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Bioassay on Toxicity of Plant Extracts against *Aphis craccivora* Koch (Hemiptera: Aphididae) in French Bean (*Phaseolus vulgaris* L.)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Original Research Article

ABSTRACT

The present experimental study was conducted under laboratory conditions in the department of Entomology, School of Agricultural Sciences and Rural Development, Nagaland University, Medziphema Campus during 2017 to 2018. For bioassay on the toxicity of plant extract on Aphids (*Aphis craccivora* Koch) in French beans, five botanicals were used namely, Neem (*Azadirachta indica*), Datura (*Datura stramonium*), Lantana (*Lantana camara*), Nigeria Eucalyptus (*Eucalyptus globules*) and Citronella (*Cymbopogon winterianus*) with one chemical *i.e.* Dimethoate 30 EC and

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control. The laboratory bioassay was done using dipping method and the LC 50 values were calculated using Probit analysis. The results from the Probit analysis at 24, 48 and 72 hours showed that the standard check Dimethoate 30 EC was the most toxic @ 0.01%. For botanicals, *L. camara* and *A. indica* extract @ 3% concentration, reported the highest mortality followed by *D. stramonium* extract @ 4%. In comparision *E. globules* and *C. winterianus* @ 5% reported the lowest mortality. Based on the study the order of toxicity of plant products based on Probit analysis are *A. indica* > *L. camera* > *D. stramonium* > *C. winterianus* > *E. globules*.

Keywords: French bean; bioassay; botanicals; probit analysis.

1. INTRODUCTION

French bean (Phaseolus vulgaris L.) provides one of the most important sources of protein [1, 2]. It is a great source of dietary fiber, two minerals, and vitamins [3,4]. French beans are delicate warm-season vegetables that cannot withstand frost, extreme heat, or rain. Its seed does not germinate below 15°C and the most favourable soil temperature for its seed germination ranges from 18 to 24°C. Green beans have been reported to contain 6.2 per cent protein, 0.2 per cent fat, and 63 per cent carbohydrate [5]. The crop is attacked by series insect pests during its life span. One of the major constraints in the production of French beans, is the attack of various insect pests such as hadda beetle (Epilachna vigintioctopunctata), the flea beetle (Longitarsus belgaumensis), aphid (Smynthurodes betae), and the bean fly (Ophiomvia phaseoli) which cause considerable damage [6-10]. Among them the sucking insect pests like, Aphid (Aphis craccivora Koch), leafhopper (Empoasca dolichi), thrips (Megalurothrips sjostedti Trybom), whitefly Gennadius) (Bemisia tabaci and mite (Tetranychus urticae Koch) are a common one.

In India, its cultivation is in 0.21 million ha with a production of 0.58 million MT and productivity is 2.8 t per ha [11]. In Nagaland French bean is cultivated under an area of 17280 hectares with a yield of 22140 MT [12]. Studies have shown that essential oils are readily biodegradable and less detrimental to non-target organisms as compared to synthetic pesticides [13]. A variety of properties, including toxicity to the pest, repellence, anti-feedant, and insect growth regulation activities against pests of agricultural value, are possessed by botanical pesticides. More than 2500 plant species belonging to 235 families have been found to possess the characteristics required for an ideal botanical insecticide.

2. MATERIALS AND METHODS

The present experimental study was conducted in the laboratory of the Department of Entomology, School of Agricultural Sciences and Rural Development, Nagaland University, Medziphema Campus from 2017 to 2018. The experimental site is located at Medziphema in the district of Chumukedima, Nagaland, India.

Five Botanicals were used for the experiment *viz.*, Neem (*Azadirachta indica*), Datura (*Datura stramonium*), Lantana (*Lantana camara*), Nigeria Eucalyptus (*Eucalyptus globules*) and Citronella (*Cymbopogon winterianus*) with one chemical *i.e.* Dimethoate 30 EC and control.

2.1 Plant Extraction Process

The extracts of the plant materials were prepared according to Singh [14] with modifications using an automated Soxhlet extractor (SOCS PLUS SCS04 AS DLS). Acetone was used as the solvent. Different plants used were collected from nearby local areas and then it was dried under shade for 2 to 3 weeks. The dried plants were then crushed evenly using an electric grinder. The crushed powder was sieved to obtain a fine powder.

For extraction, 20 g of plant powder was weighed and transferred into thimbles and placed in beakers. 80 ml of solvent (acetone) was added to the beakers. Then the beakers were loaded in the extractor and boiled at 80 °C for 1 hour. After that, the temperature was increased to recovery temperature at 160 °C and boiled for 30 minutes. The thimbles were rinsed 2 to 3 times. The beakers were taken out from the extractor and the thimbles were removed. After that the beakers were placed in a warm air oven at 100 °C for 20 to 30 minutes to remove the leftover acetone. The beakers were then removed and placed in a desiccator and cooled at room temperature. After extraction, the final extract was kept as a crude extract solution (100 %) in glass bottles. The crude extract was then used for testing insecticidal activities against the Aphid pests of French beans in Laboratory conditions.

2.2 Laboratory Experiments

In the laboratory, the different concentration of plant extract was evaluated for their toxicity against sucking pests of French bean. The method adopted for the experiment was as follows (Table 1).

2.2.1 Bioassay on toxicity of plant extract on Aphids (*Aphis craccivora* Koch.) by dipping method

The plant extracts emulsions of required concentrations were made by dilution with water and 1ml of triton X (0.1 %). The plant extracts were diluted to make 2, 4, 6, 8, and 10 %

solutions. For comparison Dimethoate 30 EC @ 0.03. 0.04. 0.05. 0.06 and 0.07 % was used in Table 1. 10 Adult Aphids (Aphis craccivora Koch.) was dipped for 10 seconds in each concentration with 3 replications. After that, the insects were removed, air-dried, and kept for observation in Petri dishes containing fresh French bean leaves. Observation count was taken before treatment and 24, 48, and 72 hours after treatment [15]. The mortality data were recorded for chemical and botanicals 24, 48, and 72 hours after treatment. Insects were observed regularly and those that did not move or react to mild touch were counted as dead. Insect mortality data was corrected by Abbott's formula [16]. The concentration mortality line was calculated using Probit analysis [17] in SPSS software with a log10 transformation of the concentrations. The results were expressed as concentration (%) per insect.

Table 1. Treatment details for bioassay in Aphids, Aphis craccivora Koch. (2017-18)

SI. No.	Treatments/ crude plant extracts	Part used	Concentration (%)
1	Neem (Azadirachta indica)	Leaf	2,4,6,8,10
2	Datura (Datura stramonium)	Leaf	2,4,6,8,10
3	na (Lantana camara)	Leaf	2,4,6,8,10
4	Nigeria Eucalyptus (Eucalyptus globules)	Leaf	2,4,6,8,10
5	itronella (Cymbopogon winterianus)	Leaf	2,4,6,8,10
6	Dimethoate 30 EC / Rogor	-	0.03,0.04,0.05,0.06,0.07
7	Control (water)	-	-

Table 2. Mortality of Aphid (Aphis craccivora) at 24, 48 and 72 hours with plant extracts treatment

Plant extracts	Concentration	Insect mortality rate (%)		
	(%)	24 HAT	48HAT	72HAT
1. Eucalyptus globules	2	10	20	40
	4	16.67	43.33	53.33
	6	26.67	56.67	63.33
	8	40	60	70
	10	60	83.33	83.33
2. Lantana camara	2	36.67	36.67	60
	4	46.67	56.67	63.33
	6	60.00	70	73.3
	8	76.67	80	86.67
	10	83.33	86.67	100
3.Cymbopogon winterianus	2	10	20	36.67
	4	20	33.33	60
	6	30	46.67	63.33
	8	40	50	73.3
	10	66.67	73.33	83.3
.Azadirachta indica	2	6.67	26.67	46.67
	4	26.67	40	66.67
	6	30.00	63.33	86.67
	8	60.00	66.67	90
	10	63.33	83.33	96.67

Plant extracts	Concentration	Insect mortality rate (%)		
	(%)	24 HAT	48HAT	72HAT
5.Datura stramonium	2	40	56.67	93.33
	4	43.33	70	100
	6	63.33	76.67	100
	8	86.67	86.67	100
	10	86.67	96.67	100
6.Dimethoate 30 EC	0.03	43.33	83.33	86.67
	0.04	83.33	86.67	93.33
	0.05	96.67	100	100
	0.06	100	100	100
	0.07	100	100	100

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*HAT: Hours after Treatment

Corrected percent mortality = Percent mortality in treatment – Percent mortality in control / 100 - Per cent mortality in control x 100%

Table 3. Probit analysis for toxicity at 24, 48 and 72 hours of plant extracts against Aphid, Aphis craccivora

Name of extract	LC50 (%)	95% fiducial limit	Slope ± SE	Goodness of fit chi squared
A. At 24 hours			-	
Eucalyptus	10	6.992-72.465	2.48±0.99	0.62
Lantana	4	0.984-6.808	1.94±0.77	0.02
Citronella	9	6.506-36.286	2.57±0.98	0.81
Neem	8	5.879-16.416	2.96±1.02	0.68
Datura	4	1.205-5.726	2.05±0.77	1.49
Dimethoate	0.01	0.031-0.02	6.53±2.04	0.72
B. At 48 hours				
Eucalyptus	6	6.021-3.878	2.36±0.84	0.53
Lantana	4	1.099-6.205	1.94±0.77	0.02
Citronella	8	5.146-39.221	2.05±0.84	0.53
Neem	5	5.466-3.187	2.20±0.80	0.39
Datura	1(NS)	-	1.58±0.77	0.40
Dimethoate	0.02(NS)	-	2.46±1.76	0.35
C. At 72 hours				
Eucalyptus	5	0.272-26.679	1.53±0.76	0.20
Lantana	3	0.000-4.765	1.52±0.76	1.19
Citronella	5	0.890-11.604	1.64±0.76	0.18
Neem	3	0.381-4.616	1.94±0.78	0.16
Datura	0(NS)	-	0.43±0.90	0.08
Dimethoate	0(NS)	-	1.60±1.66	0.11

*NS: Non significant

3. RESULTS AND DISCUSSION

3.1 Bioassay on Toxicity of Plant Extract on Aphids (*Aphis craccivora* Koch) by Dipping Method

In the present study from Table 2, the mortality at 24, 48, and 72 hours due to direct toxicity of plant extracts, *D. stramonium*, *L. camara* and Dimethoate 30 EC at different concentrations shows variations in per cent mortality ranging from 0 to 100 %. From the details presented in

Table 3, *L. camara* and *A. indica* extract @ 3% concentration reported the highest mortality followed by *D. stramonium* extract @ 4% at 24 hours after treatment. While *E. globules* and *C. winterianus* @ 5% reported the lowest mortality at 72 hours after treatment. The results show that the per cent mortality increases with the increase and time after treatment.

Based on the per cent mortality, the concentration mortality line was calculated using Probit analysis. The details of the Probit analysis for 24, 48, and 72 hours are presented in Table

3. The results from the Probit analysis at 24, 48. and 72 hours showed that the standard check Dimethoate 30 EC was the most toxic @ 0.01%. For the plant products, at 24 hours D. stramonium and L. camara were the most toxic at low concentrations followed by A. indica. The LC 50 values at 24 hours were E. globules 10%, L. camara 4%, C. winterianus 9%, A. indica 8 %, D. stramonium 4%, and Dimethoate 30 EC 0.01 %. Comparable results were obtained 48 hours after treatment where D. stramonium showed the lowest concentration mortality value among botanicals @ 1% (NS), where significant results could not be found. Therefore at 48 hours after treatment, the most toxic at the lowest concentration was observed in *L. camara* @ 4%. The LC 50 values for 48 hours after treatment were L. camara 4%, A. indica 5%, E. globules 6%, C. winterianus 8% and D. stramonium 1% (NS). However, at 72 hours, L. camara and A. indica was the most toxic at the lowest LC50 value followed by C. winterianus and E. globules. While in D. stramonium and Dimethoate 30 EC meaningful result could not be found. The LC50 values at 72 hours were E. globules 5%, L. camara 3%, C. winterianus 5%, A. indica 3%, D. stramonium 0% (Non-significant) and Dimethoate 30 EC 0% (Non-significant). Based on the study the order of toxicity of plant products based on Probit analysis was A. indica > L. camara > D. stramonium > C. winterianus > E. globules.

4. CONCLUSION

On bioassay of Aphids (*Aphis craccivora*) the most effective treatment was observed in Dimethoate 30 EC / Rogor (0.01%) followed by Botanicals Neem (3%) and Lantana (3%) followed by Datura (4%) and the least effective treatment was seen in Eucalyptus (5%) and Citronella (5%). Based on the study the order of toxicity of plant products based on Probit analysis was *A. indica* > *L. camara* > *D. stramonium* > *C. winterianus* > *E. globules.*

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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