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Research Article
Influence of Moringa and Balanite Seed-Oil on Fecundity, Hatchability and Duration of Developmental Stages of *Helicoverpa armigera* Hub. on Tomato Plant

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Abstract

Background and Objective: *Helicoverpa armigera* is among the major insect pest of tomato fruit that threatens its availability in the right quality. The management of *H. armigera* has over the years relied heavily on the use of synthetic insecticides which have negative consequences. This study therefore, aimed at examining the sub-lethal effect of Moringa and Balanite seed oils on *H. armigera* third instar larvae. **Materials and Methods:** An experiment was conducted at the Entomology Laboratory of the Department of Crop Protection, Bayero University, Kano, to test the toxicity and sub-lethal effect of three concentrations: 2, 4 and 6% of Balanite and Moringa seed oils on the third instar larvae of *H. armigera*. **Results:** The result shows that the seed oils reduced the duration of the developmental stages of *H. armigera* third instar larvae. Significant (50, 85 and 100%) reduction in the larval (most damaging stage) duration, fecundity and hatchability were observed, respectively in 6% concentration compared to the control. **Conclusion:** This underpins the sub-lethal effects of the plant oils on the third instar larvae of *H. armigera*.

Key words: *Helicoverpa*, third instar larvae, tomato, moringa, balanite, seed oils

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Tomato production is affected negatively by numerous pests which result in poor yield, low quality and financial loss. Ahmed *et al.*[1] reported 200 species of insect pest attacking tomatoes worldwide. The major pest is reported to includes aphids (*Aphis gossypii*), whiteflies (*Bemisia tabaci*), thrips (*Thrips tabaci*), leaf miners *(Liriomyza* spp.) and (*Tuta absoluta*) and tomato fruit worms (*Helicoverpa armigera*)[2,3].

Helicoverpa armigera is one of the most serious insect pests worldwide and is particularly widely distributed in Asia, Europe, Africa and Australasia. The larvae are the destructive stage, they prefer to feed and develop on reproductive structures but generally, they preferentially feed on bud, flowers and fruit which are rich in nitrogen[4]. However, first instar larvae feed on leaves, flowers and buds while the rest of the larval instars feed on fruit, they usually bore clean circular holes which serve as entry points for secondary infection by diseases causing organisms. During its developmental stages, larvae may emerge from one fruit and enter another to continue feeding and losses caused may range from 20-60%, with global annual crop losses about 5 billion dollars[5].

Leaf damage reduces leaf area, which can slow plant growth. Feeding on flowers and fruit causes the main damage. On whole plants, *H. armigera* will initially start feeding on tender leaves but eventually, their movements on plants will primarily take them to the reproductive organs[4]. The neonates clear a patch of hair by grazing before they begin to establish a feeding site on the leaf surface[6]. Flower feeding can prevent fruit formation.

Over the years, conventional synthetic insecticides have been successfully utilized to control pests and boost crop production. However, its negative consequences on both human and associated environmental health concerns as well as its tendencies to induce resistance, resurgence and secondary pest outbreaks in treated areas currently limit their utilization[7]. This scenario has triggered research towards alternative and eco-friendly methods of controlling insect pests on fruit crops like tomato. Botanical is at the forefront as a method of choice among the alternative to synthetic insecticides. The use of oils from plants is one of the botanical gaining grounds for use as a safer method of pest management strategy in organic production of particularly horticultural crops like tomatoes. Plant oils from Balanite and Moringa have shown some level of control on many insect pests. However, their efficacy in the control of *H. armigera* has not been investigated. This study aimed at examining the sub-lethal effect of Moringa and Balanite seed oils on *H. armigera* third instar larvae.

MATERIALS AND METHODS

Study area: The experiment was conducted from September to November, 2019 at the Entomology Laboratory of the Department of Crop Protection, Faculty of Agriculture, Bayero University Kano which is located at latitude: 11.98°N, longitude: 8.42°E.

Sourcing and collection of *H. armigera*: Fifty *H. armigera* late instar larvae were collected from a pesticide-free tomato farm in the study area. The larvae were transported to the Entomology Laboratory of the Department of Crop Protection, Bayero University, Kano, in a transparent plastic box measuring 25×20 and 8 cm deep line with tissue paper and covered with the muslin cloth.

Rearing of *H. armigera*: Larvae of *H. armigera* were collected from infested tomato plants in the field. The collected larvae were reared in groups of 10 larvae in a transparent plastic cup measuring 8.5 cm diameter at the top, 6 cm diameter at the bottom and 5 cm deep line with tissue paper and covered with muslin fabric. The larvae were artificially fed with young tomato leaves and immature tomato fruits. These food materials were changed and replaced with fresh ones daily. The larvae were allowed to pupate in the same containers. After emergence, 50 adults of mixed sexes were transferred to a cage measuring 45×45 cm for mass breeding to obtain a homogenous and sufficient population for the experiment. Cotton wool soaked with 10% honey solution was provided as food and muslin cloths were hanged inside the cages for oviposition. Similarly, a small fresh branch of tomato plant carrying young leaves and flowers were dipped in a bottle containing water to keep it turgid was provided to mimic their natural environment. These branches were renewed daily to ensure the availability of fresh stuff.

Muslin cloth containing eggs were removed and replaced with new ones. The eggs on muslin cloth were kept moist in the transparent plastic box of 25×25 and 8 cm deep with a perforated lid till they hatch. Neonates of *H. armigera* (first instar larvae) were collected with a camel hair brush and introduced onto young leaves and young fruits of tomato in a transparent plastic cup of 8.5 cm diameter at the top, 6 cm diameter at the bottom and 5cm deep lined with moist filter papers. Feeding and changing of food materials were continued up to the third instar larvae needed for the experiment.

Preparation of plant materials: Seeds of desert date (*Balanites aegyptiaca* Del.) and Moringa (*Moringa oleifera*) were purchased from the Kurmi market in Kano. The epicarp

(outer cover) and mesocarp (pulp) of Balanites and Moringa were removed, washed in tap water and allowed to dry and then grounded into powder using Comec (CM 290) micro grinder.

Extraction of essential oils by hydro distillation method: Fifty grams of grounded Balanites and moringa seeds (seed powder) were placed separately into 2 L round bottom flasks and distilled water was added into the round bottom flasks until all the seed powder were completely immersed. The flasks were then heated in a Clevenger-type apparatus heater and the mixture was boiled for 3 hours to evaporate the components in the seed powder. The vapours were condensed by the cold water circulating in the condenser. The condensed mixture of essential oils and water were collected in a separating funnel. After about 20 min the oil extract was separated from the water by the separating funnel and collected separately. The essential oils obtained were stored in a refrigerator at 4°C to avoid degradation of components until used.

Preparation of oil concentration and bioassays: Balanite and Moringa seed oil were prepared into three different concentrations viz: 2, 4 and 6% in acetone. Fruit and leaves of the tomato plant were sprayed with the different concentrations (2, 4 and 6%) of the two plant oils and acetone was used as the control. Thereafter, the treated fruit and leaves were placed in a plastic cup measuring 8.5 cm diameter at the top, 6 cm diameter at the bottom and 5 cm deep, lined with tissue paper and covered with muslin cloth and laid out in a complete randomized designed replicated 3 times. Ten 3rd instar larvae of *H. armigera* were introduced into the container and allowed to feed for 24 hrs on treated tomato plant material. Then, the treated food materials were replaced with untreated ones and survivals of *H. armigera* larvae were observed daily to determine the larval duration, pupation and emergence.

After emergence, three adults in the ratio 1:2 female to males of emerged adults were placed together in a plastic jar measuring 37 cm in diameter and 20 cm deep to maximize successful mating and a piece of cotton soaked in 10% honey solution was provided as a source of food. Internally, the container was covered with a soft sheet of tissue paper for oviposition. Also, the mating of adult males and females which resulted from feeding the larvae on untreated leaves were used as a control. To determine the fertility of eggs (hatchability percentage of eggs), two or three patches of eggs having not less than 100 eggs were collected during the first 3 days of oviposition and incubated under the laboratory conditions until hatching.

Data analysis: All Data obtained on fertility, hatchability, larval duration, pupation and emergence were subjected to Analysis of Variance Using Computer Software (Genstat Discovery Edition) and means with a significant difference were separated using tukey test at a 5% level of probability.

RESULTS

Effect of plant part, control materials and concentration on adult emergence, larval and pupal duration of *H. armigera* third instar larvae: The effect of Moringa and Balanites seed essential oils on the larval and pupal duration of *H. armigera* third instar larvae and emergence of the adult is shown in Table 1. The result indicates that there was a highly significant difference among plant parts, control materials and concentration on adult emergence, larval and pupal durations. Significantly shorter larval duration was recorded on tomato leaves compared to fruit. Balanites seed oil gave a significantly shorter larval duration compared to Moringa seed oil.

Based on the concentration of oils used, 6% concentrations give the shortest larval duration but were not different statistically from the durations recorded in 2 and 4% concentrations. As expected, the 0% concentration recorded the longest larval duration but was not different statistically from the durations recorded in 2 and 4% concentrations.

With regards to the pupal duration and adult emergence, a similar trend to that of larval duration was observed but the number of adults who emerged on tomato leave was not different statistically from the number of adults who emerged on tomato fruit. Similarly, the number of adults who emerged on Balanite was not different statistically from the number of adults who emerged on Moringa. When concentration is been considered, significantly high number of adults emerged in 0% concentration but was not different statistically from the number of adults emerged in 2% concentration.

Interactions of plant parts and concentrations on larval duration of *H. armigera* third instars larvae: Table 2 showed the interactions effect of plant parts and concentrations on larval duration of *H. armigera* third instar larvae. Significantly shorter larval during was observed on fruits treated with 6% concentration but was not different statistically from the durations obtained in leaves treated with 6, 4 and 2% concentration. And longer larval duration was obtained on leaves treated with 0% concentration but this duration was not different statistically from the duration obtained in fruits treated with 0, 2 and 4% concentration.

Table 1: Effect of plant part, control materials and concentration on adult emergence, larval and pupal duration of *H. armigera* third instar larvae

Treatments	Larvae (days)	Pupae (days)	Emergence
Plant parts			
Leaves	5.1[a]	7.9[a]	0.7[a]
Fruits	10.3[b]	15.3[b]	0.8[a]
SE±	3.97	5.37	0.55
Control materials			
Moringa	8.1[a]	9.0[a]	0.7[a]
Balanites	7.3[b]	14.3[b]	0.8[a]
SE±	3.97	5.37	0.55
Concentrations			
2%	7.4[ab]	13.3[ab]	0.7[ab]
4%	6.9[ab]	8.6[a]	0.6[a]
6%	5.3[a]	8.0[a]	0.5[a]
0%	11.2[b]	16.6[ab]	1.3[b]
SE±	3.97	5.37	0.47
Interactions			
Pp *Control materials	NS	NS	NS
Pp *Concentration	0.28	NS	NS
Control materials *Concentration	0.26	0.001	NS
Pp *Control materials *Concentration	NS	NS	NS

Mean followed by different letters in same column are significantly different at p = 0.05 level of probability according to Tukey test and Pp: Plant parts

Table 2: Effect of interaction between plant parts and control materials on larval duration of *H. armigera* third instars larvae

	Plant parts	
Control materials	Fruits	Leaves
Moringa	5.75[ab]	4.38[a]
Balanites	8.85[bc]	11.7[c]
LS	*	

Mean followed by different letters in the same column are significantly different at p = 0.05 level of probability according to Tukey test and LS: Level of significance

Table 3: Effect of interaction between control material and concentration on larval duration of *H. armigera* third instars larvae

	Biopesticide	
Concentrations	Moringa	Balanites
2%	6.25[a]	8.33[ab]
4%	6.17[a]	7.95[ab]
6%	5.50[a]	4.42[a]
0%	14.42[b]	8.50[ab]
LS	*	

Mean followed by different letters in the same column are significantly different at p = 0.05 level of probability according to Tukey test and LS: Level of significance

Table 4: Effect of interactions between control materials and concentrations on pupal duration of *H. armigera*

	Control materials	
Concentrations	Moringa	Balanites
2%	12.75[bc]	17.17[bc]
4%	7.92[ab]	13.75[bc]
6%	0.00[a]	8.00[ab]
0%	15.35[bc]	18.08[c]
LS	*	

Mean followed by different letters in the same column are significantly different at p = 0.05 level of probability according to Tukey test and LS: Level of significance

Interaction of control material and concentration on larval duration of *H. Armigera* third instars larvae: The interaction effect of control material and concentrations is shown in Table 3. Significantly high larval during was observed in Moringa in untreated control but is different statistically with other concentrations (Table 3). Similarly, when Balanites is been considered the untreated control has a significant high larval duration compared to the other concentrations. The least larval duration was observed in 6% concentration which is not different from 2 and 4% concentration in both Moringa and Balanites.

Interactions of control materials and concentrations on pupal duration of *H. Armigera*: Table 4 shows the effect of interaction between control materials and concentrations on the pupal duration of *H. armigera*. The result indicates that interaction of Balanites and 0% concentration gives the longest pupal duration compared to the remaining interactions but this duration was not different statistically from the durations obtained in the interaction of Balanites with 2 and 4% concentration and also with the interaction of Moringa with 0 and 2% concentration. The least pupal duration was observed in the interaction of Moringa and 6% concentration which is not different statistically from the pupal duration obtained in the interaction of Moringa and 4% concentration and interaction of Balanites with 0%.

Effect of plant part, control materials and concentration on fecundity and hatchability of *H. armigera* eggs: Table 5 shows the effect of Moringa and Balanites seed oils on the fecundity and hatchability of *H. armigera* eggs. The result

Table 5: Effect of plant part, control materials and concentration on fecundity and hatchability of *H. armigera* eggs

Treatments	Fecundity (%)	Hatchability (%)
Plant parts		
Leaves	38.0	26
Fruits	52.1	30
SE±	50.9	67
Control Materials		
Moringa	35.5	21
Balanites	54.1	35
SE±	50.9	67
Concentrations		
2%	25.4^{a}	12.08^{ab}
4%	33.3^{a}	8.83^{ab}
6%	13.58^{a}	0.00^{a}
0%	107.9^{b}	90.83^{b}
SE±	50.9	67
Interactions	NS	NS

Mean followed by different letters in the same column are significantly different at p = 0.05 level of probability according to Tukey test

reveals that there is no significant difference among plants parts and control materials on the fecundity of *H. armigera* adults but when concentrations are being considered significant difference was observed between the different concentrations and the control.

When fecundity is been considered, significantly high number of eggs were laid by *H. armigera* feed with plant parts treated with 0% concentration compared to the remaining concentrations. Likewise in hatchability, a similar trend was observed but the number of *H. armigera* eggs hatched that are laid by *H. aramigera* feed with plant parts treated with 0% concentration was not different statistically from the number of *H. armigera* eggs hatched that are laid by *H. aramigera* feed with plant parts treated with 2 and 4% concentration.

DISCUSSION

The result of this study shows that life stages and emergence of *H. armigera* adult, was short on tomato leaves compared to tomato fruits, this may due to differences in nutrients and chemical constituents of the plant parts. Tomato leaves contain several bioactive components which play a major role in host plant resistance. Ferreres *et al*.[8] reported that tomato leaves contain steroidic, alkaloids and phenolics such as hydroxycinnamic acids and flavonoids, which are known to be involved in host-plant defence.

Balanites gives significantly low larval, pupal duration and adult emergence compared to Moringa. This indicates that active constituent's present in Balanites essential oils seed affect life stages of *H. armigera*, because shorter larval duration may reduce damage. 9-octadecenoic acid methyl ester which is a fatty acid ester is among the major constituent of Balanites seed oils. Fatty acid esters were reported by Dalia[9] to have reduced pupation, egg production, hatchability and sterility. Ramos-Lopez *et al*.[10] reported that certain fatty acid esters have insecticidal properties.

Baskar *et al*.[11] in their study of bioefficacy of leaves and roots of *Aristolochia tagala* against *Spodoptera litura* report that the extract has larvicidal, pupicidal and antifeedant properties. They further report that the extract affects larval and pupal durations. A report by Chinnamani *et al*.[12] shows that selected Indian medicinal plants extract have ovicidal and larvicidal properties against *S. litura*. High larvical activities of *A. indica* against *H. armigera* larvae were reported by Wondafrash *et al*.[13]. Kavitha *et al*.[14] observed that *Argemone mexicana* extract reduced adult emergence and increase pupal mortality of *S. litura*. A report by Choudhary *et al*.[15] indicates that *Verticillium lecanii* is effective against *H. armigera* larvae.

Also, the result of these studies indicates that increases in the concentration of control material shorting the duration of the developmental stages (larval and pupal) and reduce the emergence of adults and the number of eggs laid. This may be due to the increased concentration of the bio-active constituent of essential oils as the concentrations increased. Habib *et al*.[16] report a decrease in the pupal rate of *Plutella xylostella* due to an increase in *Peganum harmala* seed extract. Wondafrash *et al*.[13] reported that *M. ferruginea* seeds and stem bark extract at 5% were effective in reducing egg lying of *H. armigera* Metah *et al*.[17] also reported that neem seed kernel extract at 5% is more effective in reducing the larval population of *H. armigera* than lower concentrations.

With regards to interactions, tomato fruits treated with Moringa seed oil reduces larval and pupal duration, the emergence of an adult than other control materials. This may be attributed to the presence of a bioactive constituent of unripe fruits of tomato, combine with the bioactive constituent of Moringa essential oil. David *et al*.[18] reported that unripe tomato fruits contain 9-octadecenoic acid methyl ester, hexadecanoic acid methyl ester and docosanoic acid methyl ester. These bioactive compounds accumulated in plant tissues and are reported to have insecticidal properties[19,20].

CONCLUSION

The present study found that the two control materials evaluated have sub-lethal effects on *H. armigera* third instar larvae by shortening developmental (larval and pupal) durations and suppressing adult emergence, fecundity and hatchability of *H. armigera*. It is recommended that a

2% concentration of Balanites seed oil should be used in managing *H. armigera* on tomato plant.

SIGNIFICANCE STATEMENT

This study discovered the effect of Moringa and Balanites seed oils in suppressing adult emergence, fecundity and hatchability of *H. armigera* and reduction in the duration of its most damaging (third instar larvae) developmental stage, that can be beneficial for the management of *H. armigera*. This study will help researchers to uncover a new input in designing an IPM programme for *H. armigera.*

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