



Invitro screening of leaf extracts of selected plants from Lamiaceae, Asteraceae and Fabaceae for mortality and repellency to *Odoiporus longicollis* (Olivier)

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ABSTRACT: A study was focused on biocidal and repellent efficiencies of leaf extracts of *Chromolaena odorata* (L) (Asteraceae), *Gliricidia sepium* (Jacq.) (Fabaceae), *Coleus aromaticus* Benth, *Hyptis suaveolens* (L.)Poit. (Lamiaceae) and *Artemisia vulgaris* L. (Asteraceae), against *Odoiporus longicollis*, banana pseudostem weevil. Ethanol leaf extracts of the above plants were primarily analysed for the mortality and repellency activity, the results showed that among the extracts *G. sepium* was most effective. Further *G. sepium* in various solvents like petroleum ether, chloroform, ethyl acetate, ethanol and water were analysed for the biocidal and repellency assays, and found that, the ethyl acetate solvent of *G. sepium* elicited maximum effectiveness. The ethyl acetate extract of *G. sepium* is found as a promising biopesticide.

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KEY WORDS: *Gliricidia sepium*, ethyl acetate extract, biopesticide, banana stem weevil

Odoiporus longicollis (Olivier) (Coleoptera, Curculionidae), the banana pseudostem weevil (BPW) is a harmful pest which attacks banana plantations in South-East Asia and all the banana growing belts of India (Padmanaban and Sathiamoorthy, 2001). Due to the attack of this pest, the plant eventually becomes fragile resulting in premature falling due to tunnels made by the grubs in stems (Ravi and Palaniswami, 2002). Prasuna *et al.* (2008) reported that *O. longicollis* causes 10–90 per cent yield loss depending on the intensity of pest attack and management efficiency.

It is desirable to develop an appropriate biopesticide

to control *O. longicollis*. The present investigation evaluated the impact of leaf extracts of five selected plants having aromatic properties such as *Chromolaena odorata* (L) (Asteraceae), *Gliricidia sepium* (Jacq.) (Fabaceae), *Coleus aromaticus* Benth, *Hyptis suaveolens* (L.)Poit. (Lamiaceae) and *Artemisia vulgaris* L. (Asteraceae), for their biocidal and repellent efficiencies against *O. longicollis*. Having higher efficacy, Ethyl acetate extract of *G. sepium* leaves was chosen for detailed studies as well as to isolate and characterize the active component. Adults as well as the fourth instar larvae were evaluated for their repellency activities. Biocidal efficiency

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studies of *C. odorata*, *G. sepium*, *C. aromaticus*, *H. suaveolens* and *A. vulgaris* on *O. longicollis*, had not been conducted earlier and hence exploration in this direction was found to be worthwhile.

O. longicollis adults were collected from the banana fields of Aruvippuram, Thiruvananthapuram district, Kerala, India. The weevils were reared in plastic containers (30cm × 20cm) along with pseudostem pieces of size 10cm × 10cm. Pseudostem accommodating eggs were reared in separate containers and the newly hatched first instar larvae were transferred onto different containers. Fresh pseudostem pieces weighing 1, 2, 3 and 4g were provided respectively for the first, second, third and fourth instars and these pseudostem pieces were regularly replaced on the third day. Mortality and repellency assays were performed using eight insects each in control and treated cultures. Each experiment was performed in eight replicates.

Leaves of *C. odorata*, *G. sepium*, *C. aromaticus*, *H. suaveolens* and *A. vulgaris* were collected from Aruvippuram, Thiruvananthapuram district, Kerala, India (8.4245° N; 77.0982° E). The plants

Table 1. LC₅₀ (mg ml⁻¹) values of selected plant extracts. (LCL: 95% lower confidence limit, UCL: 95% upper confidence limit)

No	Plant	LC ₅₀	LCL	UCL
1	<i>Artemisia vulgaris</i>	519.874	438.164	726.493
2	<i>Coleus aromaticus</i>	407.249	348.283	532.345
3	<i>Gliricidia sepium</i>	149.705	110.403	178.565
4	<i>Chromolaena odorata</i>	382.238	346.464	438.172
5	<i>Hyptis suaveolens</i>	286.273	250.637	329.365

were identified, leaves were washed thoroughly with fresh water and shade dried for 14 days. Soxhlet extraction of these leaves were done using ethanol as solvent. Serial extraction of the best effective plant leaves were done in various solvents like petroleum ether, chloroform, ethyl acetate, ethanol and water were analysed for the mortality and repellency assay to select the best solvent.

Fourth instar larvae of *O. longicollis* were selected for the experiment. Each larva was provided with pseudostem piece of weight 4g as food. The different plant extracts in ethanol solvent with varying doses of 100, 200, 300 and 400 mg ml⁻¹ were separately spread on the pseudostem pieces. Insects devoid of any plant extract treatment were used as the control group. During the experiment, the pseudostem pieces were replaced every third day. Mortality of the larvae was noted after 12, 24, 48 and 72 h respectively. The *G. sepium* leaf extracts in various solvents with varying doses of 100, 200, 300 and 400 mg ml⁻¹ were also analysed for the mortality assay.

Repellency assay was conducted using Whatman No1 filter paper with a diameter of 150mm. The filter papers were cut into two halves, followed by the uniform application of 1 mg ml⁻¹ of the ethanol plant leaf extract of selected plants on one half. The other half was treated with ethanol alone and both halves were air-dried to evaporate the solvent. After complete evaporation, both halves were re-made into the original disc shape by using cello tape. The discs were then placed in petri dishes followed by the release of larvae at the centre.

Results obtained from mortality and repellency were corrected using Abbott's correction formula (Abbott, 1925). Statistical analysis of the results was performed by the statistical software SPSS V.23 for windows using Tukey's test and one way ANOVA with Duncan's post hoc pairwise comparisons.

Probit analysis of the leaves showed LC₅₀ values of 519.874, 407.249, 149.705, 382.238 and 286.273 mg ml⁻¹ respectively for the plants *A. vulgaris*, *C. aromaticus*, *G. sepium*, *C. odorata* and

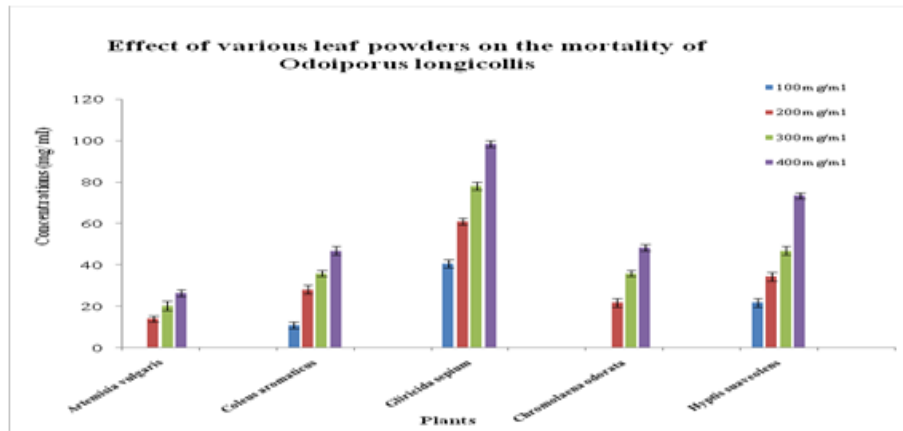


Fig. 1 Effect of various leaf powders on the mortality of 4th instar larvae of *O. longicollis*

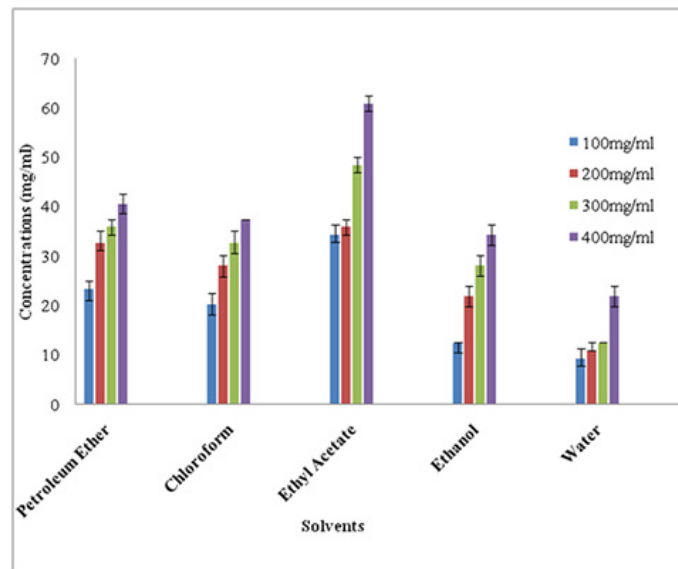


Fig. 2 Effect of various solvent leaf extracts of *G. sepium* on the mortality of fourth instar larvae of *O. longicollis*

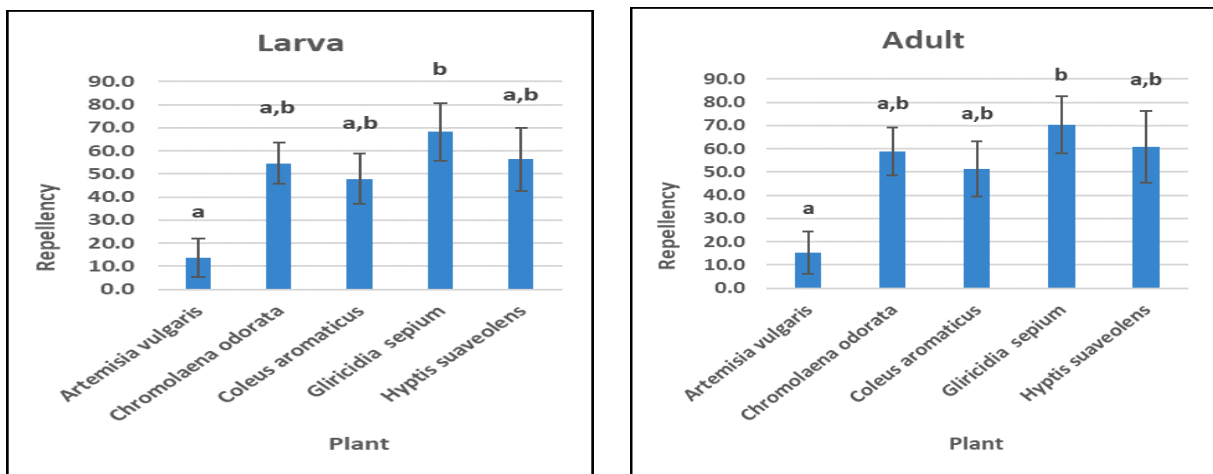


Fig. 3 Repellency activity of plant leaf extracts on the larvae and adults of *O. longicollis* (Different superscript letters in the graph indicate statistical significance at 5% level)

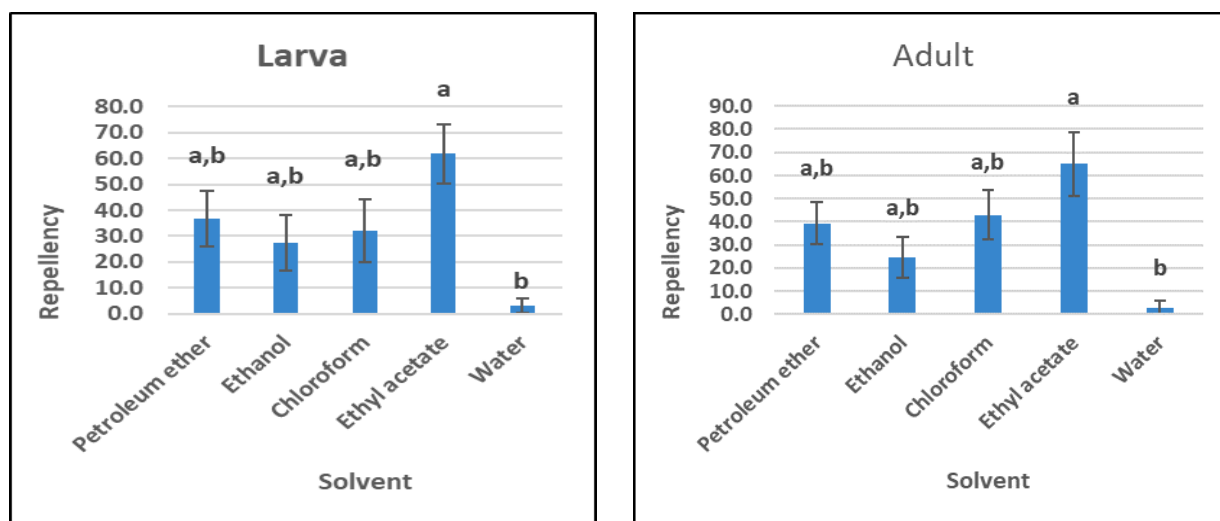


Fig. 4 Repellency activity of different solvent leaf extracts of *G. sepium* on *O. longicollis* (Different superscript letters in the graph indicate statistical significance at 5% level)

H. suaveolens (Table 1). Highest percentage of mortality with *G. sepium* leaf extract followed by *H. suaveolens*, *C. odorata*, *C. aromaticus* and *A. vulgaris*.

In the experiment on the effect of leaf powders on the mortality of 4th instar larvae of *O. longicollis*. *G. sepium* was found to elicit the maximum impact followed by *H. suaveolens*, *C. odorata*, *C. aromaticus* and *A. vulgaris* (Fig. 1).

In the leaf extract experiment maximum mortality was noted in the ethyl acetate solvent leaf extract of *G. sepium* followed by petroleum ether, chloroform, ethanol and water respectively (Fig. 2). Probit analysis of the different solvents revealed LC₅₀ of solvent petroleum ether as 808.013 mg ml⁻¹, for chloroform 863.338, ethyl acetate 292.661, ethanol 982.994.

Among the different extracts, the ethyl acetate solvent extract exhibited maximum repellency followed by petroleum ether, chloroform and ethanol respectively. *G. sepium* exhibited maximum repellency, followed by *H. suaveolens*, *C. odorata*, *C. aromaticus* and *A. vulgaris*. Repellency to larva is statistically significant against the selected plants ($F_{4,15} = 3.483$, p value = 0.033) and in adult is also

significant against the selected plants as determined by one-way ANOVA ($F_{4,15} = 3.138$, p value = 0.046). From Tukey's test, plant repellency showed maximum significance between *Artemisia vulgaris* and *G. sepium* (Fig. 3)

Solvent repellency in larva and adult was significant ($F_{4,15} = 4.294$, p value = 0.016 for larva, $F_{4,15} = 5.586$, p value = 0.006 for adult). The Tukey post hoc multiple comparison test shows that the solvent repellency has maximum significance between ethyl acetate and water (Fig. 4).

A. vulgaris, *C. odorata*, *G. sepium*, *C. aromaticus* and *H. suaveolens* elicited biocidal and repellency effects on *O. longicollis*. However, the maximum impact was displayed by the ethanol extract of *G. sepium* that the primary and secondary metabolites eluted out maximum in ethanol solvents.

The management of banana weevil is done primarily by the application of insecticides (Collins *et al.*, 1991). Biological agents are an important alternative to minimize or replace the use of synthetic pesticides (David, 2008). *C. infortunatum*, *L. camara* and *C. alata* elicited biocidal activity in pseudostem weevil, of which *C. infortunatum* was found to be the most effective (Remya and Dayanandan, 2019).

Stem injection of monocrotophos, Azadirachtin and the application of *Beuveria bassiana* recorded the highest percent mortality (Irulandi *et al.*, 2012). Essential oils from stems of *T. purpurea* and *I. carnea* could be explored as natural repellents for the control of *O. longicollis* (Sahayaraj *et al.*, 2015).

The presence of phytochemicals in the ethanol extract of *G. sepium* elicited antibacterial activity (Ajaicoba, 2002). The potency of *G. sepium* was due to the presence of saponins, phenolic compounds, essential oils, and flavonoids (Akharay *et al.*, 2012). The phytochemical analysis of *C. odorata* showed the presence of tannins, saponins, flavanoids and alkaloids in the leaves of *C. odorata* which indicates larvicidal efficacy (Man, 2013). *G. sepium* leaf and flowers contains forty-two known compounds. Of these sixteen have been identified and quantified from the flower essential oil by GC-MS analysis (Kaniampady *et al.*, 2007). The essential oil from the leaves of *G. sepium* from Columbia was analysed by GC-FID and GC-MS (Celis *et al.*, 2015). The major components are methyl-3(E)-pentenyl ether (11.55%), 3-methyl-2-butanol (10.65%), 3-methoxy hexane (10.14%), 1-(1-ethoxyethoxy)-2-hexene (9.72%), 2- decanol (8.97%), coumarin (8.07%) and hexadecanoic acid (5.16%) (Reddy *et al.*, 2010).

Based on previous reports, natural agents which have the potential to elicit repellency and mortality in both the larvae and adult populations of weevils could be effectively developed as biopesticides. The present findings demonstrated significant repellency and mortality in both the larvae and adult populations of *O. longicollis*.

Ethyl acetate extract of *G. sepium* is identified as a potent pesticide against *O. longicollis* is a preliminary indication.

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