EFFICACY OF EUPATORIUM ADENOPHORUM EXTRACT AGAINST PULSE BEETLE (CALLOSOBRUCHUS CHINENSIS) AT PAKLIHAWA CAMPUS, RUPANDEHI

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ABSTRACT

A laboratory experiment was carried out at Paklihawa Campus, Rupandehi starting from 15th December 2020 to 15th February 2021, to observe the efficacy of *Eupatorium adenophorum* extract against *Callosobruchus chinensis* (pulse beetle). A completely randomized design with two factors (altitude and concentration) was taken. Phytochemical screening and allelopathic test were completed during the research. Samples were collected from three altitudes, viz., Paklihawa, Hemja, and Tansen, and at three levels of concentration (5%, 10%, and 15%). This study explored the potential of *E. adenophorum* leaf extract as a natural pesticide against pulse beetles in stored pigeon peas. Phytochemical screening confirmed bioactive compounds like alkaloids, flavonoids, and terpenoids. Extracts from different altitudes and concentrations (5%, 10%, and 15%) were tested, revealing that higher altitude samples and higher concentrations significantly increased beetle mortality, achieving complete mortality by the tenth day at 15%. The extract also showed inhibitory effects on pigeon pea germination, suggesting allelopathic properties. Overall, *E. adenophorum* offers an eco-friendly alternative for pulse beetle management, supporting sustainable pest control practices.

Keywords: efficacy, altitude, concentration, mortality

INTRODUCTION

The genus *Eupatorium adenophorum* is related to the Asteraceae family, historically including approximately 1200 species. However a comprehensive study by Zachariades et al. (2009) led to the reclassification of about 1010 species into different genera. *E. adenophorum*, commonly known as Banmara, were to Southeast Asia from the West Indies, as noted by Biswas (1934). He suggested that it likely arrived in Singapore via cargo boats and spread to other humid tropical regions across South and Southeast Asia. Since then, it has spread to Indonesia, Philippines, Laos, Cambodia, Vietnam, China, Sri Lanka, Nepal, Bhutan and Bangladesh. It was spread in South-western part of India during the Second World War, probably through the contaminated clothing from the returning soldiers (Bennett & Rao, 1968). Robinson and King (1980) reported that the genus *Eupatorium* is native to Mexico, is now globally distributed and the plant holds value in traditional medicine, used as an antimicrobial, antiseptic, analgesic and antipyretic (Bhattarai & Shrestha, 2009).

Extraction is the separation of medicinally active portions of plant and animal tissues using selective solvents through standard procedures. The resulting products are typically complex mixtures of metabolites, in liquid or semisolid state or after removing the solvent in dry powder form and are intended for oral or external use. Common preparation types

include decoctions, infusions, fluid extracts, tinctures, pillar (semisolid) extracts or powdered extract (Akerele, 1993). However, chemical extraction methods include challenges, like health hazards to the users and consumers. It causes residual toxicity, environmental pollution and development of pesticide resistance against bruchids (Srivastava et al., 1991). While extraction methods provide valuable preparations for medicinal use, challenges with chemical methods—such as residual toxicity and environmental impact—also extend to pest control in agricultural storage, which has been concern of farmers and scientists ever.

The pulse beetle (*Callosobruchus chinensis*) is a major pest of leguminous stored seeds. Synthetic chemicals have become a common practice among the farmers and stockholders to control the storage pests of pulses (Khan et al., 2015). These pulses are the main food of one-third of the world's poorest people in Africa and Southeast Asia. Bruchids are important storage pests of grain legumes known to cause considerable economic losses, especially in pulses grown in the tropics and sub-tropics (Srivastava & Pant, 1989). Pulse beetle is the major storage pest, which attack, and causes damage to almost all the pulse crops such as pigeon pea, cowpea, lentils, green gram and black gram and affects quality of seeds badly and causes 50% damage during storage within 3 to 4 months (Raghavendra & Loganathan, 2017).

Bruchids are among the most damaging insects affecting stored chickpea, causing upto 50 % damage during three to four months. Although the infestation begins in the field, it continues and intensifies during storage. The larvae of bruchids feed and develop solely on the legume seed of legumes, while the short-lived adults (one to two weeks) do not require food or water, dedicating their time to mating and laying eggs on seeds (Kar & Ganguli, 2016).

Efforts are underway to explore alternative, non-toxic, environmentally friendly, and safe methods for controlling pulse beetles, such as using natural fumigants. Research on the efficacy of E. *adenophorum*. extract in protecting stored maize grain has shown promising result; it was found, found to be effective as certain synthetic pesticide, reducing maize weevil damage by 25 % (Rajashekar et al., 2014).

Plants contain variety of important metabolites including phenolics, alkaloids, flavonoid, terpenoids, momilactone, hydroxamic acids, brassinosteroids, jasmonates, salicylates, glucosinolates, carbohydrates and amino acids (Dahiya et al., 2017). These compounds can play a role in allelopathy, a process in which a plant can either stimulate or inhibit the growth of other nearby plants through the production of allelochemicals (Tomilov et al., 2006). Extensive research works were done to explore the inhibitory potential of different allelopathic crops (Cheema et al., 2004; Farooq et al., 2012; Iqbal et al., 2007).

Gupta et al. (1992) reported several plant extracts against the stored product pest, *C. chinensis*, Petroleum ether extract of neem (leaves and twigs), *E. adenophorum*. (leaves and flowers), *Ageratum conyzoides* (leaves, flower and buds), *Thevetia neriifolia* (leaves, twigs and young branches) and *Ipomea carnea* (leaves) diluted in benzene and mixed with seeds of green gram at 0.5, 1.0 and 1.5 parts / 100 parts seed (w/w) were highly repellent to the pest, inhibiting oviposition and protecting against infestation. A newly emerged pair of adult beetles is released onto a host seed, they do not mate immediately; instead, they first explore the host environment. On average, the initial mating takes about five minutes. After mating, the female begins moving around the seed surface and the periphery of the petri-dish initiating egg-laying approximately one hour and thirty minutes later. This behavior aligns with the observation by Wasserman and Asami (1985) who described the ovipositional behavior of

C. chinensis in three phases: patrolling the seed surface, preparation for oviposition and oviposition.

Kiradoo and Srivastava (2010) reported that egg-laying in normal sets was observed to be 37.33/pair, whereas in control sets treated with distilled water, ethanol and DEE extract the rates dropped to 30.66, 28.33 and 27.66 eggs per pair respectively. *E. adenophorum*, known for its light-stimulated germination, effectively invades barren or disturbed areas but can also thrive in shaded environments once established.

Thus, this study explores whether *E. adenophorum*, an invasive plant, is a natural pesticide against pulse beetle. Developing eco-friendly pest control strategies like this could reduce the reliance on chemical insecticides and offer a beneficial use.

MATERIALS AND METHODS

Study Site

The laboratory experiment was done in the agro-ecology laboratory of Institute of Agriculture and Animal Science Paklihawa, Rupandehi district located at 27.5065°N, 83.4377°E situated at an altitude of 87 MSL (meters from the sea level). The rationale behind the selection of this site is accessibility of plant samples, laboratory, required chemicals and other apparatus.

Collection of samples

Sample was collected from Paklihawa, Rupandehi from an altitude of 87 MSL. Another sample level from Hemja, Pokhara with an altitude of 940 MSL. Third elevation was at 1350 MSL from Tansen, Palpa.

Cleaning and drying of plant parts

The major used plant parts were leaves and twigs. The plant parts were separated, thoroughly cleaned and spread inside the room for shade drying at room temperature (30 \pm 5 °C) for twelve days.

Preparation of crude powder

After shade drying, the sample was ground using an electric mixer and then sieved to obtain a fine powder. This powder was stored in a plastic jar with an airtight lid to maintain its quality until needed.

Preparation of stock solution

Fifty grams of crude powder (CP) from each sample were soaked separately in 250 ml of different solvents (acetone, methanol, petroleum ether, and distilled water) in airtight plastic bottles and left overnight. The mixtures were then filtered using Whatman filter paper, boiled for 5 minutes, and allowed to cool.

Qualitative phytochemical screening

The presence of phytochemical constituents such as alkaloids, flavonoids, phenols and terpenoids, was screened using several standard tests. These included Mayer's test, Wagner's test, Shinoda test, Ferric chloride test, Salkowski test, copper acetate test and Foam test (Gautam et al., 2021; Khanal & Neupane, 2018; Poudel & Khanal, 2023; Visweswari et al., 2013).

Quantitative phytochemical screening

To evaluate the effects of varying concentration of phytochemicals, quantitative analysis was conducted for alkaloids, flavonoids, terpenoids and saponins. The obtained yield was expressed in terms of milligrams of gallic acid equivalent (GAE) per gram or milligram. For this study, the results were presented as percentages (Harborne, 1998; Harborne & Harborne, 1973; Khanal, 2021; Mir et al., 2013).

Allelopathic test

Pigeon pea seeds (full grain) used as a sample to observe the allelopathic effect of botanical extracts. Completely randomized design with ten treatments and five replications including control group. Nine treatments included three levels of altitude and concentration, while the tenth treatment involved distilled water. In each petri dish, twenty-five seeds were arranged in a ring structure, with sixteen seeds in the outer ring and eight in the center arrangement in each petri dish (Tsombou et al., 2022). The various parameters such as germination and root length of pigeon pea were recorded on different dates. The germination percentage was recorded through the data obtained from petri plates. Similarly, radicle length was recorded and analyzed.

Mortality test of pulse beetle against E. adenophorum extract

Pulse beetle infected pigeon pea sample were collected from Paklihawa. The obtained sample was stored in jars covered with muslin cloth for good aeration and control from other pests. These jars were kept at room temperature and allowed insects to mat. Regular inspection was conducted to monitor the oviposition process. The life cycle of Pulse beetle generally completes in 25-34 days in pigeon pea, so the development of adults was kept in keen interest.

Identification of male and female bruchids was done following established guidelines. The male adult measured 3.25 ± 0.23 mm length and is 2.16 ± 0.05 mm breadth whereas the length and breadth of female adult measured with an average 3.60 ± 0.08 mm and 2.02 ± 0.04 mm respectively. Males are having strongly serrate antennae and pygidium without dark patches, while females exhibit weakly serrate antennae and pygidium with two dark patches, one on each side of the midline. Five pairs of bruchids were taken for each treatment. Generally, female beetles are slightly larger than male beetle (Devi & Devi, 2014).

A completely randomized design with nine treatments and four replications was used during the experiment. Each treatment contains treatment of three levels of concentration and three levels of altitude. The number of dead beetles after application of leaf extract in different concentrations and from different altitude was recorded till one week after experimental set up.

Statistical analysis

The data on various traits were analyzed for CRD using R stat (version 1.1.423 - 2009 – 2018 R Studio, Inc.). Analysis of variance (ANOVA) was done. Significant difference was declared for means with P value < 0.05. Then, the least significant difference test (LSD test) function was used for multiple mean separations where means with different letter heads indicated significant differences. Package "Agricolae" was used for mean separation and "dunn's test" was used for multiple pairwise comparisons. The results were interpreted in a logical manner to present the final report. Tables and figures were used wherever appropriate.

RESULT AND DISCUSSION

Qualitative phytochemical screening of Eupatorium adenophorum.

Qualitative phytochemical screening of *Eupatorium adenophorum* was done with the various tests (Visweswari et al., 2013). Results obtained in the experiment are presented in table 1.

Table 1: Phytochemical screening of Eupatorium adenophorum

S.N.	Test	Paklihawa	Tansen	Hemja
1	Alkaloid test	+	+	+
2	Flavonoid test	+	+	+
3	Phenol test	+	+	+
4	Terpenoids test	+	+	+
5	Saponins test	+	+	+

Note: (+) = presence

The results of the preliminary phytochemical analysis of *E. adenophorum* from three different altitudes are presented in Table 1. Findings indicate that *E. adenophorum* extract contains alkaloids, flavonoids, phenols, terpenoids, and saponins, aligning with previous work by Patel et al. (2011). The extraction and presence of these compounds have been documented in several studies (Albuquerque et al., 2004; Geng, 2024; Liu et al., 2015; Yungang et al., 2020; Zhang et al., 2008). However, our findings differ from Rosuman and Lirio (2016) concerning the presence of saponins. Similar studies on *Chromolaena odorata* have reported related phytochemical compositions in both Nepal (Budha Magar et al., 2023; Giri et al., 2020) and India (Sengupta et al., 2023; Vasath & MS, 2022).

Quantitative phytochemical screening

Quantification of the constituents was done to observe the difference in altitudinal samples. The impact of high or low concentration chemical contributes to the use of the extract and its properties.

Table 2: Altitudinal variation in phytochemicals concentration in *Eupatorium adenophorum* extract

Location	Alkaloids	Flavonoids	Terpenoids	Saponins
	(%)	(%)	(%)	(%)
Paklihawa	12.66 ± 0.01	19.01 ± 0.02	5.23 ± 0.02	2.50 ± 0.01
Hemja	14.20 ± 0.01	20.23 ± 0.02	6.83 ± 0.01	2.54 ± 0.01
Tansen	14.50 ± 0.02	23.44 ± 0.05	$\textbf{7.34} \pm 0.05$	$\textbf{3.22} \pm 0.01$
LSD	6.06	13.91	6.69	2.45
CV	7.16	10.95	17.02	14.69

Note: LSD= Least Significant Difference, CV= Coefficient of variation

The highest content of alkaloids 14.50 % were recorded in Tansen, followed by Hemja with 14.20 % and least from Paklihawa with 12.66 %. Similarly, flavonoids content ranges from 23.44 % to 19.01% proportional to altitude. As well as for terpenoids, we found a similar pattern from 7.34% to 5.23% and Saponins from 3.22 % to 2.50 %.

The major compounds found were flavonoids and minor were saponins. Isolation for components of flavonoids as well as other compounds was presented with their chemical structure (Bhandari et al., 2021; Geng, 2024; Giri et al., 2022; Liu et al., 2015). While chemical concentration did not directly correlate with altitude, ecological factors, soil type,

plant sample origin, and seasonal variations likely contribute (Costa et al., 2022). A general trend of concentration increasing with altitude has been presented in other studies (Rana et al., 2020), although specific plant and chemical compositions in precise percentages were not fully explored in our study.

Allelopathic test

The Allelopathic test is an important basis for efficacy of botanicals, so we observed with the standard process. Pigeon pea seeds germination and significance was presented in Table 3.

Table 3: Allelopathic effect of Eupatorium adenophorum extract

Treatment		Number of germinated seeds		
Main effect				
Altitude	Paklihawa	15.78 ± 0.29		
	Hemja	15.00 ± 0.29		
	Tansen	14.67 ± 0.29		
Probability		1.35e-08 ***		
Concentration	5%	$17.44^{b} \pm 0.29$		
	10%	$15.00^{\circ} \pm 0.29$		
	15%	$13.00^{d} \pm 0.29$		
Interaction Effect				
Altitude	Concentration			
Paklihawa	5%	$18.33^{b} \pm 0.88$		
Paklihawa	10%	$15.00^{\rm cd} \pm 0.58$		
Paklihawa 15%		$14.00^{ m de}\!\pm0.58$		
Hemja 5%		$17.00^{bc} \pm 1.54$		
Hemja 10%		$15.00^{\rm cd} \pm 1.00$		
Hemja 15%		$13.00^{de} \pm 0.58$		
Tansen	5%	$17.00^{ m bc} \pm 0.01$		
Tansen	10%	$15.00^{\rm cd} \pm 0.58$		
Tansen	15%	$12.00^{\circ} \pm 1.54$		
Control		$24.00^{a} \pm 0.58$		
LSD		2.30		
CV		8.43		
Probability		0.732 (NS)		

Note: * = Significant at p<0.05, ** = Significant at p<0.01, *** = Significant at p<0.001, NS= Non-significant, Values bearing different superscripts within altitude, concentration and interaction effect differ significantly. LSD = Least significant difference and CV = Coefficient of variation.

The Highest gemination was recorded in samples from Paklihwa while the lowest germination occurred in samples from Tansen. Results also showed that the increase in concentration of extraction also contributed to lower germination. For instance, the samples from Tansen at 15% concentration showed the lowest germination, whereas sample from Paklihawa with 5% concentration showed the highest germination. The control group pigeon pea treated with distilled water exhibited normal, suggesting that distilled water didn't have any negative role in germination of pigeon pea.

The leaf extracts from *E. adenophorum* had an inhibitory effect on seedling development of some legume crops. It was found that amylase activity was decreased as per increasing the concentration of aqueous leaf extract which indicating that allelochemicals are present in botanical extract of *E. adenophorum* which has inhibitory effect on seed germination (Madane Atul & Patil, 2017). A similar study in radish using different botanical also presented allelopathic effect of *Eupatorium spp.* (Khadka et al., 2023).

Effect in the radicle length of pigeon pea

Radicle length of pigeon pea was recorded after third, fourth, fifth and sixth days of treatment (DAT).

Table 4: Radicle length of pigeon pea

Treatment		Radicle length (cm) of pigeon pea seedling after treatment			
		3rd DAT	4 th DAT	5 th DAT	6 th DAT
Main Effect					
Altitude	Paklihawa	$0.88^{\scriptscriptstyle b} {\pm}~0.04$	$0.95^{\text{b}} \pm 0.05$	$1.02^{b} \pm 0.06$	$1.11^{\text{b}} \pm 0.06$
	Hemja	$0.80^{\text{bc}} \!\pm 0.06$	$0.86^{\circ} {\pm}~0.06$	$0.90^{\rm c} {\pm}~0.07$	$0.99^{\circ} \pm 0.06$
	Tansen	$0.75^{\circ} {\pm}~0.04$	$0.82^{\circ} {\pm}~0.05$	$0.86^{\circ} \pm 0.05$	$0.90^{\scriptscriptstyle d} {\pm}~0.04$
Concentration	5%	$1.02^{\scriptscriptstyle b} {\pm}~0.04$	$1.15^{\text{b}} \pm 0.03$	$1.20^{\text{b}} \pm 0.03$	$1.28^{\text{b}} \pm 0.04$
	10%	$0.81^{\circ} \pm 0.02$	$0.85^{\circ} {\pm}~0.02$	0.92 ° ± 0.02	$0.97^{\text{c}} \!\pm 0.02$
	15%	$0.61^{\text{d}} \!\pm 0.02$	$0.64^{\rm d}\!\pm0.02$	$0.66^{\scriptscriptstyle d} {\pm}~0.01$	$0.76^{\scriptscriptstyle d} {\pm}~0.02$
Interaction Effect					
Altitude	Concentration				
Paklihawa	5%	$0.98^{\scriptscriptstyle b} {\pm}~0.02$	$1.10^{6} \pm 0.05$	$1.17^{\mathrm{b}} {\pm 0.05}$	$1.27^{\circ} {\pm}~0.07$
Paklihawa	10%	$0.80^{\text{cd}} \!\pm 0.04$	$0.85^{\circ} {\pm}~0.02$	$0.88^{\circ} {\pm}~0.04$	$0.94^{\circ} \pm 0.03$
Paklihawa	15%	$0.62^{\circ} {\pm}~0.04$	$0.63^{\scriptscriptstyle d} {\pm}~0.03$	$0.64^{\rm fg}\!\pm0.03$	$0.75^{\mathrm{gh}} \pm 0.01$
Hemja	5%	$1.16^{\mathrm{a}} \!\pm 0.04$	$1.28^{\mathtt{a}} \!\pm 0.02$	$1.35^a \pm 0.01$	$1.44^{\scriptscriptstyle b}{\pm}~0.01$
Hemja	10%	$0.82^{\text{cd}} {\pm 0.03}$	$0.88^{\text{c}} {\pm 0.02}$	$0.99^{\rm d}\!\pm0.04$	$1.05^{\scriptscriptstyle d} {\pm 0.01}$
Hemja	15%	$0.63^{\circ} \pm 0.01$	$0.69^{\mathrm{d}} \!\pm 0.07$	$0.72^{\rm f} \pm 0.03$	$0.82^{\rm fg} {\pm~0.04}$
Tansen	5%	$0.91^{\text{bc}} {\pm 0.05}$	$1.06^{\text{b}} \pm 0.05$	$1.08^{\circ} \pm 0.04$	$1.11^{\rm d} {\pm}~0.01$
Tansen	10%	$0.76^{\scriptscriptstyle d} {\pm}~0.02$	$0.81^{\circ} \pm 0.02$	$0.87^{\circ} \pm 0.03$	$0.89^{\rm ef} {\pm~0.04}$
Tansen	15%	$0.57^{\circ} \pm 0.03$	$0.59^{\rm d}\!\pm0.01$	$0.61^{\rm g} {\pm}~0.01$	$0.70^{\rm h}{\pm}~0.03$
Control		$1.28^{a} \pm 0.08$	$1.32^{a} \pm 0.04$	1.42°± 0.01	$1.65^{a} \pm 0.03$
LSD		0.1192	0.1069	0.08949	0.08908*
CV		10.9	9.0	7.2	6.5
Probability		0.0956 (NS)	0.202 (NS)	0.0971 (NS)	0.0227*

Note: * = Significant at p<0.05, ** = Significant at p<0.01, *** = Significant at p<0.001, NS= Non-significant, Values with different superscripts within altitude, concentration and interaction effect differ significantly. LSD = Least significant difference and CV = Coefficient of variation, DAT= Days after treatment.

Findings show the highest radicle length in Hemja with 5% concentration and followed by Paklihawa. The lowest radicle length was recorded in sample from Tansen with 15% concentration. A proportionality with altitude and radicle length can be referred to from this experiment. Although a trend of increasing radicle length with decreasing altitude was noted, the results did not consistently follow a specific order across all samples. Statistically, there was no significant interaction of *E. adenophorum* extract from different location and concentration on third, fourth and fifth days of treatment. However, significant interaction was recorded after sixth days of application of extract.

The shoot and root length of the legume species varied in responses, resulting how allelopathic effects depend on the nature of the test species and the concentration of the allelochemicals (Akhtar et al., 1978; Whittaker & Feeny, 1971). Concentration has a significant impact on seed germination, radicle length, plumule (Ahmed et al., 2007).

Mortality test of pulse beetle against Eupatorium extract

To observe the efficacy of *E. adenophorum* extract against pulse beetle, the recorded data were analyzed, focusing on mortality rates of beetle after third, fifth and seventh days of application of extract. The result of action of *E. adenophorum* extract in control of pulse beetle is presented in table 5.

Treatment		Mortality of pulse beetle			
		3 DAT	5 DAT	7 DAT	
Main Effect					
Altitude	Paklihawa	$1.89^{b} \pm 0.26$	$5.89^{b} \pm 0.48$	10	
	Hemja	$1.56^{b} \pm 0.34$	$6.89^a \pm 0.20$	10	
	Tansen	$4.00^{\mathtt{a}}\!\pm0.24$	$5.33^{b} \pm 0.87$	10	
Probability		0.35954	0.000181 ***	0.387 (NS)	
Concentration	5%	2.67 ± 0.12	$5.33^{b} \pm 0.32$	10	
	10%	2.56 ± 0.26	$5.33^{b} \pm 0.24$	10	
	15%	2.22 ± 0.26	$7.44^{a} \pm 0.18$	10	
Probability		0.00055 ***	0.009839 **	0.387 (NS)	
Interaction Effe	ect				
Altitude	Concentration				
Paklihawa	5%	$1.33^{\text{de}}\!\pm0.33$	$5^{cde}\!\pm0.58$	10^{a}	
Paklihawa	10%	$2.33^{bcd}\!\pm0.33$	$7^{\mathrm{b}} \pm 0.67$	10^{a}	
Paklihawa	15%	$4.33^a \pm 0.33$	$4^{\text{de}}\!\pm1.00$	10^{a}	
Hemja	5%	$1.66^{\text{cde}}\!\pm0.33$	$5^{bcd} \pm 0.58$	10^{a}	
Hemja	10%	$1.66^{\rm cde}\!\pm0.66$	$7^{\mathrm{b}} \pm 0.01$	10^{a}	
Hemja	15%	$3.33^{ab}\!\pm0.33$	$3^{\rm e}\!\pm0.33$	10^{a}	
Tansen	5%	$2.66^{\mathrm{bc}}\!\pm0.33$	$7^{\rm b} \pm 0.58$	10^{a}	
Tansen	10%	$0.66^{\rm e}\!\pm0.33$	$6^{bc} \pm 0.33$	10^{a}	
Tansen	15%	$4.33^a \pm 0.33$	$8^a \pm 0.33$	10 ^a	
LSD		1.14	1.65	1.52	
CV		26.85	15.93	8.88	
Probability		0.00996 **	0.000600 ***	0.433 (NS)	

Note: * = Significant at p<0.05, ** = Significant at p<0.01, *** = Significant at p<0.001, NS= Non-significant, Values bearing different superscripts within altitude, concentration and Interaction effect differ significantly. LSD = Least significant difference and CV = Coefficient of variance, DAT= Days after treatment

On the third day of treatment with 15% concentration of *E. adenophorum* extract, the samples from Paklihawa and Tansen exhibited the highest beetle mortality. On the seventh day of treatment, the same sample collected from Tansen showed the highest mortality. On the tenth day, all bruchids in the treated sample were recorded dead.

The experiment demonstrated a significant interaction between altitude and concentration on the third and fifth days of treatment with higher altitude and higher concentration showing the greatest efficacy. The higher mortality of the pulse beetle from the extract obtained from higher altitudes might be due to the presence of secondary metabolites in the plant at those altitudes.

This finding concludes that *E. adenophorum* leaf extract is efficient in controlling the pulse beetle in pigeon pea. Thus, for the control of pulse beetles, *E. adenophorum* extract can be applied. If the control is required in a short period of time, a higher concentration can be applied, whereas if the damage is not severe and a lower concentration is required, *E. adenophorum* can be applied to control the pulse beetle (Kar & Ganguli, 2016).

An ecofriendly management of pulse beetle adopted with this *E. adenophorum* extract (Khan et al., 2015). Pulse beetle being an important pest of the stored grain legumes could be controlled on oviposition stages (Raghavendra & Loganathan, 2017).

CONCLUSION

This study demonstrated that *Eupatorium adenophorum* extracts hold promising potential for eco-friendly management of pulse beetle (*Callosobruchus chinensis*) infestation in stored pigeon peas. Phytochemical analysis revealed the presence of bioactive compounds such as alkaloids, flavonoids, phenols, terpenoids, and saponins, which likely contribute to the insecticidal efficacy of the plant extracts. Concentration and altitude of origin were key factors affecting the extract's impact, with higher altitudes and higher concentrations consistently showing increased efficacy in beetle mortality and inhibition of seedling development in legumes, indicating an allelopathic effect.

The results showed that higher concentrations of *E. adenophorum* extract (15%) led to significant mortality in pulse beetles by the seventh day, with complete mortality observed by the tenth day. This suggests the suitability of *E. adenophorum* as a natural pesticide alternative, especially for managing pests without causing environmental toxicity or health hazards, unlike synthetic chemicals. Additionally, the differential effects on germination and radicle growth at various concentrations underscore its potential for controlled agricultural use.

Ultimately, *E. adenophorum* leaf extract offers a sustainable, non-toxic, and environmentally safe method for pulse beetle management. For severe infestations, higher concentrations of the extract could be applied for rapid control, while lower concentrations may be effective for minor pest issues. This study supports the utility of *E. adenophorum* as a viable botanical pesticide for integrated pest management practices, helping reduce reliance on synthetic chemicals and fostering sustainable agricultural practices.

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