Ovicidal inhibitory and insecticidal activities of plant extract *Anisomeles malabarica* against stored pest- *Callosobrunchus maculatus* (Fab) (Coleoptera: Chrysomelidae)

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Abstract:

India's population explosion has created a huge demand for vast amounts of food. Crop cultivation is just as vital as crop storage. Indian farmers' biggest concern, as the world's second-largest producer of agricultural products, is protecting their crops from grubs and adult pests. Pesticides used against them can cause a variety of acute toxicities, endocrine disorders, and even death as cancer-causing chemicals. Hence, in the present investigation the solvent extract of highly medicinal value of the plant *Anisomeles malabarica* was tested against azuki bean weevil *Callosobrunchus maculatus*. The extract was tested with different concentrations against *C. maculatus*. Result observed from the experiments found that ethanol extract of *A. malabarica* was more effective for insecticidal, repellent and ovicidal activities. The maximum ovicidal, insecticidal and repellency activity of 93.8%, 91.2% and 95.4% was noted in 96 hrs against *C. maculatus.* The study recommended using *A. malabarica* for more valuable effective control of *C. maculatus* as an alternative agent to artificial pesticides. The greatest option for them is to investigate and use resources derived from natural resources, such as biopesticides.

***Keywords*: *Callosobrunchus maculatus, Anisomeles malabarica,* Ethanol, ovicidal, insecticidal and repellent activity.**

1.0 Introduction

The pulse beetle, *Callosobruchus* sp., is a serious insect pest of economically important leguminous cereals. There are at least 20 species in the genus *Callosobruchus*, which developed primarily in Asia and Africa and originate primarily in tropical and subtropical regions of the world (Tuda *et al*., 2005). Cowpea (*Vigna unguiculata* (L.) Walp.) an associate with the Fabaceae family of crops. It0 also known as black-eyed peas or southern peas (Yusuf *et al*., 2011). It is high in protein, 1.0 percent fat, 56.6 percent carbohydrate, calcium 0.08 g, phosphorous 0.045 g, iron 5.7 mg, vitamin, thiamine 0.525 mg, riboflavin 300 mg, and fibre 2.2 gm and delivers 234 calories per 100 gm. (Srivastava and Ali, 2004). It has about three times the protein content of cereals. However, insect damage, such *as C. maculatus, Tribolium castaneum, Sitophilus oryzae, and S. zeamais,* causes significant post-harvest losses of legume grains. Due to the attack of the cowpea beetle, *C. maculatus*, huge losses in stored grains have been recognized, ranging from 20% to 50%, and occasionally even more up to100% (Udo and Harry 2013). *C. maculatus* causes an extreme deal of losses to stored cowpea seeds which has led to severe problems in farmworkers all over the world (Obembe and Ojo, 2018). Seed damage, weight loss, market value loss, and the possibility of a beetle infestation are all examples of quantitative and qualitative losses caused by the beetle (Iloba *et al*., 2016).

Anisomeles malabarica is also known as Malabar catmint erect shrub with 1.8m tall in the family Lamiaceae. It is originated in tropical and sub-tropical regions of south India and Sri Lanka. It is1.8m tall in softly white stems and branches. Flowers are purple in dense whorls of more or less broken up spikes; leaves are simple, opposite, thick, and aromatic. Antispasmodic, antipyretic, diaphoretic, anticancer, diuretic, and spermicidal effects have been described for the herb. Acrid, bitter, fragrant, aphrodisiac, stomachic, antihelminthic, febrifuge, and sudorific are some of the other properties of this plant (Prajapati and Kumar 2003).

In regions where current storage procedures have not been introduced, insect damage to stored grains and pulses could be as high as 40%. Currently, the use of gaseous and liquid insecticides to control pest infestation in grain and dry food items is heavily reliant, posing potential health risks to warm-blooded animals as well as the possibility of environmental pollution. Insecticidal plants provide an alternative to synthetic insecticides. Plant products have gained importance as a significant component of insect pest management due to their economic viability and environmental friendliness. They have the potential to minimize pesticide load in the environment as alternatives to chemical pesticides (Thiam and Ducommun, 1993). Plant-derived bio-insecticides, such as plant powder and oil, are being promoted among resource-poor farmers in developing countries since they are cheaper and more environmentally friendly than chemical insecticides (Okosun and Adedire, 2010).

The present experiment was aimed to determine the effect of powder and extract of *Anisomeles malabarica* insecticidal, repellent and ovicidal activity against cowpea weevils.

2.0Material and Methods

2.1 Collection of *Anisomeles malabarica*

The fresh and matured leaves of *Anisomeles malabarica* were collected at the Villupuram district and brought to the laboratory. The leaves were washed and shade dried at room temperature for about 10-15 days. The leaves were powdered using an electrical blender and sieved to obtain a fine powder. Then it is kept in a tightly closed glass container.

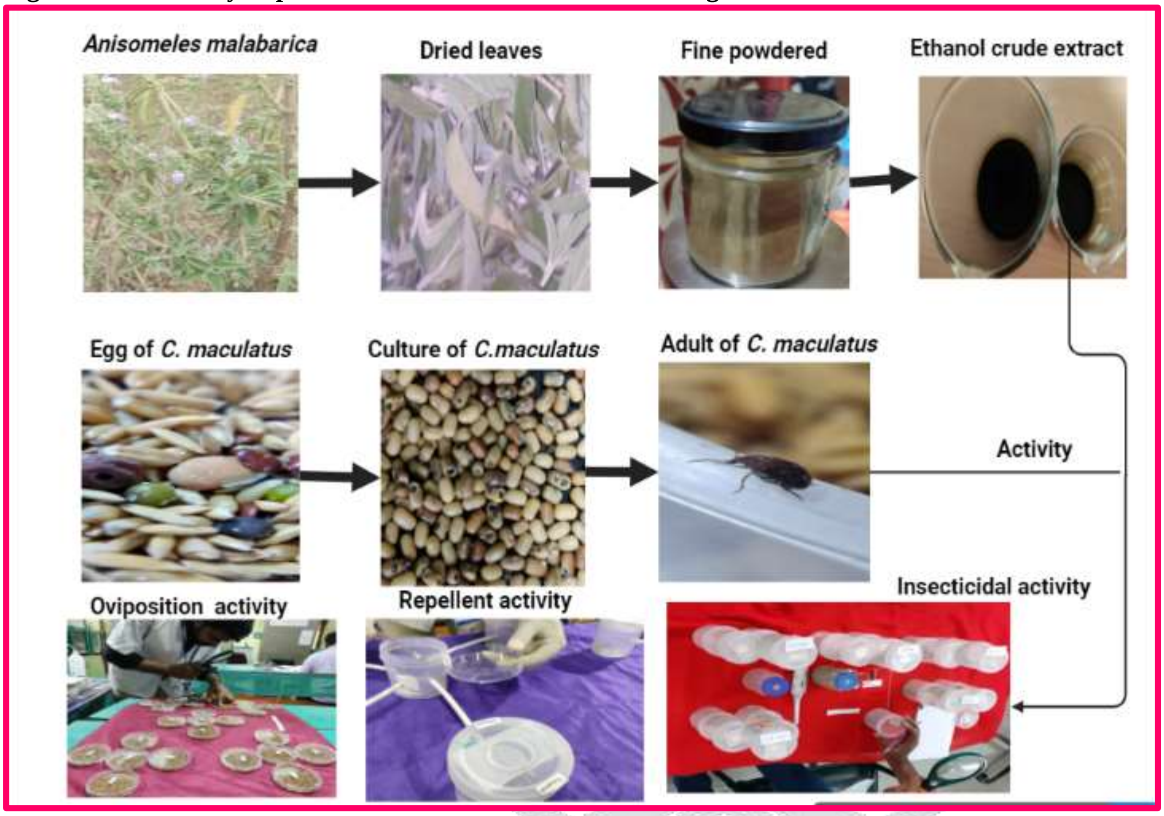
2.2 Preparation of extraction

The leaves powder was extracted with ethanol solvent by using the maceration method. The extracts were filtered through Whatman No.1 filter paper. Then the crude extract was dried using a rotatory evaporator at 50c.

2.3 Procurement of *Callosobruchus maculatus*

The insect was collected from Entomology Research unit, Department of Zoology in Government Art's college for Men (Autonomous) Nandanam Chennai.

Figure1: Bio efficacy of plant extract *Anisomeles malabarica* against *Callosobruchus maculatus* of stored grains



2.4 Ovicidal activity

Take 50g of clean and fresh cowpea seed separately in Petri plates. Then the solvent of different concentrations (100,200,300 and 400 ppm) of the crude extract in the beaker. After the crude extract and seed were mixed together by using a spatula to uniformly coating of the crude extract on the seeds. Then 20 insects were introduced in the Petri plates. Five replicates were maintained for each concentration of plant extract. Using the hand lens the number of dead eggs or unhatched eggs on the seeds were identified. In the same time untreated seeds was also set up to control treatment. Finally, calculate the percentage of ovicidal activity by using Su and Mulla (1998)

$$\text{Ovicidal activity (\%)} = \frac{Number\ of\ eggs\ hatched}{Total\ number\ of\ eggs\ treated} \times 100$$

2.5 Insecticidal activity

The insecticidal was calculated on the newly emerged adults of cow pea stored pest- *C. maculatus.* The Whatman no.1 filter paper was dipped with different concentrations (100,200,300 and 400 ppm) of plant extract was allowed dry it for15 minutes. Then the filter paper was involved to the cap of the lid internally and in all boxes, ten pairs of adults were introduced. Control was treated by Neem Azal. Mortality of the weevil when there were no antennae or leg movement even after disturbing with forceps. The insecticidal activity was determined by Abbott's formula (1925)

$$\text{\% Corrected mortality} = \frac{\%MT - \%MC}{100 - \%MC} \times 100$$

Where **MT**-mortality in treated group;

MC-mortality in control group.

2.6 Repellent activity:

The repellent activity was studied on adults of cowpea stored pests against *C. maculatus.* The experimental set up consist of five plastic boxes. All five boxes were connected with plastic tubes. In five boxes 4-boxes for treated box, 1-box for control box. After 10 pairs of unsexed adults were introduced. The different concentrations of extracts are 100ppm, 200ppm, 300ppm and 400ppm.After 96 hrs treated and the control boxes were recorded by using Lwanda *et al*., (1985).

$$EPI = \frac{Nt - Nc}{Nt + Nc} X\ 100$$

Where EPI = Effective Protection Idea;
Nt = Number of insects in the treated sample;
Nc = Number of insects in the control sample side.

3.0 Results

Table 3.1. Ovicidal activity of plant extract *Anisomeles malabarica* against *C. maculatus*

Concentrations	**Exposure periods in (Hrs)**		
	Ovicidal activity (%)		
	Ethanol extract		
	48 hrs	**72 hrs**	**96 hrs**
100ppm	15.8±1.20	20.1±2.73	24.0±3.45
200ppm	33.9±1.95	45.9±0.73	37.6±2.65
300ppm	49.4±2.85	64.2±2.30	79.1±0.89
400ppm	77.1±1.90	84.7±0.59	93.8±0.27
Neem Azal	100.0±0.00	100.0±0.00	100.0±0.00

Values expressed are mean mortality± standard deviations of five replications.

The ovicidal activity of ethanol extract of *A. malabarica* was tested against eggs of *C. maculatus* and the data were pertaining to the experiments as shown in table 3.1. It was observed that 15.8, 33.9, 49.4, and 77.1% ovicidal activity were noted in 100.200, 300, and 400ppm for 48 hrs. Likewise, % of ovicidal activity calculated for 72 hrs was 20.1, 45.9, 64.2, and 84.7 at the concentrations of 100, 200, 300, and 400ppm.Finally the highest activity of 24.0,37.6,79.1 and 93.8% was noted for 96 hrs at the concentration of 100,200,300and 400ppm respectively.

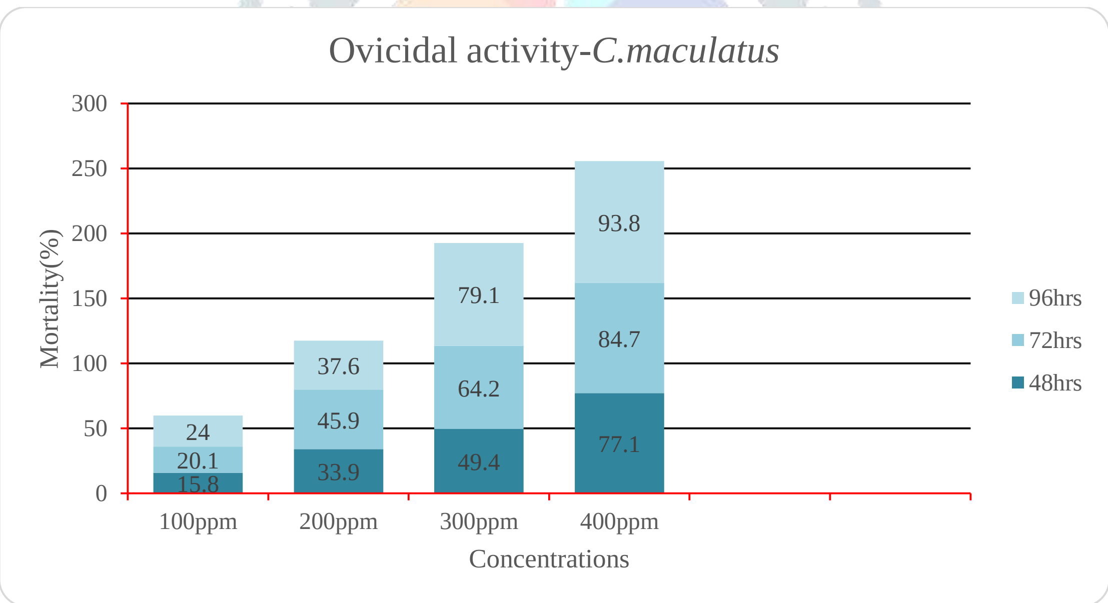


Figure 3.1. Ovicidal activity of plant extract *Anisomeles malabarica* against *C. maculatus*.

Table 3.2 Insecticidal activity of plant extract *Anisomeles malabarica* against *C. maculatus.*

Concentrations	**Exposure periods in (Hrs)**		
	Insecticidal activity (%)		
	Ethanol extract		
	48 hrs	**72 hrs**	**96 hrs**
100ppm	19.4±3.00	21.1±2.73	25.9±1.85
200ppm	36.6±1.05	49.0±1.96	33.1±2.72
300ppm	48.1±0.83	63.7±1.40	78.6±0.39
400ppm	74.3±1.62	87.2±1.19	91.2±1.07
Neem Azal	100.0±0.00	100.0±0.00	100.0±0.00

Values expressed are mean mortality± standard deviations of five replications

The insecticidal activity of ethanol extract of *A. malabarica* was tested against adult beetles of *C. maculatus* and the data were pertaining to the experiments as shown in table 3.2. It was observed that 19.4, 36.6, 48.1, and 74.3% insecticidal activity were noted in 100.200, 300, and 400ppm for 48 hrs. Likewise, % of insecticidal activity calculated for 72 hrs was 21.1, 49.0, 63.7, and 87.2 at the concentrations of 100, 200, 300, and 400ppm.Finally the highest activity of 25.9,33.1,78.6 and 91.2% was noted for 96 hrs at the concentration of 100,200,300and 400ppm respectively.

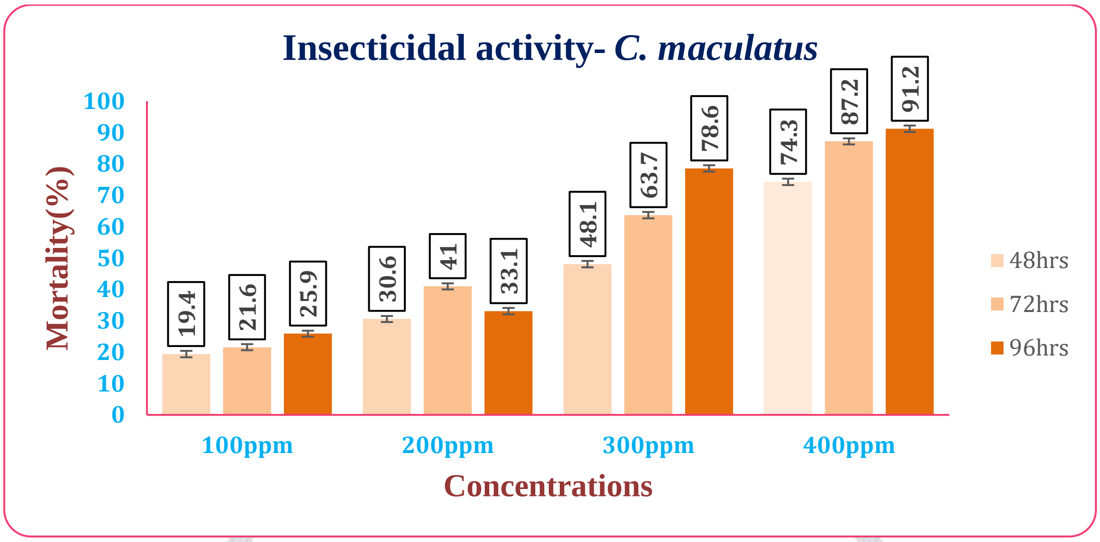


Figure3.2 Insecticidal activity of plant extract *Anisomeles malabarica* against *C. maculatus*.

Table 3.3 Repellent activity of plant extract *Anisomeles malabarica* against *C. maculatus*

Concentrations	Exposure periods in (Hrs) Repellent activity Ethanol extract		
	48 hrs	72 hrs	96 hrs
100ppm	18.1±4.52	23.8±3.39	27.2±2.59
200ppm	33.8±3.85	37.5±3.15	36.7±3.89
300ppm	42.9±1.96	68.2±2.86	74.6±1.72
400ppm	70.2±2.72	83.6±1.07	95.4±0.09
Tween 20	100.0±0.00	100.0±0.00	100.0±0.00

Values expressed are mean mortality± standard deviations of five replications

The repellent activity of ethanol extract of *A. malabarica* was tested against adult beetles of *C. maculatus* and the data were pertaining to the experiments as shown in table 3.3. It was observed that 18.1, 33.8, 42.9, and 70.2% repellent activity in 100.200, 300, and 400ppm for 48 hrs. Likewise, % of repellent activity calculated for 72 hrs was 23.8, 37.5, 68.2, and 83.6 at the concentrations of 100, 200, 300, and 400ppm.Finally the highest activity of 27.2,36.7,74.6 and 95.4% was noted for 96 hrs at the concentration of 100,200,300and 400ppm respectively.

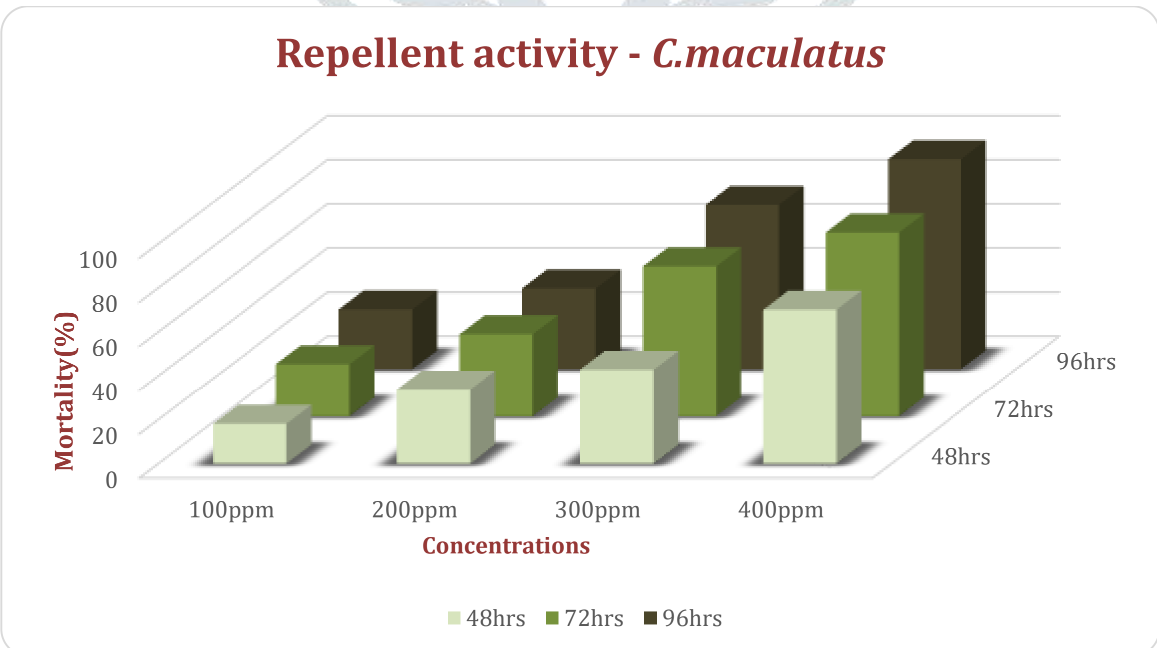


Figure 3.3 Repellent activity of plant extract *Anisomeles malabarica* against *C. maculatus.*

4.0 Discussion

Callosobrunchus maculatus is the most serious stored pests of cowpea seeds in India. In the present study the exposure of period in 92hrs of ethanol extract showed the highest (93.8%) ovicidal activity then the other two exposure periods. In the same way, a related trend was observed with insecticidal activity of *A. malabarica*. Likewise, the four different concentrations100, 200,300 and 400ppm of ethanol extract were found the maximum repellent mortality (95.4%) in 400ppm and this result agreed with result of earlier findings of several authors. This tendency was due to the presences of many phytocompounds absorbed with ethanol and thus the substantial activities were noted. Many research has reported on the effectiveness of plant extracts against the *C. maculatus*. Olusola Michael Obembe *et al*., 2020 reported that the experiments were conducted under laboratory conditions of 29 c temp and 75% relative humidity. Four different concentrations of 20g of pulses were administered to the crude extracts. The extracts from the stem bark of the shrub *K. africana,* on the other hand, were shown to have greater promise. Cowpea seeds oviposition and adult emergence were also inhibited by the extracts. As a result, natural pesticides are recommended for safeguarding cowpea seed during storage.

The insecticidal properties of three different plant powders against *C. maculatus* were investigated by Tagne Gabriel Fotso *et al*., (2019). It was discovered that at 10 gram of hexane extract, there were no *C. maculatus* F1 offspring emergences. Similarly, employing the other two extracts, acetone and methanol, totally blocked F1 production at the highest dosage during a seven-day therapy. Maiwada *et al*., (2017) used the same procedure to apply varying concentrations of two distinct plant powders (red pepper and garlic) to *C. maculatus*. In comparison to garlic, red pepper powder has a greater fatality rate. As a result, it was determined that bio pesticides can be administered to stored pests for up to eight weeks.

Similarly, chemical insecticidal are widely used to prevent and control pests' mortality, which may lead to dangerous consequences. Three distinct essential oils were examined by Viteri jumbo *et al*., (2018) and found to have insecticidal properties similar to the chemical pyrethyroid insecticide deltamethrin. At the same time that the oil dosage is increased, the growth ratio is reduced, and the bean weight is altered. Georgina *et al*., (2020) investigated the effect of five different plant products on the inhibitory effect of oviposition against *C. maculatus. S. aromaticum* had the highest eclosion failure rate, whereas *A. vasica* had the lowest. As a result, the maximum inhibitory oviposition and affecting larval growth of *C. maculutus* was determined. The highest mortality action of plant extract *Anisomeles malabarica* against both the beans weevil and the maize weevil was measured by Radha (2014) after 24 hours of treatment. Extracts of *Vitex negundo* were found to have a mild effect on mortality. *Murraya koenigii*, on the other hand, had an average mortality effect. Finally, it was discovered that the bean weevil had a higher fatality rate than the maize weevil.

Obembe and Ogugbite (2017) found that the plant powder was effective against *C. maculatus*. For a 168-hour treatment, *Z. zanthoxyloides* demonstrated the highest cytotoxic effect on adult *C. maculutus*. As a result, natural insecticides are more effective than chemical insecticides in controlling *C. maculutus*. Five different plant species were examined for their repellent efficacy against *C. maculutus* using the cup bioassay methodology and the filter paper method by Edwin and Anigboro Fidelis (2019). It was discovered that plant powder *C. millenii* had a higher repellent efficiency than *Z. officinale*. As a result, it was established that strong correlation coefficients and maximum significance were found when compared to the treatment of other species. The repellency and exposure period increased as the fraction of dosages and concentrations increased.

As a result, it demonstrated the effectiveness of ethanol crude extracts of *A. malabarica* leaf extract as a control agent for *C. maculatus* invading stored cowpea seeds, and it will serve as a viable alternative to chemical insecticides, which are costly and have side effects in the environment.

In present study the plant extract of *A. malabarica* were found effective to control *C. maculatus*. As a result, it demonstrated the effectiveness of ethanol crude extracts of *A. malabarica* leaf extract as a control agent for *C. maculatus* invading stored cowpea seeds, and it will serve as a viable alternative to chemical insecticides, which are costly and have side effects in the environment.

5.0 Conclusion

Now a days after post harvesting, the farmer and retail shop owners also facing the lot of damage loss due to stored grains weevils. In this study, *A. malabarica* was more powerful and effective for organic solvents ethanol. It gives high mortality rate and also more prominent result for the research work. So, A. *malabarica* is best natural bio pesticides when compared to chemical insecticides.

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