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Tadele Shiberu and Emanu Getu

Full Length Research Paper

Effects of crude extracts of medicinal plants in the management of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) under laboratory and glasshouse conditions in Ethiopia

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Tomato leaf miner, *Tuta absoluta* (Meyrick) is a devastating pest of tomato originating from South America which has been recorded in Ethiopia throughout tomato growing areas since 2012. The larvae damages the above ground parts of the plant especially leaf and fruits from seedling stage to maturity. Studies were conducted between 2015 and 2016 for two seasons to screen medicinal plants for the management of *T. absoluta* under laboratory and glasshouse conditions. Crude extracts of *Phytolacca dodecandra* seed and leaf, *Allium sativum*, *Nicotiana* species, *Cymbopogon citrates* and *Azadirachta indica* seed were evaluated at 5, 7.5, and 10% concentrations in the laboratory. Treatments which were found to be the best in the management of *T. absoluta* under laboratory condition were further evaluated for their efficacy under glass house condition. The mean percentage mortality of larva was recorded for 120 h after treatment application under laboratory study. The highest larval mortalities of 98.33, 96.67 and 95% were recorded for *A. indica*, *C. citrates* and *A. sativum* all at 10% concentration, respectively. In glasshouse experiment, *A. indica* gave the highest larval mortality of 66.54 % and the lowest percent mortality was recorded in *P. dodecandra* leaf and seed in both under laboratory and glasshouse conditions. With the obtained results, it can be concluded that foliar application of the mentioned medicinal plants extract on tomato plants reduced *T. absoluta* population and improved the quality and quantity of tomato fruit yield.

Key words: Medicinal plant, *Tuta absoluta*, *Lycopersicon esculentum*, crude extracts, efficacy, concentration.

INTRODUCTION

Tomato (*Lycopersicon esculentum* Mill.) belongs to the nightshade family, Solanaceae. It is one of the most

important vegetable crops in the world, and widely grown crops in Ethiopia (Lemma, 2002). It can be eaten either

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either fresh or processed into different products and helpful in healing wounds because of antibiotic properties found in ripe fruits and also a good source of vitamins A, B and C (Baloch, 1994).

Tomato production faces many problems from several factors which lead to significant yield loss. Among these factors, insect pests are the most important. *Tuta absoluta* causes a very high level of damage both in terms of quantity and quality to tomato (Garcia-Mari and Vercher, 2010; Guedes and Picanço, 2012; Megido et al., 2012), mainly if control measures are not undertaken (Desneux et al., 2010). Damage is caused by the larvae that mine leaves and bore into fruits which eventually facilitate plant pathogen invasion (EPPO, 2005). *T. absoluta* has recently invaded Europe, Africa and Asia (Desneux et al., 2011), being presently found in 63 countries of the world. It can affect tomato from seedling to fruit maturity stages. Feeding is caused by all larval stages throughout the plant growth period. On leaves, the larvae feed inside, forming irregular leaf mines which may later become dead tissue and serve as avenue for the entry of pathogens. The larvae can form extensive mines in the stems and tunnel into fruits, forming galleries which alter the normal growth of the plants and qualities of the fruits.

The first affected tomato plantation was detected in Ethiopia in 2012 (Gashawbeza and Abiy, 2012). Recently, the pest has been detected throughout the country and it may become a significant problem in greenhouses as well as in open fields. An immediate consequence of the introduction of *T. absoluta* is illustrated by the sudden increase in insecticide use in tomato fields, going from 8 to 12 applications per one season. It requires insecticide applications in every five days intervals which are needed in Dandi district West Shewa of central Ethiopia (Personal communication). However, due to the development of resistance, chemical control has shown limited efficacy even after using different types of pesticides and increasing the application frequencies (Liatti et al., 2005). Therefore, the objective of this study was the screening of different medicinal plants at appropriate doses against *T. absoluta* under laboratory and glasshouse conditions.

MATERIALS AND METHODS

Description of the study area

The research was conducted under laboratory and glasshouse conditions at Ambo University glasshouse and plant science laboratory. Ambo is at the geographical coordinate of 8°59'N latitude and 37.85°E longitude with an altitude of 2100 masl (Briggs, 2012). The temperature of the laboratory during the study period was 24±2°C and glasshouse temperature was 32±2°C.

Botanical collection and preparation

Fresh fruits and leaves of *Phytolacca dodecandra*, leaves of

Nicotiana species, and *Cymbopogon citratus* were collected and simultaneously separated from any infestation of disease and insect pest and then washed and cut into small pieces. *Allium sativum* was obtained from a local market in Ambo and used as fresh extract. *A. sativum* extraction was prepared according to the method described by Stoll (2000) using the following items: 250 g of garlic fresh bulbs were chopped and strained in grinder, then the chopped bulbs were soaked in 1 L of distilled water for 1 h. *C. citratus* extraction was also prepared according to Stoll (2000) as follows: dried leaves of lemon grass were powdered and strained. Fifty grams of powdered dried leaves were soaked in 2 L of distilled water for 6 h. All the botanicals were ground, mixed, strained and filtered through cheese cloth and made stock solution. The stalk solution mixed with water at 5, 7.5 and 10% concentration level (v/v) in 100 ml of water (Table 1).

Experiments under laboratory conditions

The experiment was conducted for 120 h under laboratory condition at room temperature of 24±2°C. Larvae were inserted in a Petri dish having 9 cm diameter within the leaves and provided with coated cotton moist that is kept as fresh leaves of tomato that was collected from the glasshouse. All medicinal plants were prepared in three doses (50, 75, and 100 ml/L) from prepared stalk solution as follows: Endod fruit crude water extract (*P. dodecandra*), Endod leaf (*P. dodecandra*), garlic clove (*A. sativum*), tobacco leaves (*Nicotiana* spp.), local variety, lemon grass leaf (*C. citratus*) and Neem seed (*Azadirachta indica*). Medicinal plant crude extracts were sprayed on *T. absoluta* larvae in the Petri dish using micro pipette. For the control treatment, larvae were treated with distilled water. After 24, 48, 72 and 120 h of exposure, the mortality rates at different concentrations were obtained. Dead larvae were removed as soon as possible in order to prevent decomposition which may cause rapid death of the remaining larvae.

Experiments under glasshouse condition

Seeds of tomato cultivar 'Coshoro' were planted on November 1st in the nursery site. The plants were transplanted into the experimental glasshouse after 40 days. The experimental pots were 20 diameters and 25 cm height. Totally, 21 pots were prepared and filled with compost, loam soil and sand soil in the ratio of 1:1:2, respectively. The distances between the plants were 40 cm and between the pots were 100 cm.

The experiment was laid in a completely randomized block design (RCBD) with three replications. Purposefully, the door and windows of the glasshouse was kept open for 24 h for the entrance of moths. Hence, plants were infested under natural conditions. Planted tomato plants were sprayed with medicinal plant insecticides at recommended doses from laboratory studies. Each potted plant was covered with nylon cloth to avoid escape of larvae. Mortality data was recorded after treatment application of 1st, 3rd, 5th and 7th days. The best promising medicinal plant extracts were selected for further field experiment.

Data collection

The glasshouse experiment was scouted every week for the signs and symptoms of *T. absoluta* occurrence until infestation of *T. absoluta* appeared. Before treatment application, pre-spray counts were made and then treatment application follows. Treatments were applied at vegetative, flowering and fruit setting stages of the crop. After treatment application, data on *T. absoluta* larva both alive and dead (per plant) were recorded at 1, 3, 5 and 7 days

Table 1. List of some crude extracts of medicinal plants used against Tomato leaf miner, *Tuta absoluta* under laboratory and glasshouse condition.

Common name	Amharic name	Scientific name	Family	Used part (s)
Soap berry	Endod	<i>Phytolacca dodecandra</i>	Phytolacaceae	Leaf and seed
Tobacco	Timbaho	<i>Nicotiana</i> species	Solanaceae	Leaf and stalk
Lemon grass	Keysar	<i>Cymbopogon citratus</i>	Gramineae	Leaf
Garlic	Nich shinkurti	<i>Allium sativum</i>	Liliaceae	Cloves
Neem	Yekinin zaf	<i>Azadirachta indica</i>	Meliaceae	Seed

intervals.

Statistical analysis

Data were subjected to analysis using SAS version 9.1 (SAS, 2009). The mean percent mortality was corrected using Abbott's formula (Abbott, 1925) and efficacy analysis was done based on data transformation to Arcsine $\sqrt{x+0.5}$ when necessary according to Gomez and Gomez (1984).

$$CM (\%) = \frac{[T - C]}{[100 - C]} \times 100$$

where CM (%) is corrected mortality, T is mortality in treated insects, and C is mortality in untreated insects.

RESULTS

Effect of botanicals on larvae of *T. absoluta* under laboratory conditions

Efficacy of medicinal plant extracts against *T. absoluta* in the laboratory is shown in Table 2. All treatments are significantly ($P < 0.01$) different on plant treated with various treatments and reduced the total numbers of *T. absoluta* larvae per treatment. Effect of medicinal plant crude extracts was evaluated against *T. absoluta* at three different concentrations (5, 7.5 and 10%) after four exposure time (24, 48, 72 and 120 h). Table 2 shows medicinal plant extract at 10% concentration. *A. indica* seed had maximum toxicity (98.33%) against *T. absoluta* after *C. citratus* (96.67%), while *P. dodecandra* leaf showed the minimum mortality (55.0%) after 120 h of application. On the other hand at 7.5% concentration, biocidal potential of *C. citratus* was high (91.67%), followed by *A. indica* seed (90.0%) and *A. sativum* (86.67%). Low toxicological effect was observed by some extracts at 5% concentration especially *P. dodecandra* leaf and seed showed least mortality (36.67%). Similarly, significant time interval for exposure of plant extracts was recorded at all concentrations as shown in Table 2. Medicinal plant extract did not show quick response after 24 h but the effect was significantly high after 72 and 120

h as more larvae of *T. absoluta* were killed.

Effect of medicinal plant on larvae of *T. absoluta* under glasshouse conditions

Under glasshouse conditions, toxicity of medicinal plant crude extracts was evaluated against *T. absoluta* at 10% concentrations after five exposure dates (1, 3, 5 and 7 days). The tested medicinal plant crude extracts at 10% concentration illustrated in Table 3 showed that significant ($P < 0.01$) differences were observed among the treatments from the untreated control. The presented data pertaining to mean percent reduction of *T. absoluta* larvae reveals that *A. indica* seed had maximum toxicity 66.54 (54.92%) against *T. absoluta* after which *Nicotiana* spp. showed 62.10 (52.58%), while *P. dodecandra* seed showed significantly lowest effect mortality 36.51 (36.84%) for the entire days. In general, the results indicated that each medicinal plant extract caused significant mortality rate against larvae of *T. absoluta* after 120 h exposure in the laboratory and after 7 days in the glasshouse. Therefore, these findings suggest that medicinal plant insecticides are a good alternative management option of *T. absoluta*.

DISCUSSION

T. absoluta are capable of reducing the total yield of tomato plants as well as facilitate it to secondary attack by pathogens. The application of botanicals on *T. absoluta* larvae resulted in good performance. These results were confirmed with those reported by Ghanim and Ghani (2014); they reported that the highest reduction of *T. absoluta* population was obtained after tomato plants treated with *A. sativum* and *C. citratus* extract. They showed the highest effects of *A. sativum* on *T. absoluta* second instar larvae under laboratory conditions. *C. citratus* extract reduced the population of *T. absoluta* on plant moderately as compared with *A. sativum*, *A. indica* and *Nicotiana* spp. The results obtained were in agreement with the work of Hussein et al. (2014); they found that the aqua extract of *C. citratus* and *A. sativum* showed good insecticidal activity and can

Table 2. Mean efficacy of some medicinal plant extracts against *T. absoluta* larvae at different rates and time of exposure in the laboratory.

S/N	Treatments	Conc. (%)	Mean efficacy percentage			
			After 24 h	After 48 h	After 72 h	After 120 h
1	Endod seed (<i>P. dodecandra</i>)	5	21.67 ^{gh}	33.33 ^{ef}	35.00 ^{hi}	40.0 ^h
		7.5	26.67 ^{fg}	36.67 ^{def}	38.33 ^h	43.33 ^h
		10	31.67 ^{ef}	36.67 ^{def}	53.33 ^{ef}	56.67 ^{fg}
2	Endod leaf (<i>P. dodecandra</i>)	5	23.33 ^h	26.67 ^g	28.33 ^j	36.67 ^h
		7.5	23.33 ^{gh}	26.67 ^g	33.33 ^{ij}	36.67 ^h
		10	38.33 ^{fg}	31.67 ^{fg}	51.67 ^{fg}	55.0 ^g
3	Garlic cloves (<i>A. sativum</i>)	5	38.33 ^d	38.33 ^{de}	46.67 ^g	66.67 ^{ef}
		7.5	38.33 ^d	53.33 ^b	63.33 ^{bc}	86.67 ^{bc}
		10	56.67 ^b	66.67 ^a	73.33 ^a	95.0 ^{ab}
4	Tobacco (<i>Nicotiana</i> species)	5	38.33 ^d	56.67 ^a	56.67 ^{def}	61.67 ^{efg}
		7.5	46.67 ^c	58.33 ^a	61.67 ^{cde}	70.0 ^{de}
		10	66.67 ^a	71.67 ^a	71.67 ^a	80.0 ^{cd}
5	Lemongrass (<i>C. citratus</i>)	5	33.33 ^{de}	41.67 ^{cd}	53.33 ^{ef}	88.33 ^{abc}
		7.5	36.67 ^{de}	46.67 ^c	56.67 ^{def}	91.67 ^{ab}
		10	38.33 ^d	56.67 ^b	63.33 ^{bc}	96.67 ^{ab}
6	Neem seed (<i>A. indica</i>)	5	46.67 ^c	46.67 ^c	58.33 ^{cde}	90.0 ^{abc}
		7.5	66.67 ^a	68.33 ^a	68.33 ^{ab}	90.0 ^{abc}
		10	66.67 ^a	68.33 ^a	71.67 ^a	98.33 ^a
7	Control	-	0.00 ⁱ	0.00 ^h	0.00 ^k	6.67 ⁱ
-	LSD	-	3.73	3.81	3.74	10.81
-	CV (%)	-	4.54	4.14	3.73	7.17
-	MSE ±	-	1.68	1.72	1.69	4.87

Means with the same letter(s) in rows are not significantly different for each other. All treatment effects were highly significant at $p < 0.01$ (DMRT).

Table 3. Mean efficacy of some medicinal plant extracts against *T. absoluta* larvae at different rates and time of exposure under glasshouse.

S/N	Treatments	Mean efficacy percentage			
		After 1 day	After 3 day	After 5 day	After 7 day
001	<i>P. dodecandra</i> seed	18.65 (25.43) ^d	24.03 (29.15) ^c	36.51 (37.14) ^{bc}	36.51 (36.84) ^b
002	<i>P. dodecandra</i> leaf	23.33 (28.86) ^{cd}	26.11 (30.61) ^c	32.72 (34.83) ^c	36.94 (37.28) ^b
003	<i>A. sativum</i> clove	43.45 (41.22) ^a	58.59 (43.95) ^a	58.93 (50.15) ^a	59.92 (50.60) ^a
004	<i>Nicotiana</i> species	33.61 (35.41) ^{ab}	45.71 (42.59) ^{ab}	45.83 (42.59) ^{abc}	62.10 (52.58) ^a
005	<i>C. citratus</i>	30.63 (33.58) ^{bc}	33.97 (35.60) ^{abc}	47.62 (43.63) ^{ab}	57.94 (49.61) ^a
006	<i>A. indica</i> seed	24.52 (29.76) ^{bcd}	46.79 (42.88) ^a	50.95 (45.55) ^a	66.54 (54.92) ^a
007	Control	0.00 (0.00) ^e	0.00 (0.00) ^d	0.00 (0.00) ^d	0.00 (0.00) ^c
-	LSD	6.17	12.0	7.98	11.64
-	CV (%)	8.91	14.98	8.83	11.59
-	SE±	2.47	4.81	3.20	4.67

Means with the same letter(s) in rows are not significantly different for each other. All treatment effects were highly significant at $p < 0.01$ (DMRT). Figures in parentheses are Arcsine percent transformed value.

be used to control *T. absoluta* in the laboratory and greenhouse. This finding is also in concordance with the report of Gonçalves-Gervásio and Vendramim (2007); they found that *A. indica* seeds' extracts made high larval mortality of *T. absoluta* under laboratory condition. Oparaeke (2007) verified that *A. indica* and *A. sativum* extracts contain insecticidal properties that are lethal to a wide range of insects. The results revealed that *A. sativum*, *Nicotiana* spp. and *C. citratus* gave promising results to minimize the impact of *T. absoluta* larvae on leaves and fruit's, similarly to the standard treatment of *A. indica* seed.

CONCLUSION AND RECOMMENDATIONS

The current experiments suggested that *A. indica*, *Nicotiana* spp., *A. sativum* and *C. citratus* showed potential effect on larvae of *T. absoluta* after the 7th day of application. Consequently, from the obtained results, it can be concluded that foliar application of the mentioned medicinal plants extract on tomato plants reduced *T. absoluta* population and improved the quality and quantity of tomato fruit yield. Therefore, further investigation is also needed to determine the active ingredients of residual effect and frequency of application against larvae of *T. absoluta*.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests

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