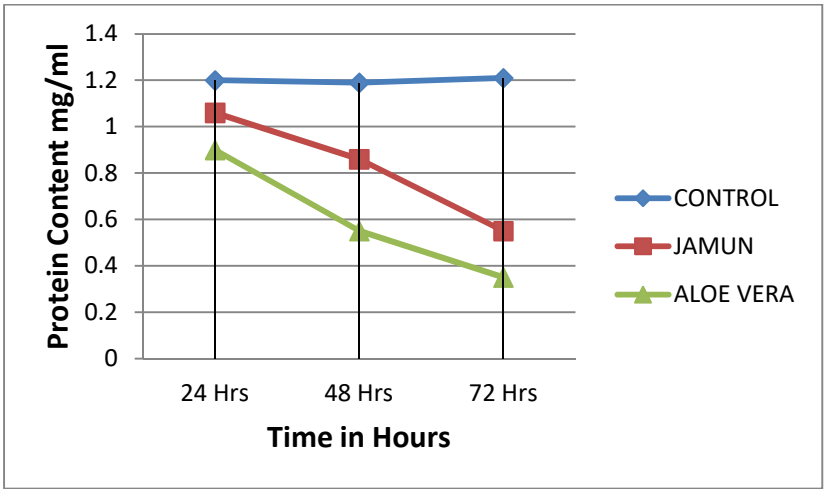

Fig. 1.0 Total Protein Estimation in adult *T. castaneum*

CONCLUSION

In the present study both the extracts were found effective against the adults of *T. castaneum*. But the Total protein content declined comparatively higher in the *Aloe vera* (Grihta Kumari) gum extract than *E. jambolana* (Jamun) seed extract. The present study revealed that both the experimental compounds possibly be eco-friendly and safe alternatives to control the infestation of *T.* castaneum. However, it is necessary to isolate the bioactive and secondary metabolites of these extracts to understand the exact mode of action. Further studies are required before the commercial applications.

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