



TOXICITY OF AQUEOUS EXTRACT OF NEEM (*AZADIRACHTA INDICA* A. JUSS) SEEDS ON GIANT POD SUCKING BUG (*ANOPLOCNEMISCURVIPES*) OF COWPEA (*VIGNAUNGUICULATA*(L.) WALP).

Kiran Singh

Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto, Nigeria.

*Correspondence to: Dr. Kiran Singh, Department of Biological Sciences, Usmanu Danfodiyo University, Sokoto, PMB 2346, Nigeria, West Africa.

Abstract: Cowpea (*Vigna unguiculata*) is the major source of protein supplement in Nigerian diet, which is attacked by many insects pests in farm and storage conditions. Studies were carried out to determine the efficacy of water extract of neem (*Azadirachta indica* A. juss) kernel against *Anoplocnemiscurvip*es adults and nymphs. Neem kernel extract at 5%, 10% and 15% w/v were evaluated using 0% as control under field conditions. The extracts were sprayed once weekly for three (3) weeks. The extract reduced the population of insects in both nymphs and adults stages. 10% and 15% of extract were found more effective than the 5% for both the stages of insect. Toxicity of neem kernel was time and dose dependent on tested insects. Nymphs of *A. curvipes* were more susceptible than the adults. After third spray of 10% and 15% of neem extract showed 100% reduction in nymph population of *A. curvipes*, while same spray caused 66.67% reduction in adult population of *A. curvipes*.

Introduction: Cowpea, *Vigna unguiculata* (L.) Walp, is cheapest source of plant protein in West Africa. This crop is known to be attacked by insect pests from the trifoliate stage to harvesting. A species complex of hemipteran pod pests called the pod-sucking bugs (PSB) attack its pods and seeds. Pod-sucking bugs leave feeding punctures that are not easily detectable but which are responsible for the premature shrivelling of young pods and the

occurrence of half-filled seeds in older pods (Singh and Jackai., 1985; Jackai et al., 1989). The giant pod sucking bug *Anoplocnemiscurvip*es is one of the most serious pests of leguminous crops in this part of world. In a study it was observed that presence of two *A. curvipes* (adults) per ten plants caused 51.7% pod loss, 24.3% seed damage and 21.5% reduction in cowpea yield (Olufemi and Odebyie, 2001). The pod-sucking bugs have also been reported to migrate into cowpea fields from a wide range of leguminous shrubs and trees, which serve as maintenance hosts (Singh *et al.*, 1985; Jackai *et al.*, 1989). After initial adult migration, the population of the bugs multiplies

For Correspondence:

ksinghATgmail.com

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in cowpea fields, though this depends on a number of ecological and agronomic factors such as climatic conditions, susceptibility to pests and diseases.

It is now realized that the use of synthetic pesticides are posing serious environmental hazards. Plant products have played an important role as herbal traditional medicine for treatment of human ailment and also served other roles including protection of crop against pests and disease vectors in Africa (Delobel and Trans, 1993). Plants are known to produce a range of secondary metabolites which possess multiple mode of action including acute toxicity, repellency, antifeedent, and inhibition of oviposition, growth, development and reproduction (Uko and Kamalu, 2001). Large numbers of plant families possess pesticidal properties (Oparaekete *al.* 2006). It has been observed that oil of neem (*Azadirachta indica*) possess lethal property against molluscs (Singh and Singh 1997). The active moiety responsible for insecticidal activity in neem is azadirachtin. In the present study we have evaluated the toxicity of aqueous extract of neem against *A. curvipes* with broader objective to control different species of cowpea pests.

Materials and Methods

The investigations were conducted under rain fed conditions in UsmanuDanfodiyo University, Sokoto, Nigeria at Dry Land Research Farm. Treatments consists of 5%, 10% and 15% of neem kernel extract w/v. Neem fruits were collected locally, depulped, dried and crushed. 250gm. 500gm. and 750gm. of crushed neem seeds were soaked in plastic buckets containing 3L. of water, stirred and kept overnight and labelled as 5%, 10% and 15% of neem extract respectively. Each extract was filtered with an additional of 2 L. of water, using muslin cloth making the total valum of extract to 5 L.

The field layout was a randomized complete block design (RCBD). Plot size was 6.0m × 5.0m and each plot had five (5) ridges (three inner rides and two discarded). The ridges were spaced 0.75m apart. There were three treatments and one untreated control. All replicated three times. The seed of cowpea variety Ba'dare (a local variety of cowpea which attracts most of the pests) was used for the experiment and 3-4 seeds were sown per hole, using the dibbling method of sowing. The seedlings were thinned to two plants per planting hill two to three weeks after sowing. A compound fertilizer (NPK 15:15:15) was top-dressed at the rate of 37.5 kg/ha.

Field application of neem extract commenced at 7 (seven) weeks after planting. This was the time of flower bud formation and onset of flowers in the cowpea variety used. Spraying was done between 8.00 a .m to 10.00 a. m each day using a pressurized low-volume knapsack (CP 3) at the discharge rate of 150L/ha. A single row of cowpea was sprayed per pass or trip. All the concentrations of spray were applied once every week for three weeks and if there was rain within two hours of application, spraying was repeated the next day.

Anoplocnemis curvipes, adults and nymphs were counted and sampled separately, before each spraying from 6.30 to 8.30 a. m. and 24 hours after the spraying of neem seed extract counting of insects was repeated and recorded. All the data were subjected to Analysis of Variance (ANOVA) and means were separated using LSD at 5% treatment level (SAS), 1989.

Results

Table 1 showed that the treatments (5%) did not reduce insect count significantly after first spray, but there was 72.73% reduction in the nymph population after first spray of 10% and 15% of neem extract. Counting after second spray

showed that there is significant reduction in the population of *A. curvipes* nymph treated with 5%, 10% and 15% of aqueous extract of neem seed, but there were no difference between sprayed plots. Treatments 10% and 15% showed similar effect against nymphs. After third spray

100% reduction in *A. curvipes* nymph population, but there is significant difference between the 10% and 15% treatments as compared to 5%, as well as there was significant difference between 5% treatment and the control.

Table 1: population of *A. curvipes* (nymph) as affected by neem seed extracts on cowpea, after first, second and third spray.

Treatments (conc. %)	Mean population (survival) of <i>A. curvipes</i> (nymph) after		
	1 st spray	2 nd spray	3 rd spray
0%	3.667 (100%)	3.667 a (100%)	4.000 a (100%)
5%	2.333 (63.62%) +	2.000 b (54.54%) +	1.667 b (41.68%) +
10%	1.000 (27.27%) +	1.333 b (36.27%) +	0.000 c (0.0%) +
15%	1.000 (27.27%) +	1.000 b (27.27%) +	0.000 c (0.0%) +
SE	0.9	0.4	0.9
Sig.	NS	*	*

Means in a column followed by the same letters are not significantly different using LSD at 5%.

NS: not significant

*: Significant

+ : Survival (%) of *A. curvipes*(nymphs) after spraying of neem seed extract.

Table 2 showed that no significant difference between 0%, 5% and 10% was observed, but there was a significant reduction in *A. curvipes* population at 15% treatment level after first spray. No significant difference after second spray, but there was a significant difference in

the number of *A. curvipes* at 10% and 15% treatment level after third spray as compared to control. There was 66.67% reduction in adult of *A. curvipes* population. No significant difference was observed between 0% and 5% treatment level even after third spray.

Table 2: population of *A. curvipes* (adults) as affected by neem seed extract on cowpea, after first, second and third spray.

Treatments (conc. %)	Mean population (survival) of <i>A. curvipes</i> (adult) after		
	1 st spray	2 nd spray	3 rd spray
0%	2.667 a (100%)	3.000 (100%)	3.000 a (100%)
5%	2.000 a b (74.99%)+	2.667 (88.9%)+	2.333 a b (77.67%)+
10%	1.000 a b (37.50%)+	1.667 (55.57%)+	1.000 b (33.33%)+
15%	1.609 b (60.33%)+	1.333 (44.33%)+	1.000 b (33.33%)+
SE	0.4	0.5	0.5
Sig.	*	NS	*

Means in a column followed by the same letters are not significantly different using LSD at 5%.

NS: not significant

*: Significant

+: Survival (%) of *A. curvipes*(adults) after spraying of neem seed extract

Discussion

The results clearly demonstrate that aqueous extract of *A. indica* seeds has potential insecticidal properties against *A. curvipes* nymph and adults. The nymphs of *A. curvipes* were more susceptible than the adults. Table 1 and 2 show significant reduction in the number of insect both nymph and adults. After spray the number of insects were higher in 0% where no neem extract was applied, while higher reduction in insects population was obtained with 10% and 15% neem seed extract treatments. Reduction in adults and nymphs of *A. curvipes* population may be due to toxicity of azadirachtin and salanin which are the chief active components present in neem (Schmutterer and Ascher, 1989).

Azadirachtin inhibits the release of prothoracicotropic hormones, allatotropins, affecting metamorphosis in insects (Schmutterer and Rembold, 1995; Bunken and Stark, 1997). Azadirachtin caused significant inhibition in many vital enzymes in the snail's body like acetylcholinesterase, alkaline phosphatase and lactic dehydrogenase activity (Singh and Singh, 2000). It seems that inhibition of same enzymes in the insect body can cause

death in *A. curvipes* and contribute in reduction of population of insects.

A part from the toxic action, reduction in the *A. curvipes* population by neem seed extract can be due to its antifeedent activity (Warthen et al., 1978; Mordue and Nisbet, 2000). Azadirachtin cause inability to ingest food, resulting to primary antifeedant at the sensory level (Schmutterer, 1985). Antifeedency is correlated with the sensory response of chemoreceptor and insect mouthparts. The feeding behaviour of these insects depends on their neural input, oral cavity and the combination of this sensory code by the nervous system. Azadirachtin stimulates the chemoreceptor inhibiting the firing of sugar cells, which stimulates feeding. This results in the starvation and death of insects by prevention of feeding (Mordue et al., 1998). The secondary antifeedancy is targeted in the gut and results to food consumption reduction (Schmutterer, 1985). Secondary antifeedency is as a result of an imbalance in hormone and other physiological system such as stomatogastric nervous system, inhibition of digestive enzymes production, movement of food through the gut (Koul and Isman, 1991; Timmins and Reynolds, 1992; Trumm and Dorn, 2000). These factors

may cause reduction in the insect population or swarming of insects as there were no insects after third week spray of neem seed extracts. Higher reduction in the insects, treated with 10% and 15% extract, is due to the high concentration of azadirachtin and other active ingredients in the neem seed extract, which can cause mortality and /or antifeedancy in insects. Higher reduction in the *A. curvipes* nymph population after spray of neem seed extract as compared to adults may be due to their relatively soft body integument, which allow the extract to penetrate through the insect integument and reach to the site of action, or the nymphs have a less developed mixed function oxidases system (MFOs, which is responsible for detoxification of drugs in the insect body) than the adult *A. curvipes* have, which cause a rapid metabolism and / or higher toxicity of azadirachtin and salanin which are the chief active components present in neem (Schmutterer and Ascher, 1989).

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