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Larvicidal activity and GC–MS analysis of *Ophiorrhiza mungos* against *Spodoptera litura*

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ABSTRACT

The highly polyphagous insect *Spodoptera litura* has become a serious pest in recent years, infesting a variety of crops due to the pest's extensive presence; pesticides are constantly being used to try to control the infestation. Several botanical/ plant extracts have been employed to regulator the *S. liure*. The current study was aimed to assess the chemical constituents from the methanolic extract of *O. mungos* and exploring the larvicidal activity of *O. mungos* against *S. litura*. Our observations indicated that the total phenolic content of *O. mungos* was found to be 16.22±0.115 mg GAE/g in the stems and 21.17±0.011 mg GAE/g in the leaves. The total flavonoid content was found to be 42.80±0.096 mg QE/g in the stems, and 28.52±0.120 mg QE/g in the leave of *Ophiorrhiza mungos*. Following a 24-hour exposure period, larval mortality was observed. A concentration of 5 mg/mL was shown to have the greatest larval death rate. The GC-MS analysis of O. mungos leave revealed seven phytocompounds, whereas the stem sample analysis revealed ten phytocompounds, respectively. These results conclude that methanolic crude extracts *O. mungos* can be utilized as an effective pesticidal agent for managing lepidopteran pests..

Keywords: *Ophiorrhiza mungos, Spodoptera litura*, Bio-pesticides, GC-MS
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INTRODUCTION

Insect pests are critical agents in wreaking agricultural crops alongside other natural causes. Among the lepidopteran family, *Spodoptera litura* Fab. (Lepidoptera: Noctuidae) highly destructive and detrimental pest, inflicting damage to more than 150 plant species within South Asian countries (Huang *et al.,* 2014; Selin *et al.,* 2016;). A strong flier *S. litura* is recorded to cover extensive distances during summer season. It is known as a foremost agricultural pest throughout tropical nations such as China, Japan, and India imposing considerable economic losses across diverse vegetable and grain crops (Wheeler and Isman, 2001; Selin *et al.,* 2016; Saraswathi *et al.,* 2023).

Chief agent managing insect pests, the chemical pesticides are widely accepted but also have shown resistance by the pests (Vasantha-Srinivasan *et al.,* 2017). Resistance to widely used commercial insecticides has been linked to altered metabolism in insects notably in the up regulation of detoxifying enzymes or structural modifications of enzymes (Vasantha-Srinivasan *et al.,* 2016; Edwin *et al.,* 2016; Annamalai *et al.,* 2017; Dinesh-Kumar *et al.,* 2018; Shin *et al.,* 2018). Majorly with the problem of pest resistance in addition to environmental degradation, negative impact on biodiversity and human illness has made us to revert to the use of botanical insecticides which were widely used from millenia.

"Plant extracts" the natural repellants, antifeedants and growth inhibitors encompasses a wide variety of phytochemicals, many of which have proven effective in controlling insect pests (Chellappandian *et al.,* 2019). These specialized secondary metabolites serve as a potent defense mechanism in plants, safeguarding them against herbivorous predators, microbial pathogens, and other ecological threats (Senthil-Nathan 2013). Contemporary research in insect toxicology predominantly suggests the usage of green extracts and their principal derivatives for the control of detrimental pests with agricultural and medical relevance. Essential oils and bioactive compounds are playing a crucial role in integrated pest management (IPM) strategies, including the control of mosquitoes and lepidopteran pests (Benelli et al 2016; Karthi *et al.,* 2020; Ponsakar *et al.,* 2020).

Global research suggests that plant extracts especially rich in bioactive compounds not only control the spread of agricultural pests but also the disease vectoring arthropods, exhibiting comparable toxicity to its chemical counter (Senthil-Nathan *et al.,* 2008; Thanigaival *et al.,* 2012; Lija-Escaline *et al.,* 2015). Several authors suggested that natural products from the plants display convincing toxicity against pests, acting as insect growth regulators, larvicidal agents, insect repellents, adulticides, and oviposition attractants (Senthil-Nathan 2015; Selin *et al.,* 2016; Senthil-Nathan, 2020; Amala *et al.,* 2020). Plant derivatives may deliver as alternative sources of commercial pesticides, since they are enriched with bioactive insecticidal compounds that are easily degradable (Senthil-Nathan *et al.,* 2005; Murfadunnisa *et al.,* 2019).

Ophiorrhiza mungos a species from the Rubiaceae family, the genus *Ophiorrhiza* is extensively distributed throughout the wet forests of tropical and subtropical Asia, Australia, New Guinea, and the Pacific Islands (Gopalakrishnan *et al.,* 2018). It is one of the Indo-Malaysian genera (Hareesh *et al.,* 2018). Currently the genus recorded 321 species, five variations, and one subspecies in the *Ophiorrhiza* L. genus. Specifically, the Western Ghats and northeastern states of India are home to 46 species and 5 variants, whereas the Indian state of Kerala is home to 16 species and 3 varieties (Sibi *et al.,* 2012).

The *Ophiorrhiza mungos* is known for its bioactive compounds like alkaloids (indole alkaloids and quinoline alkaloids), secoiridoid monoterpenes, sesquiterpenes, steroids, quinones and phenylpropanoids (Krishnakumar *et al.,* 2020). These identified compounds contribute in antimicrobial, antiviral, anti-ulcer, anti-helminthic, anti-venomous and also gastroprotective properties (Taher *et al.,* 2020; Patowary and Sharma, 2023). In the field of pest management utilizing *O. mungos* extracts as a botanical insecticide offers a sustainable and eco-friendly approach to management of *Spodoptera litura* infestation in host plants. Currently no data regarding the larvicidal activity of O.mungos against S.litura are available. Hence, the current study was aimed to assess the chemical constituents from the methanolic extract of *O. mungos* and exploring the larvicidal activity of *O. mungos* against *S. litura*.

MATERIALS AND METHODS

Collection of *Ophiorrhiza mungo*

Disease-free, healthy parts of *O. mungos* (leaves and stems) were collected from Pullurampara village (Topography: Hilly terrain with gentle slopes and valleys; Climate: Tropical monsoon climate with high humidity and moderate temperature; Soil Type: Laterite soil with high iron and aluminum content, Vegetation: Tropical evergreen forest with diverse plant species) in Kozhikode district, Kerala, India. The samples were washed in deionized water, shade-dried, powdered and stored in airtight bottles until use.

Plant Extract Preparation

About 20gms of both stem and leaves plant powder was processed for extract preparation in Soxhlet extractor using methanol as solvent. Upon completion of the extraction process, the organic solvents were concentrated under reduced pressure at a temperature of 40-50°C (Arora *et al.,* 2017).

Phytochemical Aanalyses

Qualitative tests for analysis and identifying of phytochemicals of alkaloids, carbohydrates, flavonoids, glycosides, proteins, tannins, and free amino acids were executed for *Ophiorrhiza mungos* extracts following a modified procedure as described by Harborne and Sazada et al (2009).

TPC

The total phenolic content (TPC) of *Ophiorrhiza mungos* was estimated using the Folin-Ciocalteu reagent, based on the method of Sánchez-Rangel *et al.* (2013) with slight modifications. Standard gallic acid solution of concentrations ranging from 20-100 µg/mL was mixed with 7% Na_2CO_3 (w/v) and incubated at room temperature for 90 minutes. The optical density at 550 nm was measured using a UV-Vis spectrophotometer. For TPC estimation, 1 ml of crude extracts of *O. mungos* was used, and a reagent blank was prepared with distilled water, undergoing the same treatment (Singleton & Rossi, 1965). All experiments were conducted in triplicates. The TPC was calculated as gallic acid equivalent (mg/g) using the standard calibration curve equation and the results were expressed as mean ± SD values.

TFC

The total flavonoid content (TFC) of *Ophiorrhiza mungos* leaves and stems was determined using the standard aluminum chloride method (Zhishen *et al.,* 1999). About 1 mL of crude methanolic extracts from *O. mungos* leaves and stems was used. A series of standard Quercetin solutions at concentrations of 20-100 µg/mL was prepared in distilled water. To this mixture, 0.3 mL of 5% sodium nitrite was added and incubated at room temperature (RT) for 5 minutes, followed by the addition of 0.3 mL of 10% aluminum chloride. The resulting reaction mixture was then treated with 1M sodium hydroxide (2 mL), and the optical density (OD) was determined at 510 nm. TFC was expressed as Quercetin equivalent (mg/g) using the standard calibration curve (Aiyegoro & Okoh, 2010)

***Spodoptera litura* larvae**

S. litura (Accession no: NBAII-MP-NOC-02) disease-free third instar larvae were collected from the National Bureau of Agricultural Insect Resources (NBAIR), Hebbal, Bengaluru, Karnataka. The larvae were reared on castor leaves as a natural diet, and the rearing conditions were maintained according to Mamtha *et al.* (2019).

Larvicidal Activity

The larvicidal activity of *O. mungos* extracts was evaluated using the leaf dip method (Park *et al.,* 2002). Castor leaves discs of 6.0 cm in diameter were used to assess the larvicidal activity of the extracts. The castor leaves were cleaned with 70% double-distilled alcohol and allowed to air dry for fifteen minutes before being dipped into the plant extracts. Four different concentrations of *O. mungos* extracts (0.5, 1, 2, and 5 mg/L) were utilized. Leaf discs were immersed in each concentration for 30 seconds and allowed to evaporate for one hour. Distilled water-treated castor leaves were used as negative control and carbofuran (1%) as positive control. For each concentration 20 larvae were used per treatment. The mortality rate was observed and recorded every 24 hrs, compared with the positive control. All moribund larvae were considered dead.

Characterization of Plant Volatiles

The crude extract of *Ophiorrhiza mungos* (1 µL) was dissolved in HPLC grade methanol and subjected to analysis using a Clarus 680 Gas Chromatograph (PerkinElmer) an equipment with a fused silica column packed with Elite-5MS (5% biphenyl 95% dimethylpolysiloxane, 30 m × 0.25 mm ID × 250 µm df), and helium as the carrier gas at a constant flow rate of 1 mL/min. With the injector temperature set at 260°C, the oven temperature program was as follows: 60°C for 2 minutes, then increased to 300°C at a rate of 10°C per minute, and held at 300°C for 6 minutes. The mass detector conditions were: transfer line temperature at 240°C, ion source temperature at 240°C, and ionization by electron impact at 70 eV, with a scan time of 0.2 seconds and a scan interval of 0.1 seconds, analyzing fragments from 40 to 600 Da. The molecular structure, molecular weight, and formula of the individual compounds in the extracts were determined by interpreting the mass spectra from GC–MS using the database of the

National Institute of Standards and Technology (NIST) (Amala *et al.,* 2021).

Statistical analysis

The larval mortality data is shown as a percentage. Values are shown as mean ± SD (standard deviation) in the tables and graphs. Graph Pad Prism 10 and Microsoft Excel were used to create the graphs. Statistically significant when $P < 0.05$. *P <0.0001 in comparison to the positive control treatment group (one-way ANOVA with Holm-Sidak post-hoc test). The average of n = 3 separate experiments is represented by all the data. ****; P <0.0001; ***, P <0.001; **, P < 0.01; ns: not significant.

RESULTS

Phytochemical Screening

Qualitative analysis of the methanolic crude extracts of *O. mungos* revealed a range of compounds such as phenolics, saponins, terpenes, steroids, carbohydrates, and alkaloids. The presence of these compounds contributes to numerous biological activities such as free radical scavenging, antimicrobial, insecticidal, larvacidal and anticancer properties. The methanolic extracts from the leaves of *O. mungos* reveal a higher concentration of phytocompounds compared to extracts from their stems (Table 1).

Table 1. Phytocompounds identified from the methanolic extract of O.mungos through GC-MS analysis.

Phytochemical	***O. mungos* stem**	***O. mungos* leaves**
Carbohydrates	+	+
Proteins	+	+
Phenolics	-	+
Flavonoids	+	+
Steroids	+	+
Sterols & Triterpenoids	-	+
Tannin	+	+
Terpenoid	+	+
Glycosides	+	+
Saponins	+	+

TPC

The phenolic content in *Ophiorrhiza mungos* was measured as gallic acid equivalent (GAE) using the Folin-Ciocalteu reagent. The results were derived from a calibration curve ($y = 0.0085x$, $R^2 = 0.9758$, where x represents absorbance and y indicates gallic acid equivalent in mg/g). The plant extracts are rich in phenolic compounds, which contain hydroxyl groups essential for their redox properties, thereby producing antioxidant activity through free radical scavenging. For *Ophiorrhiza mungos*, the total phenolic content was found to be: 21.17±0.011 mg GAE/g in the leaves and 16.22±0.115 mg GAE/g in the stems respectively (Figure 1). The high levels of phenolic content recorded in *O. mungos* leaves also indicate a strong potential for antioxidant activity.

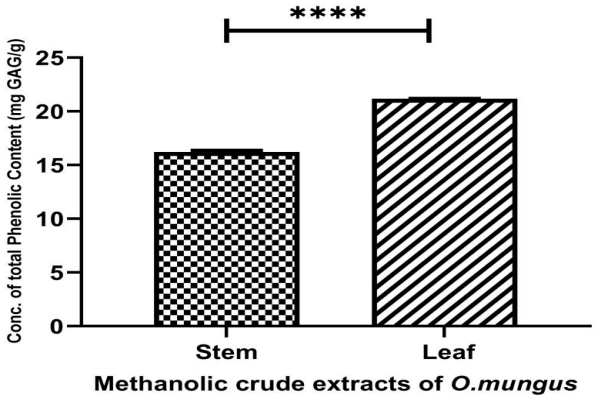

Figure 1. Concentration of total phenolic contents in Ophiorrhiza mungos leave and stem. Values are represented in Mean ± SEM from three independent experiments. *P<0.0001 (one-way ANOVA followed by Holm-Sidak post-hoc test) compared to the Positive Control treated group. All the data represents an average of n=3 independent experiments. Here, ****, p<0.0001(highly significant).

TFC

The total flavonoid content in the methanolic extracts of *Ophiorrhiza mungos* was measured in terms of quercetin equivalent (QE) using a colorimetric assay with aluminum chloride. The flavonoid content in the extracts was quantified based on absorbance readings, plotted against a calibration curve ($y = 0.0066x$, $R^2 = 0.9939$, where x represents absorbance and y denotes quercetin equivalent in mg/g).

Table 2. Phytocompounds identified from the methanolic extract of *O. mungos* leaves through GC-MS analysis

Sl.No	Retention Time	Height	Area	Area (%)	Norm (%)	Compound Name	Compound formula	Properties
1	19.611	53,262,640	3,290,682.2	1.628	2.03	Z-1,9-Dodecadiene	$C_{12}H_{22}$	Antioxidant (Ghosh *et al.*,2013)
2	20.176	470,152,608	161,732,944.0	80.033	100.0	Bicyclo[3.1.1]Heptane, 2,6,6-Trimethyl-,[1R-(1.Alpha.,2.Alpha.,5.Alpha.)]	$C_{10}H_{18}$	Antioxidant (Momoh *et al.,* 2023)
3	20.901	89,553,672	17,377,020.0	8.599	10.74	Z-1,8-Dodecadiene	$C_{12}H_{22}$	Antioxidant (Ghosh *et al.*,2013)
4	21.216	38,074,600	6,961,357.0	3.445	4.30	1,6-Octadiene, 3,7-Dimethyl-,(S)	$C_{10}H_{18}$	Larvicidal (Nagella *et al.,* 2012)
5	25.143	62,168,280	4,596,650.5	2.275	2.84	6-Octen-1-Ol, 3,7-Dimethyl-,(R)	$C_{10}H_{20}O$	Anticancer (Swantara *et al.,* 2022)
6	27.104	26,262,992	3,889,542.2	1.925	2.40	2-Methyl-6-Methylene-Octa-1,7-Dien-3-OL	$C_{10}H_{16}O$	Larvicidal (Pandeeswari *et al.,* 2022)
7	27.494	37,359,448	4,234,398.5	2.095	2.62	2-Methyl-6-Methylene-Octa-1,7-Dien-3-OL	$C_{10}H_{16}O$	Larvicidal (Pandeeswari *et al.,* 2022)

Table 3. Phytocompounds identified from the methanolic extract of *O.mungos* stem through GC-MS analysis**.**

Sl.no	Retention Time	Height	Area	Area (%)	Norm (%)	Compound Name	Compound Formula	Properties
1	17.905	933,543,168	49,051,028.0	27.198	70.76	1,3-Dioxolane, 4,4,5,5-Tetramethyl	$C_7H_{14}O_2$	N/A
2	19.535	1,665,284,864	69,318,920.0	38.436	100.0	3-Hexyne, 2-Methyl	C_7H_{12}	Antioxidant (Sunday *et al.,* 2021)
3	19.791	112,659,920	4,413,941.0	2.447	6.37	Meso-2,5-Dimethyl-3,4-Hexanediol	$C_8H_{18}O_2$	Antioxidant (Khan *et al.,*2016)
4	20.211	122,794,176	44,651,108.0	24.758	64.41	Z-1,9-Dodecadiene	$C_{12}H_{22}$	Antioxidant (Ghosh *et al.,*2013)
5	23.557	49,319,920	1,504,665.6	0.834	2.17	3-Methylpent-2-ene-1,5-Diol	$C_6H_{12}O_2$	Antioxidant (Chandy *et al.,* 2022)
6	24.277	59,958,888	2,078,578.8	1.153	3.00	3-Methylpent-2-ene-1,5-Diol	$C_6H_{12}O_2$	Antioxidant (Chandy *et al.,* 2022)
7	24.988	67,225,512	2,441,762.2	1.354	6.52	4-Nonene,2,3,3-Trimethyl-,(E)-	$C_{12}H_{24}$	Antimalarial (Singh *et al.,* 2023)
8	25.663	57,003,032	1,748,286.2	0.969	2.52	Oxirane, 2,2'-(1,4-Butanediyl)Bis-	$C_8H_{14}O_2$	Antibacterial (Imsong *et al.,* 2022)
9	26.328	51,457,524	2,151,136.5	1.193	3.10	2-Methyl-6-Methylene-Octa-1,7-Dien-3-OL	$C_{10}H_{16}O$	Larvicidal (Pandeeswari *et al.,* 2022)
10	29.915	40,914,044	2,990,272.2	1.658	4.31	2-Nitro-2-Ethyl-1,3-PropaneDiOl	$C_5H_{11}O_4N$	Antioxidant (Imsong *et al.,* 2022)

For the methanolic extracts of *Ophiorrhiza mungos*, the total flavonoid content was found to be: 28.52±0.120 mg QE/g in the leaf and 42.80±0.096 mg QE/g in the stems respectively. Flavonoids, known for their antioxidant properties, are secondary metabolites whose effectiveness is influenced by the number and arrangement of free hydroxyl (OH) groups (Figure 2).

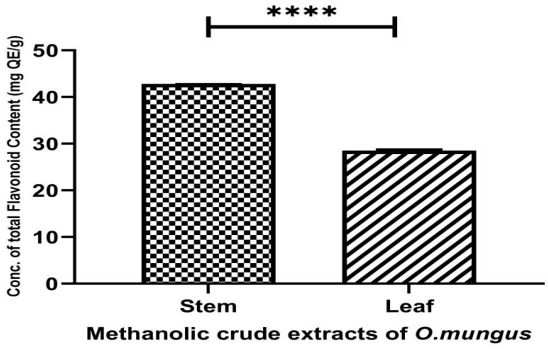

Figure 2. Concentration of Total Flavanoid contents in Ophiorrhiza mungos leave and stem. Values are represented in Mean ± SD from three independent experiments. *P<0.0001 (one-way ANOVA followed by Holm-Sidak post-hoc test) compared to the Positive Control treated group. All the data represents an average of n=3 independent experiments. Here, ****, p<0.0001(highly significant).

Identification of Compounds

The *Ophiorrhiza mungos* chemical constituents identified by the Gas chromatography/Mass spectrometry (GC-MS) obtained results were included in the Tables 2-3 with their molecular formula, retention time, and peak area (Figures 3-4). The results of *O. mungos* revealed the presence of seven phytochemical compounds in the leaves and ten compounds in the stem from the methanolic crude extracts. The major compound isolated in the leave extract is Bicyclo [3.1.1] Heptane, 2,6,6-Trimethyl-, [1R-(1.Alpha.,2.Alpha.,5.Alpha.)], and the minor compound Z-1,8-Dodecadiene, both the compounds indicating the antioxidant activities. Similarly, the major compound identified in the stem extract is 3-Hexyne, 2-Methyl, while the minor compound was 1,3-Dioxolane, 4,4,5,5-Tetramethyl.

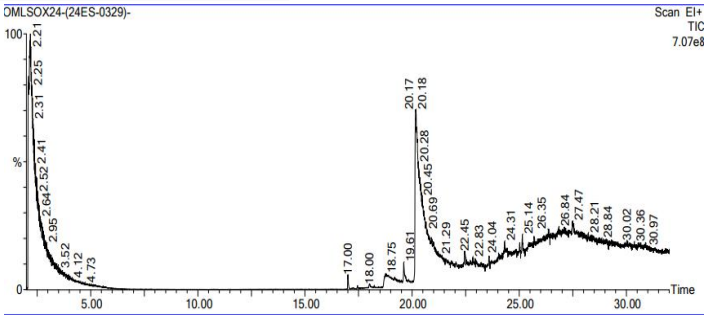

Figure 3: GC–MS chromatogram of the methanolic extract of *O. mungos* leave.

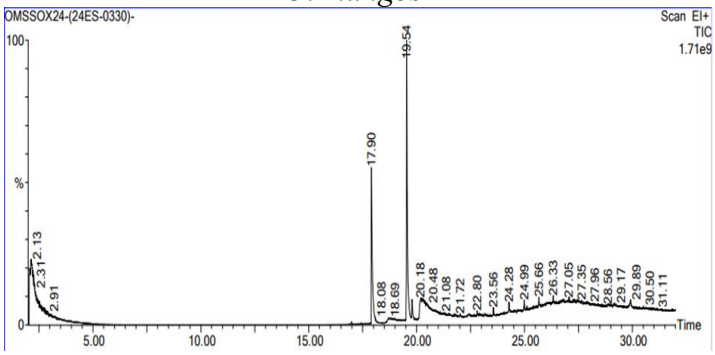

Figure 4. GC–MS chromatogram of the methanolic extract of *O.mungos* stem.

Methanol extracts of *O.mungos* against *S. litura* larvae

The results recorded during the experiment states that the methanolic extracts of *Ophiorrhiza mungos* were found to have a larvicidal activity on *S. litura* larvae. The crude methanolic extract of *Ophiorrhiza mungos* leaves and stem showed larvicidal effects after 24 hrs of exposure. The mortality rate of *S. litura* larvae increased with the increase in concentration of the *Ophiorrhiza mungos* crude extracts. The highest mortality of 95% for *O. mungos* stem extract was found to be at 5 mg/mL concentration, while the lowest mortality rate of 35% was recorded at 0.5 mg/mL concentration of the stem extract as indicated in Fig 5, Similarly for leaves extract highest mortality of 91.65% was observed at 5 mg/mL concentration, while lowest mortality of 13.3 % was noted at 0.5mg/mL concentration as indicated in Fig 6.

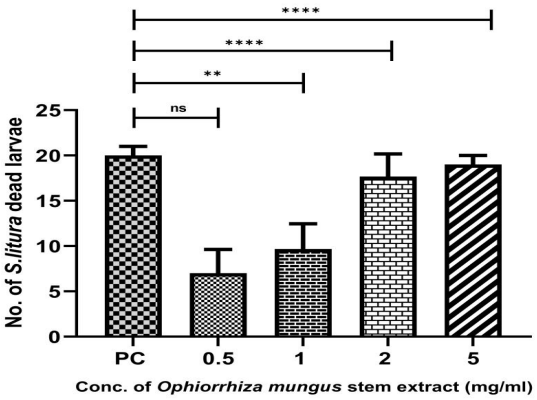

Figure 5. Effect of *O. mungos* stem extract against S. *litura.*

Values are represented in Mean ± SEM from three independent experiments. *P<0.0001 (one-way ANOVA followed by Holm-Sidak post-hoc test) compared to the Positive Control treated group. All the data represents an average of n=3 independent experiments. Here, ****, p<0.0001(Highly significant); **, p < 0.01 (significant), ns: not significant.

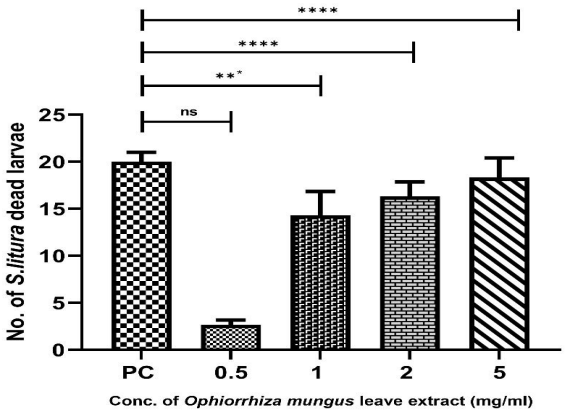

Figure 6. Effect of *O.mungos* leave extract against *S.litura.*

Values are represented in Mean ± SEM from three independent experiments. *P<0.0001 (one-way ANOVA followed by Holm-Sidak post-hoc test) compared to the Positive Control treated group. All the data represents an average of n=3 independent experiments. Here, ****, p<0.0001(Highly significant); **, p < 0.01(significant), ns: not significant.

DISCUSSION

Chemical-based insecticides and pesticides are turning increasingly ineffective against mosquitoes and agricultural pests, as these are demonstrating a growing capacity to develop resistance to commercial chemicals (Hemingway *et al.,* 2000; Silva *et al.,* 2008). The key factors responsible in inducing insecticidal resistance include biological factors pertaining to insect's developmental cycle, rate of growth of insect population, genetic makeup of the insect, its behavioral factors including movement pattern and more importantly repeated exposure rate to chemicals (Polson *et al.,* 2011). Typically, commercial chemicals consist of synthetic mixtures of two or three compounds. This issues is addressed with the replacement and extensive evaluation of natural-based insecticides primarily sourced from plants for their effectiveness in controlling various species of disease vectoring mosquitos and agricultural pests (Piesik *et al.,* 2007; Piesik *et al.,* 2013; Piesik *et al.,* 2014; Thanigaivel *et al.,* 2019; Vasantha-Srinivasan *et al.,* 2021;). Botanical extracts and their principal derivatives are crucial for managing insect pests, as these extracts are rich in diverse array of natural chemicals, including phenolic compounds, alkaloids, tannins, flavonoids, steroids, terpenes, coumarins, terpenoids, and lignins (Chellappandian *et al.,* 2018). The bioactive compounds isolated from the plant extracts are natural, facilitating their decomposition in the environment post-application, which reduces pollution and mitigate environmental harm in comparison to the synthetic pesticides (Liu *et al.,* 2017). Botanical insecticides, with their diverse insecticidal compounds and distinct mechanisms of action, makes less likely for pests to develop resistance, though developed can be tackled by increasing ecological heterogeneity and by the use of diversified bio pesticides (Mangan *et al.,* 2023). Also bioactive compounds from plants that are insecticides are generally found to be highly selective, therefore exhibiting low toxicity to humans, livestock, and natural predators, and are relatively cost-effective to produce and utilize

(Chellappandian *et al.,* 2018). In this study, a qualitative evaluation of the crude methanol extract of *O. mungos* leave and stem revealed the existence of various compound classes such as alkaloids, saponins, terpenes, steroids, carbohydrates, and phenolics. These compounds are associated with different biological activities, likely due to their free radical scavenging, antimicrobial, insecticidal, anticancer, anti-venom, mosquito larvicidal properties and many more (Tanaka *et al.,* 1989; Rajani *et al.,* 2001; Krishnan *et al.,* 2014;Taher *et al.,* 2020;).

The total phenolic content in *O. mungos* was recorded to be 21.17±0.011 mg GAE/g in the leaves and 16.22±0.115 mg GAE/g in the stems. The high phenolic content in the leaves of *O. mungos* suggests a significant potential for antioxidant activity. A similar study by Jayadev *et al.* (2013) reported a significant value of 46 mg tannic acid/ml in the leaves and 17 mg tannic acid/ml in the stems. Flavonoids, which are secondary metabolites widely regarded for their antioxidant properties, have their effectiveness influenced by the number and arrangement of free hydroxyl (OH) group (Smith *et al.,* 2023). The total flavonoid content in the *O. mungos* extracts, expressed in mg quercetin equivalent, was recorded to be 28.52±0.120 mg QE/g in the leaves and 42.80±0.096 mg QE/g in the stems. Similar study by Mohan *et al.* (2012) on *O. mungos* leave extract indicated the maximum flavonoid content of 56 µg/0.1 mg, followed by the root and stem with 23 µg/0.1 mg and 18 µg/0.1 mg, respectively. The results of the GC-MS analysis of phytochemical compounds in *Ophiorrhiza mungos* indicated the seven in leaves and ten compounds in the stem. This work is the first report for the larvacidal study on *Spodoptera litura* as promising biopesticide with highest mortality of both the extracts for the concentration of 5mg/ml. However further studies to purify the extract and evaluate the phyto-compound for their larvicidal activity is to be done to make them a potential commercial bio pesticide.

AUTHOR CONTRIBUTION

Saraswathi Saraswathi: Conceptualization, Supervision, Formal analysis, Methodology, Writing - review & editing. Jagadisha Tavarekere Venkataravanappa: Conceptualization, Resources, Supervision, Validation, Writing - review & editing. Esther Shoba R, Ashok Dhayalan, Debojyoti Chatterjee, Jikki M Roy, Mahima Manoharan, Rupsa Dey, Naveen Kumar S, and Thyloor Rama: Validation, Writing – review.

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