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RESEARCH ARTICLE

The effects of alkaloid extracted of *Solanum nigrum* leaves on the biological performance of *Dialerodus citri*

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Abstract

In laboratory bioassays to determine the toxicity of alkaloid extracts of *Solanum nigrum* leaves to whitefly *Dialerodus citri* . alkaloids applied at concentrations of 0.1 , 0. 2 , 0.5 , 1 % . The larva was generally the more susceptible stage to all test treatments.The results indicated that the concentration of 1 % was the most effective accumulative mortality of *D. citri* which treated with concentration mention earlier. Mortality rate reached to 51.05 , 58.69 , 62.31 and 67.15% at concentration of 1% in larvae , nymphs , pupae and adults respectively . Development time of immature stages of *D. citri* also , affected by the application of alkaloid extracts of *S. nigrium* leaves , generally development period prolonged in all treatments of alkaloids as compared with control treatment . As well as two compounds were separated by using TLC technique and compound no. 1 was more effected as compared with compound no.2

Introduction

Today almost 60% of the synthetic pesticides are derived from plant sources , many of them have effects on the different systems of the body (Perry , 1980) . One of the largest groups of chemicals arsenals produced by plants are the alkaloids , It can occur naturally in any part of the plant, including the leaves, fruit, and tubers (Wang *et al*.,2006) glycoalkaloids are a poisons found in species of the nightshade family (Solanaceae) such as *S. nigrum* plant (Jisha *et al*., 2011 ; Bai et al., 2012) , many of the gluco alkaloids such as solanine has fungicidal and pesticidal properties, and solanine hydrochloride (a salt of solanine) has been used as a commercial pesticide, as well as it is one of the plant's natural defenses (Alexander *et al*., 1948) .

Whitefly *D. citri* become a problem when they continually sap the plant of energy needed for growth. This occurs when there is constant flushing and availability of new growth, such as after severe hedging and topping (Fasulo and Brooks, 2010) . Citrus is the most important host, but the following are also food plants --- allamanda, banana shrub, cape jasmine, chinaberry, laurel cherry, crape myrtle, coffee and others (IIE , 1996).

Material and Methods

Alkaloid extraction from *S. nigrium* leaves was done according to Harborn (1973) . Fresh leaves of plant were collected and washed to remove the dust . The green leaves were dried at room temperature for two weeks and ground to fine size (powder) with a mill and stored in plastic containers at 10°C (Haikal and Omer , 1993) .The samples were extracted by maceration with 5% acetic acid , then was filtered through filter paper to remove cellular debris . The extract was transferred to electrical oven at 70 °C and then cooling and add Con. Ammonium hydroxide drop wise until the pH is10 . Then it was transferred to centrifuge and discard the supernatant by filtration .Wash the precipitate with 1% Ammonium hydroxide and recentrifuge and refiltration , the extract was dried by rotary evaporator . In the end the extract was purified by dissolving in boiling methanol , filtering and concentration until the alkaloid starts crystallizing . Concentration of 0.1 , 0.25 , 0.5 , 1 and 0.0 % of alkaloid extract were prepared by dissolving one gm of dried alkaloid extraction in 5ml methanol , the volume completed to 100 ml by using distilled water . Thin Layer Chromatography (TLC) technique on silica gel was used to separate the chemical compounds by using acetic acid – ethanol (1:3) , Dragendorff reagent for general alkaloids and

Marquis reagent for glucoalkaloids were used to detect the compounds by UV (Harborn , 1973 ; Mohammed *et al.* , 2009) , then spots were crushed and dissolved in same solvent (methanol) by magnetic stirrer and the filtered through filter paper , the extract was dried by rotary evaporator . Concentration of 0.5% was prepared from compound 1 (Rf = 45) and compound 2 (Rf = 57) .

D . citri adults were collected from the citrus plants (Diyala region) and kept in a cage containing young *Citrus aurantium* L. as host plant , after an ovipositional period of one day the adults were removed . The egg- bearing leaves on plant were incubated at room temperature (23 ± 3 C°) . The effects of alkaloids extract with their different concentrations were tested against eggs , larval stage (mobile stage) , nymphal stages (immobile stages) pupae and adults of *D . citri* by taking 50 individuals from each stage (three replicates were made for each treatment) . Mortality rates were recorded and corrected to Abbots formula . To calculate the developmental period , fifteen newly hatched larvae (five each replicates) that lived from previous treatment with concentration that mentioned before , The developmental period (days) from larval stage to nymphal stage was counted and the mean of each was calculated . All the above steps were used to calculate the developmental period (days) of pupae . As well as cumulative effects of alkaloids were calculated , all the steps mentioned above were applied except for eggs which were left after treatment . Mortality rates of eggs, larva , nymphs , pupae and adults were recorder and corrected according to Abbots formula . The effects of alkaloid compounds 1 and 2 on the mortality of *D. citri* were tested , Mortality rates were recorded and corrected to Abbots formula . The Statistical Analysis System- SAS (2010) was used to effect of difference extract in study parameters. Least significant difference-LSD and Duncan multiple range test was used to compare between means in this study.

Result and Discussion

Table 1 shows that alkaloids of *S.nigrum* leaves were affected different stages mortality of *D. citri* , also the data showed a direct correlation between mortality and extract concentrations , significant difference were found among the concentrations of $P < 0.05$. Larval stage was more susceptible , the mortality rate range between 22.86 – 87.85% at concentration of 0.1 – 1.0 % respectively . Concentration of 1.0 % is the best treatment of all stages , mortality rate reached 87.85 , 63.31 , 51.75 , 50.0 and 38.46% in larvae , adults , nymphs , pupae and eggs respectively .Rathi (2013) found that the hexane extract were less effective than methanolic extract at different concentrations of *Schanginia aegyptiaca* against different stage of *D. citri* , and extracts were more effective in larval stage than other stages . Zhang and Kubo (1993) ; Blackford and Dinan (1997) mentioned that the toxic extracts inhibit growth and development of many species of insects because it interfere with molting hormone and converted to physiologically inactive ecdysteroids . The extracts were more effective in larval stage (crawlers) than nymphal stage , this may attributed to the crawlers usually move a few centimeter in search of a feeding site (exposed to toxic extracts more than nympal stage) while ,the nymphal stage is immobile . Also , eggs mortality may be due to embryo asphyxia inside the egg because the extract was formed as layer on the external shell Sexena *et al*. (1980). Adults mortality may be attributed either to an indirect effect of strong deterrence causing energy depletion or dehydration Veierov (1996) ,or to direct toxicity of the crude extract to very susceptible adults Metacalf *et al*. (1951).

Study results indicated that alkaloids of *S.nigrum* leaves significantly affect accumulative mortality ($P < 0.05$) . Figure 1 shows the percentages of accumulative mortality of *D. citri* which treated with concentration mention earlier. Mortality rate reached to 51.05 , 58.69 , 62.31 and 67.15% at concentration of 1% in larvae , nymphs , pupae and adults respectively , El- Shafie and Basedow (2003)found that the cumulative mortality reached 78 % and 61 % in nymphal stages of *Aphis gossypi* and *B. tabaci* respectively when treated with Neem Azal – T/S® while it was 57.1% and 52.4 % when treated with neem oil .

The development period of larvae , nymphs , pupae and adults was significantly increased ($P< 0.05$) as the concentration of alkaloid increased (Table 2) . The developmental time increased from about 3.4 , 7.2 and 4.8 days in control treatment to about 6.1 , 13.5 and 9.3 when treated at concentration of 1% of crude alkaloid respectively. Al – Mansour (1995) mentioned that the development period of white fly *B. tabaci* generally prolonged in all treatment of alkaloid extracts as compared with control treatment . The increasing developmental period of immature stages that treated with extracts may be attributed to decreased larval efficiency of food conversion which affected negatively on growth and increased developmental period , or due to the interference with the action of endocrine system . Al – Sharook and Girjee (1993) .

Study results indicated that the two compounds of the TLC technique are alkaloids (Rf1 = 45 , Rf2 = 57) due to their positively reaction (orange colure) with Dragendroff reagent (general test of alkaloids) , as well as compounds 1 and 2 are glucoalkaloids due to their positively reaction (yellow colure) with Marquse reagent (test of glucoalkaloids) . Table 3 shows that compound no. 1 was very toxic as compared with compound no.2 at concentration of 0.5% and Larval stage was more susceptible .

Table 1: The effects of alkaloid extracts of *S. nigrum* leaves on the mortality of different developmental stages of *D. citri*

Extract con. (%)	Eggs mort. (%)	larval mort.(%)	Nymphal mort. (%)	Pupal Mort. (%)	Adults Mort. (%)
Control	4.66 d	6.66 e	4.66 d	2.66 d	6.00 d
0.1	10.48 c	22.86 d	22.38 c	19.86 c	28.36 c
0.25	20.97 b	40.00 c	30.07 c	29.45 b	36.87 bc
0.5	23.77 b	68.56 b	40.00 b	36.30 b	44.68 b
1.0	38.46 a	87.85 a	51.75 a	50.00 a	63.31 a
LSD Value	7.54 *	12.09 *	8.53 *	8.02 *	11.74 *

* ($P<0.05$).
Means having different letters at the same column are significant different.

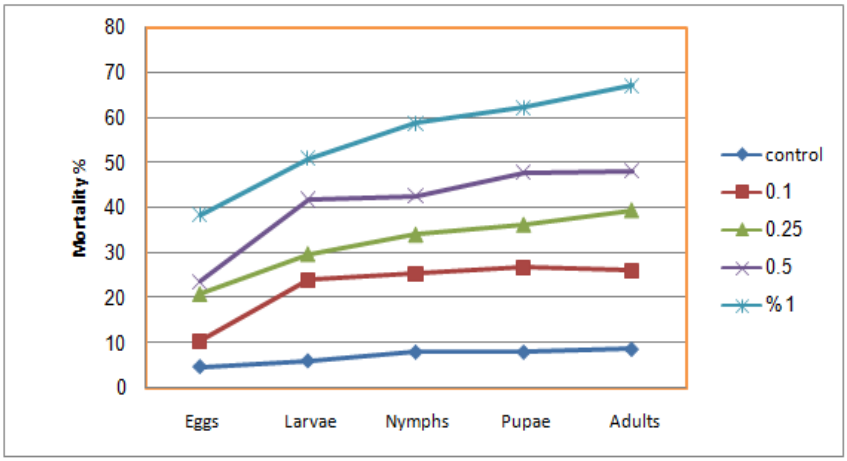

Fig.1: The effects alkaloid extracts of *S. nigrum* on the cumulative mortality of *D. citri*

Table 2: The effects of alkaloid extracts of *S.nigrum* on the developmental period of immature stages of *D. citri*

Extract conc. (%)	Developmental period (days)		
	Larvae	Nymphs	Pupae
Control	3.4 b	7.2 c	4.8 b
0.1	3.7 bc	8.9 c	6.5 b
0.25	5.1ab	9.4 bc	7.4 ab
0.5	5.4 a	12.1 ab	8.3 a
1	6.1 a	13.5 a	9.3 a
LSD Value	1.75 *	3.69 *	2.85 *

* ($P<0.05$).
Means having different letters at the same column are significant different.

Table 3: The effects of separated alkaloids compounds of *S.nigrum* leaves on the mortality of different developmental stages of *D. citri*

Extract 0.5%	Eggs mort. (%)	larval mort.(%)	Nymphal mort. (%)	Pupal Mort. (%)	Adults Mort. (%)
Control	3.33 b	4.0 c	2.0 c	2.0 c	1.33 c
Comp.1 (Rf=45)	35.86 a	66.66 a	34.01 a	39.44 a	39.19 a
Comp.2 (Rf = 57)	11.76 b	29.85 b	19.04 b	17.0 b	24.32 b
LSD Value	10.56 *	14.82 *	12.09 *	14.53 *	12.69 *

* ($P<0.05$).
Means having different letters at the same column are significant different.

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