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REPELLENT ACTIVITIES OF SOME INDIGENOUS MEDICINAL PLANTS AGAINST THE STORED GRAIN PESTS *TRIBOLIUM CASTANEUM* (HBST.) (COLEOPTERA: TENEBRIONIDAE)

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ABSTRACT: The petroleum (Pet.E), methanol (MeOH) and chloroform (CHCl₃) extracts of *Rauvolfia canescens* (whole plant), *Desmodium heterocarpon* (whole plant) and *Vitex negundo* (leaf CHCl₃ MeOH and root Pet. E.) have been thoroughly screened through residual film assay against *T. castaneum* adults and the repellent activity tests were also carried out on the same to find biological activities. The residual film assay offered promising results with remarkable activity against the adult beetles of *T. castaneum*. A perusal of the data achieved in this experiment clearly showed the presence of insecticidal properties in *Rauvolfia canescens* (whole plant), *Desmodium heterocarpon* (whole plant) and *Vitex negundo* extracts as well as traces of repellent potential. For repellency *D. heterocarpon* (whole plant /Pet. Ether) extract and *V. negundo* (leaf /Pet. Ether) extract were weakly active (P <0.05) and *R. canescens* (whole plant /Pet. Ether) extract and *V. negundo* (root /Pet. Ether) extract were mildly active (P<0.01); while the rest showed no repellent activity against the adults beetles of *T. castaneum*. Comprehensive phytochemical analyses of the test plants for its repellent as well as the physiological studies of the active ingredients are very much to be solicited for their effective use in the future pest control and pharmaceutical endeavors.

INTRODUCTION: Pests are a major problem in stored grain field crops all over the world because they reduce the quantity and quality of grain. The overall damage caused by insect pests, microbial deterioration, and other factors is estimated to be 10- 25% of worldwide production annually¹.

Tribolium castaneum (Herbst) is considered as a serious pest of stored grains feeding on flour, cereals, meal, crackers, beans, spices, pasta, dried pet food, dried flowers, chocolate, nuts, and dried museum specimens²⁻⁴. To overcome these problems, pesticides are often considered to be the most potent control technology for pests. Large use of Chemicals as pesticides in crop protection could be environmental pollutants and have adverse effects on animals and human beings.

Also, continuous or heavy use of some pesticides has often created serious problems which occurring

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from factors such as direct toxicity to parasites, bacteria, predators, pollinators, fish, and man^{5,6}. Consequently, there is a pressing need to build up suitable, harmless, environmentally friendly and low-cost alternatives. Plant-derived materials played a significant role in pest management which is a rich source of bioactive chemicals and considered non-pollutant, less toxic and easily biodegradable. Many plants commonly considered as safe contain noxious compounds, which may provide them unsafe for both animals and humans to consume^{7,8}. Insecticidal activity of many plants against numerous insect pests has been demonstrated^{9,10}. The plants used for pest insect control and set up that there is a strong association between medicinal and pesticidal plants¹¹.

Medicinal plants have been a major resource of a cure for human diseases since time immemorial. It is no wonder that the world's one-fourth population, i.e. 1.42 billion people, are reliant on traditional medicines for the treatment of various ailments¹². The insecticidal activity of essential oils and other plant extracts has been evaluated against some major agricultural pests¹³.

Despite the well-known appreciation that many plants have insecticidal properties, the commercialization of new botanicals can be hindered by some issues, and only a handful of pest control products directly obtained from plants are in use¹⁴. Essential oils found from different plant species hold ovicidal, larvicidal, and repellent properties against diverse insect species and are considered as environmentally compatible pesticides¹⁵.

This study aims to evaluate the presence of pest control potentials (insecticidal action and repellent) of the selected medicinal plants through dose-mortality assay and repellent activity test.

MATERIAL AND METHODS:

Insect Collection and Rearing: *T. castaneum* were used to find out the repellent property of decided three medicinal plants. Heterogeneous samples were collected from different godowns of Rajshahi and were reared on wheat flour and grain at 30 °C ± 0.5 °C in the laboratory.

Selection of Plant Materials: In this whole investigation plant of *Rauvolfia canescens* and

Desmodium heterocarpon and leaf stem bark and the root of *Vitex negundo* have been selected for the presence of toxic, as well as, bio-active constituents since the plants are well known as medicinal plants and also considered to contain toxic constituents. In the case of very small plants, such as herbs, shrubs, grass, etc.

Normally, the whole plant is subjected to extraction because the distribution of constituents generally not varies too much. Being a large timber plant, the distribution of compounds in different parts of this plant is obviously different. The presence of constituents in the heartwood may disappear in the leaves; similarly, constituents in the roots may not be the same that present there in the fruits.

Collection of Plant Materials: *Desmodium heterocarpon* (whole plant) and *Rauvolfia canescens* (whole plant), and *Vitex negundo* leaf, stem bark and roots were collected from Natore district in Bangladesh.

Preparation of Plant Materials for Extraction:

Whole Plant: The whole plant *Desmodium heterocarpon* and *Rauvolfia canescens* were collected. Excess soil from the roots removed, without washing. The plant material then spread out to dry without heaping the material together. This was done under the shade avoiding direct sunlight or in a well-ventilated room.

Leaves: After collection of leaves of *Vitex negundo* were spread out to dry without heaping the material together. It was done under the shade avoiding direct sunshine.

Root: Roots were collected by digging up without damaging them and shake and brush away excess soil without washing them with water. Root was cutting into small pieces as thin as possible. After collection roots were dry thoroughly in well-ventilated place. After drying well, the plant materials were powdered in a grinder machine avoiding excess heat during grinding.

Chemical Extraction of the Collected Materials:

There are two methods for extracting compounds from plant materials. Which one to choose, depends on whether the aim is to extract the more polar compounds (especially glycosides) which are present in the cell vacuole, or to obtain the less

polar aglycones present on the surface of the plant, in aerial parts heartwood or roots.

In the present study, three solvents were selected to extract the materials collected. The ground dried materials, viz. whole plant, leaves, root, and stem were extracted with a sufficient amount of solvent (CHCl₃, MeOH and Pet. E) for each of the items. Separate extracts have been collected by the cool method after 48 h of plunging for each of the material. Extracts, thus, obtained are filtered and concentrated on a rotary evaporator at 40 °C and only as residue is left and kept in a refrigerator after labeling.

Repellent Activity of the Extracts: The repellency test used was adopted from the method (No. 3) of McDonald *et al.*, (1970) with some modifications by Talukder and Howse (1993, 1994). No significant difference was detected between the repellency of only solvent impregnated and untreated filter papers in tests designed to check for any possible influence of Pt. E. CHCl₃ and MeOH. The average of the counts was converted to percentage repellency (PR) using the formula of Talukder and Howse (1993, 1995):

$$PR = (N_c - 5) / 20$$

Where 'c' is the percentage of insects on the untreated half of the disc. Positive values expressed repellency and negative values for attractant activity.

Preparation of Doses with the Crude Extracts for the Repellency Test: A general concentration for each of the plant's extracts was selected as the stock dose for repellency application to make other successive doses by serial dilution to give 0.629, 0.315, 0.157, 0.079 and 0.039 mg cm⁻² concentrations.

Application of Doses in the Repellency Test: Half filter paper discs (Whatman No. 40, diameter 9 cm) were prepared and selected doses of all the Pet. E. CHCl₃ and MeOH extract separately applied onto each of the half-disc and allowed to dry out as exposed in the air for 10 min. Each treated half-disc was then attached lengthwise, edge-to-edge, to a control half-disc with adhesive tape and placed in a petri dish (diameter 9 cm), the inner surface of which was smeared with fluon to prevent insects

escaping. Control experiments by applying the only solvent into the Petri dishes were also set at the same time under the same condition. Three replications were maintained same as the surface film test. Being volatile the solvent was evaporated out within a few minutes. Then ten insects were released in the middle of each filter paper circle.

The orientation of the same was changed in the replica to avoid the effects of any external directional stimulus affecting the distribution of the test insects. Each concentration was tested five times. Insects that settled on each half of the filter paper disc were counted after 1 h and then at hourly intervals for 5 h.

Observation and Analyses of Repellency Data: Repellency was observed for 1 hour interval and up to 5 h successive hours of exposure, just by counting the number of insects in the treated and non-treated part of the filter paper spread on the floor of the 90 mm petri dish. The values in the recorded data were then calculated for percent repellency, which was again developed by arcsin transformation for the calculation of ANOVA.

RESULTS:

Bioassay on *T. castaneum* Adults:

Effects of *D. heterocarpon* (Fruit, Aerial Part, and Root) Pet. E. Extract against *T. castaneum* by Residual Film Assay: All the *D. heterocarpon* Pet. E. extracts (fruit, aerial part, and root) were tested against the adult beetles of *T. castaneum* through residual film assay, and results found promising.

The data was then subjected to probit analysis and the result has been presented in **Table 1**. To trace acute toxicity an observation of lethality was made after ½ h of application of the doses.

Effects of (whole plant) Extracts (MeOH) of *D. heterocarpon* against *T. castaneum* Adults: All the *D. heterocarpon* MeOH extracts (whole plant) were tested against the adult beetles of *T. castaneum* through residual film assay, and results found promising.

The data was then subjected to probit analysis, and the result has been presented in **Table 2**. To trace acute toxicity an observation of lethality was made after ½ h of application of the doses.

TABLE 1: LD₅₀ VALUES 95% CONFIDENCE LIMITS AND REGRESSION EQUATIONS OF FRUIT, AERIAL AND ROOT EXTRACTS (PET. E.) OF *D. HETEROCARPON* AGAINST *T. CASTANEUM* ADULTS

| Plant part | Exposure | LD ₅₀ value mgcm ⁻² | 95% confidence limit | | Regression equation | x ² value (df) |
|---|----------|--|----------------------|-------|-----------------------|---------------------------|
| | | | Upper | Lower | | |
| <i>D. heterocarpon</i> (Pet. E) (Whole plant) | 12 | 2.719 | 15.228 | 0.485 | Y=2.5694 + 1.6942X | 26.210(4) |
| | 24 | 1.912 | 4.475 | 0.817 | Y=1.7882 + 2.505X | 30.362(4) |
| | 36 | 1.608 | 2.884 | 0.897 | Y=1.6782 + 2.753X | 27.782(4) |
| | 48 | 1.246 | 1.697 | 0.915 | Y=1.6048 + 3.098X | 16.675(4) |

The LD₅₀ values were 2.719912, 1.91276, 1.608702 and 1.246667 mg cm⁻² for 12, 24, 36, 48 h of exposure respectively, while the regression equations were Y = 2.56944 + 1.694296 X, Y = 1.788212 + 2.505958 X, Y = 1.678262 + 2.753257 X and Y = 1.604852 + 3.098469 X; the χ² values along with their df were 26.21 (4), 30.36 (4), 27.78 (4) and 16.68(4) and the 95% confidence limits were 0.4858095 to 15.22803, 0.8174345 to 4.475772, 0.8971584 to 2.884578 and 0.9157986 to 1.697074 cm⁻² respectively

TABLE 2: LD₅₀ VALUES 95% CONFIDENCE LIMITS AND REGRESSION EQUATIONS OF FRUIT AND AERIAL EXTRACTS (MEOH) *D. HETEROCARPON* AGAINST *T. CASTANEUM* ADULTS

| Plant part | Exposure | LD ₅₀ value mgcm ⁻² | 95% confidence limit | | Regression equation | x ² value (df) |
|---|----------|--|----------------------|-------|----------------------------|---------------------------|
| | | | Upper | Lower | | |
| <i>D. heterocarpon</i> (MeOH) (Whole plant) | 12 | 0.919 | 1.07 | 0.786 | Y=2.081453+3.0283 456X | 0.669 (4) |
| | 24 | 0.571 | 0.73 | 0.446 | Y =2.644622 + 3.11253 X | 5.106 (4) |
| | 36 | 0.328 | 0.57 | 0.187 | Y =3.678537 + 2.55588 X | 1.087 (4) |

The LD₅₀ values were 0.9191304, 0.5711366 and 0.3288726 mg cm⁻² for 12, 24, 36, 48 h of exposure respectively, while the regression equations were Y = 2.081987 + 3.028942 X, Y = 2.644622 + 3.112533 X and Y = 3.678537 + 2.555884 X; the χ² values along with their df were 0.6697445 (4), 5.106449 (4) and 1.087931 (4) and the 95% confidence limits were 0.7869408 to 1.073525, 0.4466829 to 0.7302655 and 0.1877441 to 0.5760883 cm⁻² respectively

Effects of *Rauvolfia canescens* (Whole Plant) Pet. E. Extract Against *T. castaneum* by Residual Film Assay: All the *Rauvolfia canescens* Pet. E. extracts (whole plant) were tested against the adult beetles of *T. castaneum* through residual

film assay, and results found promising. The data was then subjected to probit analysis, and the result has been presented in **Table 3**. To trace acute toxicity an observation of lethality was made after ½ h of application of the doses.

TABLE 3: DOSE MORTALITY EFFECT OF THE *RAUVOLFIA CANESCENS* (PET.E) (WHOLE PLANT) AGAINST *T. CASTANEUM* ADULTS

| Plant part | Exposure | LD ₅₀ value mgcm ⁻² | 95% confidence limit | | Regression equation | x ² value (df) |
|--|----------|--|----------------------|--------|-----------------------------|---------------------------|
| | | | Upper | Lower | | |
| <i>R. canescens</i> (Pet. E) (Whole plant) | 12 | 2.717491 | 3.5488 | 2.0808 | Y = 3.284913 + 3.950284X | 1.115 (4) |
| | 24 | 2.314858 | 3.3348 | 1.6068 | Y = 3.73563 + 3.468548X | 11.802 (4) |
| | 36 | 1.760789 | 2.1910 | 1.4150 | Y = 3.836417 + 4.735647X | 36.202 (4) |
| | 48 | 1.40103 | 2.0008 | 0.9810 | Y= 4.393501 + 4.141413 X | 36.202 (4) |

The LD₅₀ values were 2.717491, 2.314858, 1.760789 and 1.40103 mg cm⁻² for 12, 24, 36 and 48 h of time exposure respectively, while the regression equations were Y = 3.284913 + 3.950284 X, Y = 3.73563 + 3.468548 X, Y = 3.836417 + 4.735647 X and Y = 4.393501 + 4.141413 X the χ² values along with their df were 1.115901(4), 11.80273(4), 19.96361(4) and 36.20296(4); and the 95% confidence limits were 2.080863 to 3.548891, 1.606854 to 3.334818, 1.415036 to 2.191024 and .9810495 to 2.000802 mgcm⁻² respectively

Effects of (Whole Plant) Extracts (MeOH) of *Rauvolfia canescens* against *T. castaneum* Adults: All the *Rauvolfia canescens* MeOH extracts (whole plant) were tested against the adult beetles of *T. castaneum* through residual film assay

and results found promising. The data was then subjected to probit analysis and the result has been presented in the **Table 4**. To trace acute toxicity an observation of lethality was made after ½ h of application of the doses.

TABLE 4: DOSE MORTALITY EFFECT OF THE RAUVOLFIA CANESCENS (MEOH) (WHOLE PLANT) AGAINST T. CASTANEUM ADULTS

| Plant part | Exposure | LD ₅₀ value mgcm ⁻² | 95% confidence limit | | Regression equation | x ² value (df) |
|--|----------|--|----------------------|--------------|----------------------------|---------------------------|
| | | | Upper | Lower | | |
| <i>R. canescens</i> (MeOH) (Whole plant) | 30 min | 6.516 | 2.665 | 1.593366E-02 | Y=2.931211 + 1.140429 X | 21.7135 (3) |
| | 12 | 0.692 | 0.79607 | 0.60294 | Y=1.8109779 + 3.793674X | 4.24917 (3) |
| | 24 | 2.097 | 1.11883 | 0.03932 | Y=4.5267778 + 1.470826X | 15.913 (3) |
| | 36 | 0.114 | 1.72659 | 7.63423 | Y=4.925497 + 1.2435748X | 12.987 (3) |
| | 48 | 1.82 | 1.44985 | 2.33105 | Y=5.634997 + 0.863283X | 2.3141 (3) |

The LD₅₀ values were 6.516959, 0.6928131, .2097686, 0.1148097 and 1.838398E-02 mg cm⁻² for 30 min, 12 h, 24 h, 36 h and 48 h of time exposure respectively, while the regression equations were Y = 2.931211 + 1.140429 X, Y = 1.810977 + 3.793674 X, Y = 4.526778 + 1.47082 X, Y = 4.925412 + 1.243574 X and Y = 5.634997 + .8632832 X; the χ² values along with their df were 21.71351(3), 4.249176 (3), 15.91378 (3), 12.9875 (3) and 2.314148 (3); the 95% confidence limits were 1.593366E-02 to 2665.474, 0.6029444 to 0.7960765, 0.0393291 to 1.118837, 7.634237E-03 to 1.726599 and 2.331059E-04 to 1.449859 mg cm⁻² respectively.

Bioassay with *Vitex negundo* (Leaf CHCl₃) Extract: All the *Vitex negundo* leaf (CHCl₃) was tested against *T. castaneum* adults. The data was then subjected to probit analysis, and the result has

been presented in **Table 5**. To trace acute toxicity an observation of lethality was made after 6 h of application of the doses.

TABLE 5: DOSE MORTALITY EFFECT OF THE CHCl₃ EXTRACTS OF AGAINST VITEX NEGUNDO (LEAF) T. CASTANEUM ADULTS

| Plant part | Exposure | LD ₅₀ value mgcm ⁻² | 95% confidence limit | | Regression equation | x ² value (df) |
|---|----------|--|----------------------|--------------|-------------------------------|---------------------------|
| | | | Upper | Lower | | |
| <i>V. negundo</i> (CHCl ₃) | 24 | 4463.364 | 6.859321E+13 | 2.904313E-07 | Y = 2.829286 + 0.4668542 X | 0.30506 (2) |
| | 36 | 131340.2 | 1.366983E+20 | 0.261914E-10 | Y = 2.979722 + 0.3301973 X | 0.45906 (3) |
| | 48 | 55332.94 | 9.081862E+13 | 3.371273E-05 | Y = 3.042121 + 0.3409167 X | 0.45132 (4) |

The LD₅₀ values for were 4463.364, 131340.2 and 55332.94 mg cm⁻² for 24, 36 and 48 h of exposure respectively, while the regression equations were Y = 2.829286 + .4668542 X, Y = 2.979722 + .3301973 X and Y = 3.042121 + .3409167 X, the χ² values along with their df were .3050642 (2), .459066 (3) and .4513229 (4); and the 95% confidence limits were 2.904313E-07 to 6.859321E+13, 1.261914E-10 to 1.366983E+20 and 3.371273E-05 to 9.081862E+13 mg cm⁻² respectively.

Bioassay with *Vitex negundo* (Root) Pet. E. extract: Pet. E. (Root) extracts of the *Vitex negundo* were tested against *T. castaneum*. The data was then subjected to probit analysis, and the

result has been presented in the **Table 6**. To trace acute toxicity an observation of lethality was made after 6 h of application of the doses.

TABLE 6: DOSE MORTALITY EFFECT OF THE CHCl₃ EXTRACTS OF AGAINST VITEX NEGUNDO (LEAF) T. CASTANEUM ADULTS

| Plant part | Exposure | LD ₅₀ value mgcm ⁻² | 95% confidence limit | | Regression equation | x ² value (df) |
|-----------------------------|----------|--|----------------------|-----------|-----------------------------|---------------------------|
| | | | Upper | Lower | | |
| <i>V. negundo</i> (root) | 24 | 1.185641 | 1.882845 | 0.7466071 | Y = 2.582428 + 2.251096X | 9.385418(3) |
| | 36 | 0.8622064 | 1.320118 | 0.5631313 | Y = 1.069454 + 2.030649X | 10.47551(4) |
| | 48 | 0.6729956 | 1.285587 | 0.3523083 | Y = 1.881685 + 1.705851X | 19.5863(4) |

The LD₅₀ values for were 1.185641, .8622064 and .6729956 mg cm⁻² for 24, 36 and 48 h of exposure respectively, while the regression equations were Y = 2.582428 + 2.251096 X, Y = 1.069454 + 2.030649 X and Y = 1.881685 + 1.705851 X, the χ² values along with their df were 9.385418(3), 10.47551(4) and 19.5863(4); and the 95% confidence limits were .7466071 to 1.882845, .5631313 to 1.320118 and .3523083 to 1.285587 mg cm⁻² respectively.

Bioassay with *Vitex negundo* (leaf Pet E) extract:

All the *Vitex negundo* MeOH extracts (leaf) were tested against the adult beetles of *T. castaneum* through residual film assay, and results found

promising. The data was then subjected to probit analysis, and the result has been presented in **Table 7**. To trace acute toxicity an observation of lethality was made after ½ h of application of the doses.

TABLE 7: DOSE MORTALITY EFFECT OF THE PET. E EXTRACTS OF *VITEX NEGUNDO* (LEAF) AGAINST *T. CASTANEUM* ADULTS

| Plant part | Exposure | LD ₅₀ value mgcm ⁻² | 95% confidence limit | | Regression equation | x ² value (df) |
|--|----------|--|----------------------|--------------|-----------------------------|------------------------------|
| | | | Upper | Lower | | |
| <i>V. negundo</i> (leaf CHCl ₃) | 12 | 1775.623 | 9.584444E-03 | 3.28953E+08 | Y =3.053877 + 0.4579813X | 2.109092(3) |
| | 24 | 4740.038 | 2.027758E+09 | 1.108018E-02 | Y =3.485439 + 0.3239162X | 3.044313(4) |
| | 36 | 150.1223 | 26254.13 | 0.8584059 | Y =3.498762 + 0.4726158X | 2.269408 (4) |
| | 48 | 7408.159 | 3.141738E+11 | 1.746836E-04 | Y =3.923931 + 0.2209718X | 1.311824 (4) |

The LD₅₀ values for were 1775.623, 4740.038, 150.1223 and 7408.159 mg cm⁻² for 12, 24, 36 and 48 h of exposure respectively, while the regression equations were Y = 3.053877 + .4579813 X, Y = 3.485439 + .3239162 X, Y = 3.498762 + .4726158 X and Y = 3.923931 + .2209718 X; the χ² values along with their df were 2.109092(3), 3.044313(4), 2.269408 (4) and 1.311824 (4); and the 95% confidence limits were 9.584444E-03 to 3.28953E+08, 1.108018E-02 to 2.027758E+09, 0.8584059 to 26254.13 and 1.746836E-04 to 3.141738E+11 mg cm⁻² respectively

Repellent Effect of the Test Plants on *T. castaneum* Adults:

All the extracts of the selected plants, *R. canescens* (whole plant), *D. heterocarpon* (whole plant) and *Vitex negundo* (leaf, st. b and root) were tested against *T. castaneum* adults for their repellent activity.

The extracts of *D. heterocarpon* (Pet. E), *Rauvolfia canescens* (Pet. E), *Vitex negundo* (leaf) Pet. *Vitex negundo* (root) Pet. E was found to show repellent activity against the adult's beetles of *T. castaneum* even for concentration from 0.0785- mg cm⁻² to as less as 0.0049- mg cm⁻² (0.0785-, 0.0393-, 0.0196-,

0.0098- and 0.0049- mg cm⁻² for ½ of 90 mm filter paper for all the plant extracts).

The data was read with a 1 h interval for up to 5 h of exposure and was subjected to ANOVA after transforming them into percentage value **Table 8**. While *D. heterocarpon* (Pet. E) and *Vitex negundo* (leaf) Pet. E is weakly active (P<0.05) and *R. canescens* (Pet. E) *Vitex negundo* (root) Pet. E is mildly active (P<0.01) and the rest were not active (*i.e.*) they didn't show repellent activity against the adult's beetles of *T. castaneum*. They had an attractant nature.

TABLE 8: ANOVA RESULTS OF REPELLENCY BY SOME PLANT EXTRACTS

| Types of extract | Source of variation (df) | | | F-ratio with level of significance | | P-value | |
|---|-----------------------------|-----------------------------|-------|---------------------------------------|-----------------------------|------------------|-----------------------------|
| | Between doses | Between Time interval | Error | Between Doses | Between time Interval | Between Doses | Between time interval |
| <i>D. heterocarpon</i> (Pet. E) | 4 | 4 | 16 | 16.17267* | 1.091619 | 1.77 E-05 | 0.3939 |
| <i>D. heterocarpon</i> (CHCl ₃) | 4 | 4 | 16 | 4.295318 | 3.262399 | 0.0150 | 0.0389 |
| <i>D. heterocarpon</i> (MeOH) | 4 | 4 | 16 | 2.062677 | 0.463112 | 0.1336 | 0.7618 |
| <i>R. canescens</i> (Pet. E) | 4 | 4 | 16 | 63.3918** | 1.964467 | 1.31E-09 | 0.1487 |
| <i>R. canescens</i> (CHCl ₃) | 4 | 4 | 16 | 2.453775 | 0.535133 | 0.0880 | 0.7119 |
| <i>R. canescens</i> (MeOH) | 4 | 4 | 16 | 2.32684 | 0.567867 | 0.1006 | 0.6896 |
| <i>Vitex negundo</i> (leaf) Pet. E | 4 | 4 | 16 | 10.69439* | 0.554647 | 0.0002 | 0.6986 |
| <i>Vitex negundo</i> (leaf) CHCl ₃ | 4 | 4 | 16 | 7.324571 | 1.231001 | 0.3369 | 3.0069 |
| <i>Vitex negundo</i> (root) Pet. E | 4 | 4 | 16 | 63.4694** | 3.035947 | 1.3E-09 | 0.0485 |
| <i>Vitex negundo</i> (St.B) CHCl ₃ | 4 | 4 | 16 | 7.13562 | 0.707396 | 0.0016 | 0.5985 |

*= (P <0.05), **= (P <0.01) and ***= (P <0.001)

DISCUSSIONS: The petroleum ether and methanol extracts of *R. canescens* (whole plant), and *D. heterocarpon* (whole plant); and the Pet. E. and CHCl_3 extracts of *Vitex negundo* leaf and the Pet. E. extract of the root has been found insecticidal against the stored product pest *T. castaneum*. For repellency *D. heterocarpon* (whole plant /Pet. Ether) extract and *V. negundo* (leaf /Pet. Ether) extract were weakly active ($P < 0.05$) and *R. canescens* (whole plant/Pet. Ether) extract and *Vitex negundo* (root /Pet. Ether) extract were mildly active ($P < 0.01$).

These findings receive supports from the prior works. An investigator found out the same result in *C. viscosa* against *T. castaneum* for repellency test¹⁶. *Desmodium gangeticum* and *Desmodium adscendens* have emerged as a good resource of traditional medicine¹⁷. *Desmodium gangeticum* can scavenge the free radicals generated during ischemia and ischemia-reperfusion thereby preserving the mitochondrial respiratory enzymes that eventually lead to cardioprotection and has potential prophylactic and therapeutic efficacy against Leishmania infection. *Desmodium adscendens* is helpful against chronic bronchitis and asthma.

A researcher reported that this plant is of interest as being an ingredient of the Dasamula Kvatha so often mentioned in Sanskrit works; it is considered to be febrifuge and anti-catarthal¹⁸. In the Dasamula it is placed among the five minor plants a decoction of these is directed to be used in catarrhal fever, cough, and other diseases. No earlier works were done on *R. canescens* so far internet browsing had been done thoroughly.

In conclusion, the present study was an attempt to screen two medicinal plants *R. canescens*, *D. heterocarpon*, and *Vitex negundo*. This experiment clearly showed the presence of insecticidal properties in the test plants have repellent, cytotoxic and antibacterial potential.

This may be due to the potential compounds was present an extract of the *R. canescens*, *D. heterocarpon* and *Vitex negundo*. These plants could be potential sources of new phytotoxic and insecticidal agents. As effective insecticidal activities were observed, more research should be

directed towards the isolation of insecticidal bioactive compounds as well as further field trials can be carried out to confirm the present findings.

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