

Bioactivities of the Peacock Flower *Caesalpinia pulcherrima* (L.) Sw. Against Mosquitoes and Three Stored Product Pests

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Abstract

Petroleum ether, chloroform (CHCl_3) and methanol (CH_3OH) extracts of the flowers and leaves of *Caesalpinia pulcherrima* (L.) were evaluated for their larvicidal activity against *Culex quinquefasciatus* Say and their repellent activity against three stored grain pests: *Tribolium castaneum* (Hbst.), *Sitophilus oryzae* (L.) and *Callosobruchus chinensis* (L.). The LC_{50} values of the petroleum ether and CHCl_3 extracts of the flowers were 119.80, 37.34, 25.06, 17.12, and 10.36 ppm and 101.04, 76.67, 66.67, 58.24, and 51.82 ppm, respectively, after 24, 30, 36, 42, and 48 h of exposure. The CHCl_3 leaf extract showed LC_{50} values of 361.21, 321.10, 245.41, 207.09, and 203.19 ppm after 24, 30, 36, 42, and 48 h of exposure, respectively. Depending on the intensity of larvicidal activity on the mosquito, the extracts can be classified in descending order as follows: petroleum ether (flower) $>\text{CHCl}_3$ (flower) $>\text{CHCl}_3$ (leaf). Extracts of flowers with petroleum ether demonstrated a highly significant repellent effect against adult *T. castaneum* ($p<0.01$), extracts of leaves with CHCl_3 and CH_3OH showed a moderately significant repulsion against adult *T. castaneum* ($p<0.05$), and extracts of leaves with petroleum ether also showed a moderately significant repulsion against adult *S. oryzae* ($p<0.05$).

Keywords: *Caesalpinia pulcherrima*, *Culex quinquefasciatus*, *Tribolium castaneum*, *Sitophilus oryzae*, *Callosobruchus chinensis*, Larvicidal activity, Repellent activity.



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Introduction

Medicinal plants are attracting increasing interest due to their minimal side effects, allowing them to manage or cure a wide range of ailments and thus playing an important role in the discovery of new drugs. *Caesalpinia pulcherrima* (L.) Sw. is one such plant, widely used in traditional medicine to treat various diseases, which has led researchers to discover the different phytochemical compounds it produces (Bhagya 2024). It is also known as peacock flower, is an ornamental and medicinal plant found along roadsides. Different parts of this plant have been studied for their toxicological properties. The essential oil from its root has been shown to have insecticidal properties against the stored-food pests *Tribolium castaneum* and *Callosobruchus analis*, with mortality rates of 20% and 40%, respectively (Erharuyi et al. 2017). The ethanolic and acetone extracts of its leaves possess ovicidal activity (Krishnaveni et al. 2023). Both ethanolic and aqueous extracts exhibit antibacterial effects against various bacterial strains (Gbenga et al. 2024). This plant has a long history of use in various medicinal contexts, making it one of the most common medicinal plants and playing a key role in the treatment of fever, microbial infections, cancer, and in several other traditional medicine systems (Deepika et al. 2020).

Leaf extracts of this plant are used for wound healing, likely due to the presence of phenolic compounds and their antioxidant, antimicrobial, analgesic, anti-inflammatory, and astringent properties (Pusztahelyi et al. 2015). The plant produces secondary metabolites as a chemical defense mechanism against microbial destruction and oxidative stress, notably through the formation of triplet chlorophyll radicals, single hydroxyl radicals and oxygen radicals (Gechev et al. 2006, Rao et al. 2007). A detailed and thorough review of this plant was published by Krupanidhi et al. (2024) reporting all its potential anti-arthritic, anti-asthmatic, analgesic, antibacterial, anticonvulsant, antidiarrheal, antidiabetic, antifertility, antifungal, anthelmintic, anti-inflammatory, antimarial, antimetastatic by molecular docking, antinociceptive, antioxidant, antituberculosis, antitussive, antiulcer,

antiviral, anxiolytic, arginase inhibitor, cytotoxic, immunosuppressive, immunomodulatory, larvicidal, leishmanicidal, hepatoprotective, purgative and insecticidal activity.

A similar publication by Sangeetha et al. (2024) confirms the findings of Krupanidhi et al. (2024), which mention the numerous therapeutic properties of this plant. Numerous phytoactive compounds, including glycosides, rotenoids, benzoic acid, tannins and hydrocyanic acid, isoflavones, flavanones, chalcones, flavanols, flavones, and sterols, have been identified in *C. pulcherrima*, and even the authors described this plant as "marvelous" in the title of their publication. These properties are reportedly used to treat cholera, ulcers, fever, tumors, asthma, miscarriages, promote menstruation, act as a purgative (or laxative), boost energy, relieve chest pain, treat bronchitis, and treat malarial fevers. Furthermore, the plant is said to possess antimicrobial, analgesic, anti-inflammatory, antioxidant, anthelmintic, antiulcer, cytotoxic, antiviral, anticancer, antidiabetic, immunosuppressive, and vasorelaxant properties. Thus, information on the chemistry and biological potential of *C. pulcherrima* is abundant, particularly concerning the biological activities of extracts from its roots, leaves, wood, and even flowers on various test organisms. However, this study aimed to better define its activity against stored-food pests, using the leaves and flowers as test material. It was therefore conducted to evaluate the larvicidal activity of *C. pulcherrima* extracts against the mosquito *C. quinquefasciatus* and their repellent activity against three stored-food pests: *T. castaneum* (Hbst.), *S. oryzae* (L.), and *C. chinensis* (L.).

The southern house mosquito *C. quinquefasciatus* is a well-known vector for human lymphatic filariasis, caused by the parasitic nematodes like *Wuchereria bancrofti*, *Brugia malayi* and *Brugia timori*, which are transmitted from person to person by the mosquito (Bockarie et al. 2009, Chhabri et al. 2017). The red flour beetle *T. castaneum* (Hbst.) is recognized globally as a pest of stored products, particularly cereals, and is widely used as a model organism in food safety research (Ridley et al. 2011, Grünwald et al. 2013). The rice weevil *S. oryzae* (L.) mainly infests stored rice, reducing its nutritional value and converting it into unusable protein residues (Benhalima et al. 2004, Akhtar et al. 2015, Zakladnoy 2018). The pulse beetle *C. chinensis* (L.) is a pest which is generally associated with legume processing industries and inflicts significant damage to stored legumes (Ahmed et al. 2018, Seni et al. 2025).

Materials and Methods

Collection of plant and preparation of extracts

Fresh leaves and flowers of *C. pulcherrima* were collected in September 2024 in Chowddopai, Rajshahi. The plant's phylogenetic position was verified by a taxonomist from the Department of Botany at Rajshahi University. The plants were chopped, shade-dried, and then ground into a fine powder using an electric grinder. The resulting powder was weighed and placed in an Erlenmeyer flask. The solvent was added in two stages, at a rate of 60 g × 180 mL, and the mixture was stirred for 48 hours. After filtration through Whatman filter paper (made in China), the petroleum ether extract was collected in a round-bottom flask. The same procedure was repeated with chloroform (CHCl_3) and methanol (CH_3OH) (Mohammad et al. 2025) (Fig. 1&2). After evaporation, the extracts were transferred into glass vials, labeled and kept refrigerated at 4°C until use.

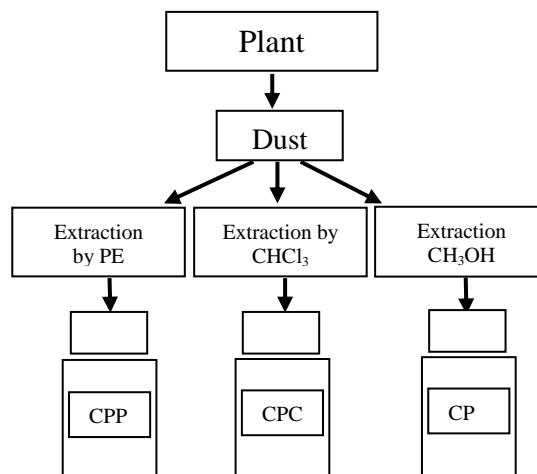


Fig. 1: Collection of *C. pulcherrima* extracts using organic solvents.



Fig. 2: Extracts in bottles with labels.

Collection of the test insects and their culture

Egg masses of *C. quinquefasciatus* were collected from campus gutters and reared at the Crop Protection and Toxicology Laboratory of the Zoology Department at Rajshahi University. These egg masses were placed in 500 ml beakers containing filtered pond water. After 24 h, the eggs hatched, and the larvae were collected and used for experiments. Adult beetles of *T. castaneum*, *S. oryzae*, and *C. chinensis* were collected in the same laboratory. Stocks and subcultures were maintained throughout the experimental period to ensure a continuous supply of insects for testing (Chhabi et al. 2017).

Larvicidal activity tests

Extracts of *C. pulcherrima* flowers in petroleum ether (concentrations of 50, 30, and 10 ppm) and extracts of flowers in chloroform (concentrations of 80, 70, 60, and 50 ppm) were used for the larvicidal activity test against *C. quinquefasciatus* (Fig. Extracts of *C. pulcherrima* leaves in chloroform (concentrations of 300, 250, and 200 ppm) were also tested. Each test tube contained ten newly hatched larvae at the indicated concentrations. Mortality was observed after 6, 12, 18, 24, and 30 h of exposure (Fig. 3).



Fig. 3: Bioassay with plant extracts on *C. quinquefasciatus* larvae.

Repellent activity tests

The repellent activity test was adapted from the method of McDonald et al. (1970) with some modifications. Filter paper half-discs (Whatman No. 40, 9 cm in diameter) were used. Each half-disc was cut in half, and one of the halves was treated with specific doses of petroleum ether/CHCl₃/CH₃OH extracts: 0.1571, 0.0785, 0.0392, 0.0196, and 0.0098 mg cm⁻². Each treated half-disc was then taped lengthwise to a control half-disc, and the assembly was placed at the bottom of Petri dishes intended to hold *T. castaneum*. For the repellent activity test on *C. chinensis* and *S. oryzae*, similar Petri dishes were divided into three zones by placing two

narrow sticks on the ground using adhesive tape, and the two parts on each side were filled with treated and untreated food, in the central part where ten beetles of the same age were released and the doses were the same as those mentioned above. Their orientation was modified to prevent external directional cues from affecting the distribution of the tested insects. Ten adult insects were placed in the center of each circle of filter paper. The same procedure was applied to all extracts. The concentration of the extracts in each solvent was tested five times. Insects deposited on each untreated portion of the Petri dishes were counted after 1 h, and then data were collected hourly five times. Using the formula of Talukder and Howse (1993, 1995), the count means were converted to percentage repulsion (PR) = $(N_c - 5) \times 20$, where N_c represents the hourly mean number of insects observed on the untreated halves of the Petri dishes (Fig. 4).

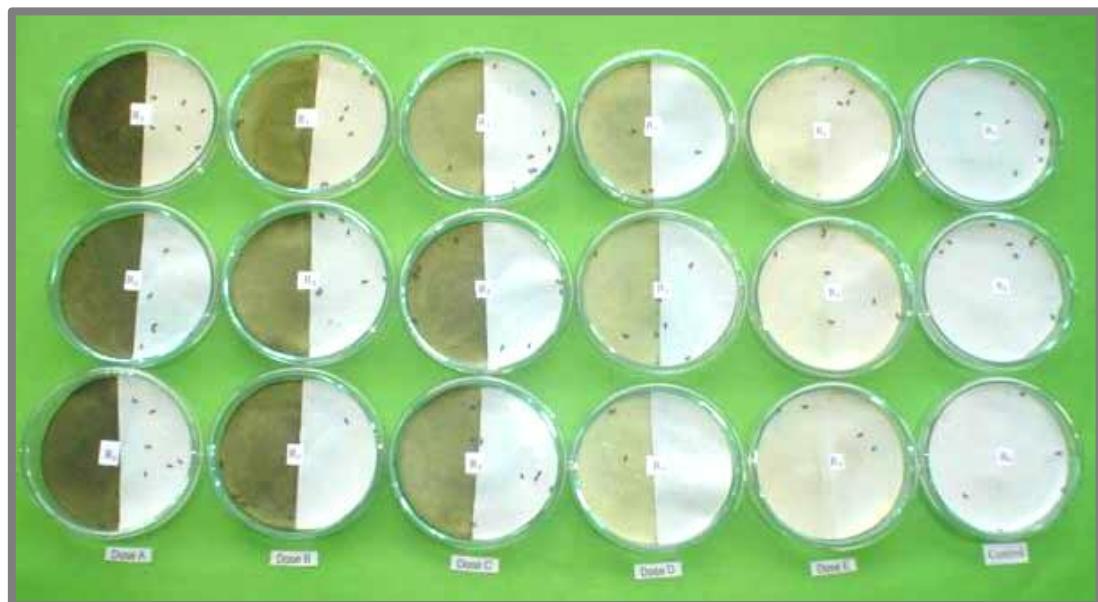


Fig. 4: Experiment on the repellent activity test with an extract of *C. pulcherrima* against *T. castaneum* adults according to McDonald et al. (1970).

Statistical analyses

Larval mortality (%) was corrected using Abbott's formula (Abbott 1925). The data were then subjected to probit analysis according to Finney (1947) and Busvine (1971). Data relating to the percentage of repulsion were subjected to analysis of variance (ANOVA) to compare means at significance levels of 5% and 1% (Chhabhi et al. 2017).

Results

Larvicidal effects on mosquito

Petroleum ether, CHCl_3 , and CH_3OH extracts of *C. pulcherrima* flowers and leaves were evaluated for their larvicidal activity against *C. quinquefasciatus*. Both the petroleum ether and CHCl_3 flower extracts, as well as the CHCl_3 leaf extract, showed larvicidal activity against the tested larvae. The CHCl_3 leaf extract exhibited LC_{50} values of 361.21, 321.10, 245.41, 207.09, and 203.19 ppm after 24, 30, 36, 42, and 48 h of exposure, respectively. The petroleum ether flower extract showed LC_{50} values of 119.80, 37.34, 25.06, 17.12, and 10.36 ppm at the same exposure intervals. Meanwhile, the CHCl_3 flower extract showed LC_{50} values of 101.04, 76.67, 66.67, 58.24, and 51.82 ppm after 24, 30, 36, 42, and 48 hours of exposure, respectively (Fig. 5).

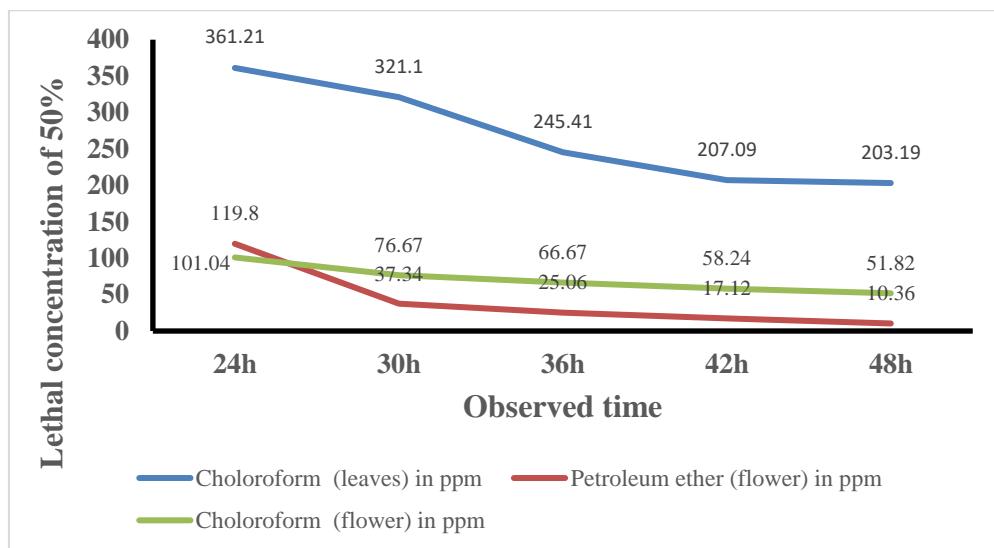


Fig. 5: Larvicidal activity of *C. pulcherrima* against *Culex quinquefasciatus*.

Repellent activities against stored pests

Only the petroleum ether flower extract demonstrated a significant repellent effect against adult *T. castaneum* ($p<0.01$). The other extracts showed no repellent activity against *T. castaneum*, *S. oryzae*, or *C. chinensis*. Among the leaf extracts, the CHCl_3 and CH_3OH extracts showed moderately significant repellent activity against adult *T. castaneum* ($p<0.05$), and the petroleum ether leaf extract also showed moderate repellent activity against adult *S. oryzae* ($p<0.05$) (Table 1).

Table 1: Summary of the repellent activities of *C. pulcherrima* leaf and flower extracts against three stored-food pests.

Plant parts	Test agents	Solvent of extraction	Sources of variation			F-ratio with level of significance		P- values	
			Bd	Bti	Error	Bd	Bti	Bd	Bti
Leave	<i>T. castaneum</i>	Pet. ether	4	4	16	4.98	1.52	0.008	0.242
		CHCl_3	4	4	16	13.44*	1.05	5.5E-05	0.413
		CH_3OH	4	4	16	12.31*	1.56	9.2E-05	0.233
		Pet. ether	4	4	16	17.68*	1.96	1.0E-05	0.151
	<i>S. oryzae</i>	CHCl_3	4	4	16	3.12	2.74	0.045	0.066
		CH_3OH	4	4	16	3.43	0.47	0.033	0.755
	<i>C. chinensis</i>	Pet. ether	4	4	16	2.20	1.62	0.115	0.217
		CH_3OH	4	4	16	4.79	10.38	0.011	0.0002
Flower	<i>T. castaneum</i>	Pet. ether	4	4	16	25.30**	2.83	9.5E-07	0.060
		CHCl_3	4	4	16	2.12	3.60	0.125	0.028
		CH_3OH	4	4	16	1.23	2.96	0.337	0.052
		Pet. ether	4	4	16	5.61	3.24	0.005	0.041
	<i>S. oryzae</i>	CHCl_3	4	4	16	1.55	4.28	0.235	0.015
		CH_3OH	4	4	16	4.08	0.38	0.018	0.821
	<i>C. chinensis</i>	Pet. ether	4	4	16	2.72	0.11	0.067	0.978
		CHCl_3	4	4	16	4.96	2.81	0.009	0.061
		CH_3OH	4	4	16	4.19	0.38	0.016	0.821

Discussion

Laboratory experiments were conducted to evaluate the larvicidal and repellent activity of petroleum ether, CHCl_3 , and CH_3OH extracts of *C. pulcherrima* flowers and leaves against mosquitoes and three stored-food pests. These experiments yielded satisfactory results. The CHCl_3 leaf extract exhibited LC_{50} values ranging from 361.21 to 203.19 ppm after 24 to 48 h of exposure, respectively. The petroleum ether flower extract exhibited LC_{50} values ranging from 119.80 to 10.36 ppm after 24 to 48 h of exposure, respectively. Meanwhile, the CHCl_3 flower extract showed CL_{50} values ranging from 101.04 to 51.82 ppm after 24 to 48 h of exposure, respectively.

Previous studies on the biological potential of *Caesalpinia* spp. have corroborated the findings of this research. Krishnaveni et al. (2023) demonstrated the ovicidal activity of ethanolic and acetone extracts of *C. pulcherrima*, and their results showed that these extracts significantly reduced the hatching rate of mosquito eggs. These results are consistent with those of Cai et al. (2004) and Badami et al. (2003), who demonstrated that extracts prepared from ethyl acetate, methanol, and water all exhibited strong antioxidant activity, reducing harmful free radicals, both *in vitro* and in living organisms. Administered at certain doses (50 and 100 mg/kg body weight) to living models, these extracts not only neutralized free radicals, but also improved levels of important protective enzymes such as superoxide dismutase (SOD) and catalase. These enzymes help our cells manage oxidative stress, that is, damage caused by free radicals (Badami et al. 2003, Cai et al. 2004). Further experiments conducted by these same authors showed that *C. pulcherrima* extract possesses anti-inflammatory activity. Kim et al. (2004) highlighted that extracts, including chloroform, methanolic, and aqueous solutions, were able to lower the minimum concentration required to inhibit the growth of microorganisms. You et al. (2005) described how an important active compound in this plant, braziline, reduced blood glucose levels in diabetic animals.

Krupanidhi et al. (2024) demonstrated the full range of pharmacological properties of *C. pulcherrima*, including antiarthritic, antiasthmatic, analgesic, antibacterial, anticonvulsant, antidiarrheal, antidiabetic, antifertility, antifungal, antiflamminic, anti-inflammatroy, antimarial, antimetastatic, antinociceptive, antioxidant, antitubercular, antitussive, antiulcer, antiviral, anxiolytic, arginase inhibitor, cytotoxic, immunosuppressive, immunomodulatory, larvicidal, leishmanicidal, hepatoprotective, purgative, and insecticidal effects. The oil extracted from the roots of this plant induced a mortality rate of 20% in *Tribolium castaneum* and 40% in *Callosbruchus analis*. Sangeetha et al. (2024) confirmed the properties mentioned above, specifying that different parts (leaves, flowers, bark, seeds) of this beautiful, colorful ornamental plant with distinctive foliage are used in traditional medicine to treat ailments such as intestinal worms, coughs, and as a purgative or abortifacient. It contains compounds with potential anti-inflammatory, antimicrobial, antihypertensive, antiulcer, and anticancer properties, among others. Uadia et al. (2023) conducted a phytochemical study, an analysis of the immediate composition, acute toxicity, anti-inflammatory and antinociceptive activities of extracts from *Caesalpinia pulcherrima* flowers which could be considered as further confirmation of the potential of peacock flower. This study focused on the larvicidal and repellent activity of the tested plant, corroborating previous findings (Govindarajan et al. 2011). These results suggest that crude extracts of *C. pulcherrima* could be a promising source for mosquito control. Furthermore, repellent activity tests were conducted on stored products, confirming the plant's repellent effect and suggesting another potential application for protecting stored goods.

Conclusion

The results of this study on the repellent activity tests and larvicidal potential of *C. pulcherrima* have highlighted the need to continue research on its potential future applications in the mosquito, storage pests and vector control sectors in order to make biodegradable and environmentally friendly repellents and agents for the treatment of drainage and stagnant water available on the market.

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Conflict of interest: The authors declare no conflicts of interest.

Author's contribution: SS, PCR and AK collected samples and data, conducted experiments, writing the draft, analysis was done statistically. NI designed the conceptualization and supervised the study, writing review and editing the manuscript. All authors have read and approved the final manuscript.

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Data availability: The data used the current study are available from the corresponding author upon reasonable request.

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