

Biological Studies On Cotton Leafworm (*Spodoptera Littoralis*) And Evaluation The Efficacy Of Certain New Means For Its Control

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The studies were conducted to evaluate the bioactivity of certain plant extracts against the eggs, 2nd and 4th instar larvae and pupae of cotton leafworm *S. littoralis*. Extractions by two solvents (petroleum ether and acetone) from four plants (Radish, White Mustard, Poinciana seeds and Cloves flowering buds) were used to study their bioactivity on the cotton leafworm. Also two insecticides (lannate and Dipel 2x) were tested after dispersing the required amount of the formulated insecticides in water. The cotton leafworm larvae were obtained from the laboratory colony maintained on castor plant leaves. The host plants used in this experiment included cotton, berseem, castor-oil, soybean and maize. Isolation, identification and bioassay processes were performed on the constituents of Clove flowering buds extracts, fennel, (*Syzygium aromaticum*) They were subjected to further analytical investigations for identifying their chemical and structural formula using GC/MS and GLC. A field experiment was conducted to evaluate the efficacy of the extracts (petroleum ether and acetone) of cloves flowering buds, essential oils of clove, Dipel 2x, W.P. based on *B.t. subsp. kurstaki* (32x103) potency per mg.) and Lannate 90 %W.P. alone/or combined with Lannate against *S. littoralis* larvae. Bio-residual activity of certain plant extracts, insecticides and their combinations under field conditions. Also, the effect of spraying insecticides and bio agents on chlorophyll content of Tomato leaves and yield were determined. The major results of the present study could be summarized as follows: - Out of 8 different crude extracts with regard to antifeedant effects against 4th instar larvae of the cotton leafworm larvae. Acetone extract of Cloves flowering buds and Poinciana seeds were recorded as the highest antifeedant activity. While Cloves flowering buds gave the highest antifeedant activity with petroleum ether extracts. From biological activity test Radish and White Mustard seeds acetone extracts were the most effective against 4th instar larvae of *S. littoralis* induced higher larval mortality means. While Cloves petroleum ether extract was the most effective on larval mortality. The longest larval duration mean were obtained by Cloves flowering buds and Radish seeds acetone extracts. While Cloves flowering buds and Radish seeds petroleum ether extract showed the highest larval duration. The highest mortality percentages of pupae was recorded by White Mustard seeds acetone extract. While zero pupal mortality was recorded for Radish seeds acetone extract but the highest mortality percentage was recorded by Cloves flowering buds, White Mustard and Radish seeds petroleum ether extracts. The higher pupal durations were recorded for the acetone extract of Cloves flowering buds, Radish and Poinciana seeds, compared with control. While, the duration of pupae for White Mustard seeds of pupae for White Mustard seeds acetone extract was similar to that of control. The pupal duration recorded for the petroleum ether extracts of different plants were similar to that of control. All tested plant acetone and petroleum ether extracts gave normal pupae except white Mustard and Poinciana seeds acetone extracts which gave malformed pupae. The lowest mean percentage of successful adult formation was recorded by White Mustard seeds acetone extracts but higher percentages adult emergence resulted with Cloves flowering buds and Poinciana seeds acetone extracts. While the lowest percentage of formation resulted after treatment with Cloves flowering buds and White Mustard seeds petroleum ether extracts. The highest percentage of adult emergence resulted by

Radish seeds and Poinciana seeds petroleum ether extracts. Concerning adult sex ratio, the male ratio was generally greater than female one by Radish seeds, Cloves flowering buds and Poinciana seeds acetone extracts and also White Mustard and Poinciana seeds petroleum ether extracts. The female was greater than male one, by White Mustard seeds acetone extracts and untreated and also Radish seeds petroleum ether extracts. The highest number of eggs was recorded by Cloves flowering buds acetone extract and also White Mustard seeds petroleum ether extract. While Radish seeds acetone extract and Cloves flowering buds petroleum ether extract expressed the lowest number of deposition. Summary 209 White Mustard seeds acetone extract and Cloves flowering buds petroleum ether extract gave the lowest egg hatchability %. While, The highest hatchability % was recorded by the control. The higher sterility value detected by acetone extracts of White Mustard, Radish and Poinciana seeds and Cloves flowering buds petroleum ether extract. Results indicated that Cloves and Radish seeds extracts were the most effective extracts against the 2nd larval instar of *S. littoralis* followed by the extract of white Mustard. While, Poinciana seeds extract was the least effective one under study. This result indicate that the petroleum ether extracts of Cloves and Poinciana seeds were the most effective against 2nd instar larvae of *S. littoralis* followed by Radish extract, while white Mustard petroleum ether extract was the least effective one. Date indicated clearly that Cloves extract was the most effective one followed by Radish seeds extract, while the acetone extracts of Poinciana seeds and white Mustard seeds were the least effective extracts against 4th instar of *S. littoralis*. This result indicated that petroleum ether extract of Cloves flowering buds was only the effective extract against 4th instar larvae of *S. littoralis*. While the other three plant extracts indicated lower mortality values. The probit analysis obtained by the exposure of 2nd instar larvae of *S. littoralis* to different concentrations of four plant extracts. Cloves flowering buds in petroleum ether was the most effective one recording the lowest value (LC50 0.18) followed by the Summary 210 same plant but with acetone having the value of LC50 (0.2) and then Radish seeds with acetone having the value of LC50 (0.7). The two extracts of cloves flowering buds were the most effective against 4th instar larvae of *S. littoralis* having LC50 values of 1.6 and 3.3 % for petroleum ether and acetone extracts, respectively. The LC50 values of acetone extract of Radish seeds was 7.6 %. Data of extracts indicate that newly laid eggs were more affected by the tested extracts compared with older ones. Poinciana seeds acetone extracts gave the lower means of eggs hatchability percentage of *S. littoralis* on newly laid 24 hrs, 48 hrs and 72 hrs. While White Mustard seeds and Poinciana seeds petroleum ether extracts had the lower hatchability percentage treated a newly laid and treated after 24 hrs., respectively. However, The White Mustard seeds and Radish seeds acetone extracts recorded the second (Moderately) hatchability percentages of *S. littoralis* eggs when treated newly laid and eggs 24 hrs old. However, the Cloves flowering buds acetone extracts recorded the highest means for eggs hatchability % of 48 hrs and 72 hrs old. The data obtained from eggs 48 hrs and 72 hrs old, White Mustard seeds gave the second effects (moderately) on hatchability percentages, while, both of Radish seeds and Cloves flowering buds recorded the highest ones. Radish seeds and Cloves flowering buds petroleum ether extracts recorded the highest hatchability percentage when treated eggs newly laid and after 24 hrs. For eggs of 48 and 72 hrs old, the Poinciana seeds petroleum ether extracts gave the lower hatchability percentage Summary 211 The biological activity of different plants extracts on *S. littoralis* after dipping the pupae for 5 seconds in each plant extract concentration. the highest used concentration (10%). Cloves flowering buds acetone extract gave the highest pupal mortality%, (100%) followed by Poinciana seeds extract (93%), Cloves flowering buds and Poinciana seeds petroleum ether extracts gave the highest percentage of pupal mortality %. While the lowest mortality values was recorded for White Mustard seeds acetone extract (53.6%) and Radish seeds and Whit Mustard petroleum ether extracts gave the lowest mortality. pupal duration were increased after the treatments of Radish seeds, White Mustard seeds and Cloves flowering buds acetone extracts Cloves flowering buds and Poinciana seeds petroleum ether extracts. On the other hand, the Poinciana seeds acetone extract and Radish seeds and White Mustard seeds petroleum ether extracts reduced significantly the duration periods of pupae compared with other plants extracts and the control. The lowest percentage of adult formaton resulted at the highest used concentration (10%). all tested plans: extracts in acetone showed lower percentage of adult emergence in

comparison to control. Various treatments of acetone extracts on the adult longevities indicated no significant effect. But the longevities of *S. littoralis* male and female adults were 9.00 female and 8.67 male days for control. Various treatments of petroleum ether extracts were significantly shortened of longevity. Summary 212 At the highest used plant concentration (10%), egg-number was zero for Poinciana and Cloves acetone extracts, while it was 695, 748 and 1967 for Radish, White Mustard acetone extract and control. And also 10% concentration of all plant extract and 5% concentration of both plant extracts Cloves flowering buds and Poinciana seeds petroleum ether extracts recorded (0.0) number of egg per female. All concentrations of Poinciana seeds acetone extracts and 10% concentration of both plants Cloves and Radish seeds acetone extracts exhibited zero hatchability %. However, the 1.25 % conc. of Radish seeds acetone extract and untreated gave the highest hatchability % of eggs laid by females. concentration 10% of all plant extracts and 5% concentrations of both of cloves and Poinciana petroleum ether extracts had zero % of hatchability. However the untreated gave the highest hatchability % of eggs laid by females but without superiority than those obtained by 1.25% concentration of all plant extracts except Radish seeds petroleum ether extracts. The highest rates of sterility above 90% were obtained with adult females treated as pupae with all concentration of Poinciana seeds, gave 100% sterility followed by 10 and 5 conc. of Radish seeds and 10% conc. of White Mustard seeds gave 98.08, 91.69 and 90.76, respectively. When the Cloves flowering buds extracts was used at the concentration of 1.25 it caused 2.25% sterility at treated females. 100% sterility was recorded at 10% concentrations of different plant extracts in petroleum ether. While, lower sterility values were obtained at the lowest tested plant concentration. Summary 213 post-treatment, the recorded mortality of 2nd and 4th larval instar caused by lannate treatment at 50ppm were around 63% and 60% for 2nd and 4th instar larvae, respectively. The effect of lannate in acetone on fourth instar larvae was relatively similar to effect of lannate in water. The highest used concentration (one gram/ 100 ml) increased from around 3% to 67% for 2nd larval instar and from 2% to 62% for the 4th instar larvae by increasing the treatment period from one to 7 day. LC50 values of Dipel2x were 0.39 and 0.69 g/ 100 ml for 2nd and 4th instar larvae. In case of lannate these values were 10.1 and 18.1ppm causing 50% bill for 2nd and 4th instar larvae. Lannate in acetone showed lower LC50 value (10.3ppm). For 4th instar larvae of *S. littoralis*. Data indicated that increasing lannate and di pel2x concentrations increased mortality percentages of larvae. lannate did not give any significant effect on the durations of larvae. But, all Dipel2x concentrations exhibited longer larval and pupae duration when compared with untreated. While, insignificant effect of lannate sublethal concentrations was shown on percentages of malformed pupae. The results showed that percentages of adult emergence and sex ratio resulted from the treatments of lannate sublethal concentrations were similar to control. adult deformation percentage and this rate increased with increasing lannate concentrations. In addition, the untreated (control) did not show any adult malformation. no. of eggs/ female and hatchability % were decreased. with increasing concentrations of lannate and Dipel:Zx. Female sterility Summary 214 percentage increased by increasing lannate and Dipel2x concentrations. While, the effects of dipel2x on duration of pupae emergence rates of adults. longevity and sex ratio was insignificant. Generally, the hatchability percentage of treated newly laid and after 24, 48 and 72 hrs decreased significantly by increasing lannate concentrations from 5 to 25ppm and it was significantly lower than control. Also, mean reduction rate of eggs hatchability obviously higher at high concentrations of lannate (25 and 15ppm). And also dipel2x concentrations from 0.125 to 1 g mean eggs hatchability was higher at the highest tested concentration of Dipel2x (1g/ 100ml). The increasing lannate conc. increased mortality percentage from 6.9 at 5ppm to 41.4 at 50ppm. conc. had significant pupal mortality effect percentage compared with control except at 5ppm and increasing dipel 2x conc. increased mortality percentage. The pupal period were obtained due to dipping *S. littoralis* pupae for five second in lannate and dipel 2x treatments compared with control. The lowest value of emergence percentage was detected at all concentration of lannate except 5ppm of lannate no insignificant difference in adult emergence and the control was detected. The decrease in adult % emergence was recorded with rising the concentration of dipel2x from 0.125g to 1g /100 ml. The lowest value of emergence percentage was detected at the highest tested concentration of Dipel 2x (1g / 100ml). The higher malformed percentage was obtained at 50ppm

followed by 15ppm. On the other side, the lowest value was obtained by 10ppm. No malformation was recorded in case of Dipel 2x and the control pupae. Summary 215 The highest reduction in no. of eggs and hatchability was obtained at higher concentrations. Also, higher concentrations of lannate and Dipel 2x gave the highest sterility percentage, while, sterility percentage decreased by decreasing lannate and Dipel concentrations. All the mixtures of the lannate and acetone plant extracts produced additive effect with exception of Poinciana seeds at 5 and 1.25% concentrations which produced antagonistic effect. The effect of five host plants (i.e. Castor oil, cotton, berseem, maize and soybean) was studied. Larvae did not able to complete its life cycle when feed on maize and soybean. This result indicated clearly that Castor oil leaves was the most suitable food for rearing *S. littoralis* larvae followed by Berseem and Cotton leaves. Lower duration means of eggs, larvae and pupae in the second generation was retarded by castor oil leaves. Results showed that larvae fed on castor oil leaves and cotton gave the highest pupation percentage, adult emergence%, number of eggs per female, hatchability percentage, weight of female pupae and weight of male pupae. On the other hand, larvae fed on castor oil and cotton leaves gave shorter longevity of male and females, compared with berseem. Also, significant higher percentages of adult emergence, no. of eggs per female and hatchability were recorded in the second generation in case of castor oil. At the highest tested extract concentration (10%), data revealed higher mortality percentages at 72 hrs. Post-treatment for essential oils (97%) followed by petroleum ether extracts of cloves flowering buds (70), But in case of the acetone extract of cloves flowering buds the corresponding mortality percentage was very low (around 17 %). Gas Chromatography-Mass spectrometry (GC/MS) analysis of Cloves flowering buds essential oils. Data indicated that phenol, 2-methoxy 4-(2-propenyl)- (Eugenol) was the most abundant (78.13%) among all the other components followed by Beta-caryophyllene (11.83%) and Eugenyl acetate, recording 5.69% which followed by other constituents of oil sample. Data showed that the unsaponifiable Fraction of petroleum ether extract of Cloves flowering buds has obviously higher toxicity against *S. littoralis* larvae compared to the saponifiable one. compiled the fatty acids of unsaponifiable fractions of Cloves which were distinguished by having. Penlacosanoic acid as a major unsaturated fatty acids. Constituted 16.019% of the total fatty acids. Melissic acid (Triacontanoic) was found to be the second abundant unsaturated fatty acid comprising 14.612% followed by cerotic (Hexacosanoic) which constituted 12.029%, tetracosanoic acid (5.295%), docosanoic acid which constituted 5.263, Actacosanoic acid which constituted 4.143 and Anachidic (heptacosanoic acid which constituted 2.276 but a know which consisted 37.889. During the three sprays, effects of most treatments showed similar effect or mortality percentages. In the first spray, after the first day, the mortality percentage ranged from (0.00) untreated to (53.33) dipel2x 2.5g + lannate 0.75g. Also, the mortality percentage after two days ranged from 1.33 to 91.89 by the same previous treatments. After three days, the highest mortality percentage recorded from treated with petroleum ether extract of Clove 50g + 1 annet 0.75g. After 5`h days, all treatments recorded significantly increased mortality percentage compared with untreated except essential oil cloves 25g + lannate 0.75g, petroleum ether extract of cloves 25g, acetone extract of cloves (5 g or 25 g) and dipel2x 1.25g. In the second spray, after one day from spray, the mortality percentage ranged from (0.00) untreated to (72.0%) lannate 1.5g or petroleum ether extract of cloves 50g + lannate 0.75g. After two days from spray, the mortality percentage ranged from 0.00 of untreated to 92.33 % by 1.5g of lannate. After 3 and 5 days from spray, the two treatments (25 g of petroleum ether extracts of Cloves and 1.25g of dipel2x showed insignificant effects on mortality percentage compared with untreated. The highest mortality percentage was recorded by essential oil of Cloves 50 g + lannate 0.75g. While, the petroleum ether extracts of clove 25 g gave the lowest one. In the third spray, generally the same trend was obtained as shown in the first and second spray. After 14 days from spray, twelve treatments i.e. (lannate 1.5 or 0.75g, essential oil of Cloves 50 or 25g with 0.75g of lannate, petroleum extract of clove 50g, petroleum ether extract of clove 50 g or 25 g with lannate 0.75g., acetone extract of clove 50g, acetone extract of Clove 50 or 25g with 0.75g lannate and dipel2x 2.5 or 1.25g with 0.75 lannate) gave significant higher mortality percentage. The highest larval mortality percentages were recorded for the treatment of petroleum ether extract of clove + lannate (50 g+0.75 g) Summary 218 followed by acetone extract of Cloves + lannate (5 g + 0.75 g) and then by dipel2x + lannate (2.5g +0.75 g).

With the exception of 25 g of essential oil of cloves + 0.75 lannate and 25 g of essential oil of cloves, all treatments exhibited significantly higher mortality percentages compared with control. Results showed that the bio-residual activity of the various treatments to the 2nd instar larvae of *S. littoralis* was significantly decreased by time elapse. At the same time, data indicated lower mortality values for different treatment 14 days from treatment. All treatments gave significantly lower chlorophyll content of tomato leaves than untreated, except 25 g of essential oil of Cloves flowering buds. The highest mean values of total yield was obtained by 50 g of acetone extract of clove (1763.33 g) followed by 25 g from this extract (1513.3 g) followed by dipel2x 2.5 g + lannate mixture 0.75g (1250). On the other hand, the lowest total yield (652.33 and 635.33 g) was obtained for the treatment of essential oil + lannet (50 g + 0.75 g) and (25 g + 0.75 g), respectively, followed by untreated (813.67 g).