

## TOXICOLOGICAL AND BIOLOGICAL EFFECTS OF CERTAIN PLANT EXTRACTS ON LARVAL STAGE OF BLACK CUTWORM UNDER LABORATORY CONDITIONS

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The black cutworm (BCW), *Agrotis ipsilon* (Hufnagel) (Lepidoptera: Noctuidae) is distributed in many countries of the world and cause great losses on most vegetables and field crops. Under laboratory conditions, the toxicity of plant extracts (Camphor, Sea ambrosia and the Thorn apple) and pyriproxyfen as insect growth regulator (IGR) were investigated on the larval stage of black cutworm *Agrotis ipsilon*. The new 3<sup>rd</sup> larval instar were treated with different concentrations (ppm) of the tested materials. Results clearly indicated that the pyriproxyfen (LC<sub>50</sub> value was 275.40 ppm) was the most effective than other plant extracts (PEs). The LC<sub>50</sub> values were 3850.36, 4230.15 and 4375.22 ppm for Damsissa, Camphor and Datoora leaf extracts, respectively. Treatments reduced food consumption, feeding ratio and larval weight, whereas increased larval duration and larval mortality %. The most effective compound was pyriproxyfen followed by Damsissa, Datoora and the least was Cafoor leaf extracts, respectively. The joint action of PEs at LC<sub>25</sub> with pyriproxyfen at LC<sub>25</sub> values showed that the mixture with Damsissa leaf extract was potentiation, followed by the mixture with Datoora and the mixture with Cafoor leaf extract (additive action). The corresponding rates of Co-toxicity factor were 39.51, 27.87 and 17.65% of the mixture with Damsissa, Datoora and Cafoor leaf extracts, respectively. This study concluded that the plant extracts and pyriproxyfen have insecticidal activity against *Agrotis ipsilon* larvae and the mixture of previous plant extracts with pyriproxyfen has a good result on larvae, especially with Damsissa leaf extracts and is possibly used as an alternative to conventional chemical insecticides.

**Keywords:** *Agrotis ipsilon*, toxicity, food consumption

## INTRODUCTION

The seedlings of most crops, including cotton, potatoes, tobacco, cereals, and crucifers, are attacked by the polyphagous black cutworm *Agrotis ipsilon* (Hufnagel). Around the world, it is regarded as a cosmopolitan pest that occasionally affects a wide variety of crops. This pest causes an important danger to Egypt's grain, vegetable, and cotton crops. The older larvae eat at night at the base of crop plants or on the underground roots or stems, while young larvae consume the leaves of numerous crops. Usually cut through at ground level, seedlings might be destroyed by a single caterpillar (Fahmy, 2008; Sallam, 2008; Jeyasankar, 2012; Sharaby and El-Najiban, 2015; Nasr et al., 2021 and Farouk et al., 2023). Chemical insecticides may control these pests to some extent, but in recent years, their risks have led to a rising attack on these pesticides. The prolonged use of synthetic pesticides, which are the primary methods used in developed countries for crop production, may result in serious problems. These problems include insect species' genetic resistance, hazardous residues in agricultural products, handling risks, worker health risks. The frequent and extensive use of synthetic pesticides causes direct toxicity to non-target organisms, including predators and beneficial parasites. Food chains also contain concentrations of some chemicals. Consequently, it might be beneficial to find natural pesticide supplements (Davies et al., 2007; Sallam, 2008; Ali et al., 2017; Ebeid et al., 2017; Vattikonda and Sangam, 2017; Shaurub et al., 2020; Abdel-Kader et al., 2021 and Mohammed et al., 2023).

Thus, search on the insecticidal effect of agent naturally produced by plants has been conducted for many years. Farmers are familiar with plants that produce compounds that have insect growth regulators (IGRs), feeding deterrents, repellents and confusants because they typically grow in the same general area. These plants also frequently have other uses, such as household insect repellents or medicinal plants. The active product's quick breakdown could be useful because it lowers the possibility of residues on food. Before harvesting, some of these products might be put to use. Even though they don't kill insects over time, several of these products work really quickly to stop the insects from feeding. They may be less harmful to natural enemies and more selective to insect pests because many of these compounds have a stomach action and break down quickly. Many of these substances quickly affect, are not phytotoxic, and are not harmful to plants or mammals. However, compared to synthetic pesticides, resistance to these chemicals does not develop as quickly (Copping and Duke, 2007; Sallam, 2008; Duke et al., 2010 and Aremu et al., 2024).

As a result, finding new, ideally ecologically friendly insecticides became a crucial challenge (Sallam, 2008). Several plant extracts (PEs) that are view as promising natural insecticides (Lambrano et al., 2014). Many researches on insecticidal activity of plant extracts on the black cutworm

(BCW) insect pest (El-Shershaby 2010; El-Hosary et al., 2013; Moustafa et al., 2021; Shaurub et al., 2022; Farouk et al., 2023 and Nasr et al., 2024) were recorded insecticidal activity of PEs on black cutworm (BCW) insect pest. Also, other researches showed the insecticidal activity on Lepidopteran insect pests such as on *Spodoptera littoralis* (El-Kholy and Shaheen, 2004; El-Kholy et al., 2014 and Taha-Salaime et al., 2020); on *Spodoptera Frugiperda* (Alves et al., 2018), on *Tota absoluta* (Pinto et al., 2020) and on *Sesamia certica* (Ismail et al., 2023). Also, Lambrano et al. (2014) found that the effects of plant products on insect pests can be manifested in several manners including toxicity, mortality, insect growth regulators (IGRs), antifeedant, growth inhibition, reduction of reproductive behavior (fecundity and fertility).

Insect growth inhibitors (IGIs) and insect growth regulators (IGRs) are active against many insect species. From the IGIs, pyriproxyfen is effective on insect pests. This compound is used to control BCW insect (El-Sappagh, 2015; Ibrahim and Gaber, 2020 and Bakr et al., 2021) and on *S. littoralis* (Ali, 2009 and Thabet et al., 2024) and on *S. littura* (Khan and Naveed 2020 and Basu and Maddheshiya, 2023). Due to the use of pyriproxyfen in many countries in the world, it to know how this compound behaves in environment and the side effect on non-target organisms. Also, used to control insect pests of many vegetables and field agricultural plants all over the world.

Therefore, the aims of this study are to evaluate the insecticidal activity of three plant extracts in comparison with the pyriproxyfen compound (standard) and the joint action of these treatments on 3<sup>rd</sup> larval instar under laboratory conditions for possible use as a safe method for alternative approaches to conventional chemical insecticides within Integrated pest management programs (IPM).

## MATERIALS AND METHODS

### 1- The Black Cutworm (BCW) Insect

The larvae of the BCW were obtained from the culture maintained in laboratories of the Department of Economic Entomology and Pesticides, Faculty of Agriculture, Cairo University without exposure to any pesticides or microbes. The larvae were reared in laboratories of Department of Plant Protection, Faculty of Agriculture, Cairo, Al-Azhar University, Egypt. The rearing technique was similar to obtained by Abdin (1979). The fresh and clean castor bean leaves (*Ricinus communis* L. Malpighials: Euphorbiaceae) were used as food for larvae.

These feedings were performed inside jars (1L.) and supplied with a layer of sawdust at the bottom to absorb moisture as a recommended by Abdel-Rahim (2002) and El-Hosary et al. (2013) with some modifications. Under laboratory condition at 25±2°C and 65±5% R.H. and a 12-h photoperiod, these larvae were supplied daily with fresh leaves of castor bean

up to the third instar, then they were reared in the plastic cups, individually (5 cm in diameter and 5 cm height), (Gesraha et al., 2018), with saw dust on the bottom and covered with muslin, to avoid the crowding and cannibalism until the pupation. The newly molted 3<sup>rd</sup> instar larvae of BCW were weighed before and after feeding daily until pupation. The pupae were kept in glass jars (1 L) until the emergence of moths. The adult moths were supplied with a 10% sucrose solution (Shaurub et al., 2018).

## 2. The Tested Materials

Three plant extracts were used along with pyriproxyfen insecticide as a comparable key in this study as shown in Table (1). Pyriproxyfen (Admafin 10% EC) was supplied by Kanza Group Co. for Chemicals and Pesticides.

**Table (1).** The tested plants.

| Arabic name | English name             | Scientific name               | Family     | Crude extracted yield (g Kg <sup>-1</sup> leaves) | Source of plants                              |
|-------------|--------------------------|-------------------------------|------------|---|---|
| Cafoor      | Camphor                  | <i>Eucalyptus globules</i> L. | Myrtaceae  | 80.52   | Fac. of Agric. (Cairo) Al-Azhar Univ.         |
| Damsissa    | Sea ambrosia             | <i>Ambrosia maritima</i> L.   | Compositae | 111.64  | Farm of Fac. of Agric. Mostorod Kalubiah Gov. |
| Datoora     | Thorn apple or Worm wood | <i>Datura stramonium</i> L.   | Solanaceae | 94.40   | Fac. of Agric. (Cairo) Al-Azhar Univ.         |

These plants were identified in Faculty of Agriculture (Cairo), Al-Azhar University in the Department of Agricultural Botany.

Samples from three local plant species belonging to different families were used for extraction processes according to El-Torkey (2008). The young fresh leaves were cleaned from any dust and debris with sterilized distilled water (SDW) three times. Leaves were dried with tissues and left at room temperature for 7 days. Dried leaves were ground individually in a mill and then immersed in 95% ethanol for seven days, during which time the soaked leaves powder was shaken for an hour per day with an electric shaker. Filter paper Whatman's No. 1 and the anhydrous Na<sub>2</sub>SO<sub>4</sub> were used to filter the PEs. A rotary evaporator and hot water bath were used to evaporate the solvent under vacuum, and it was then stored in a refrigerator at -4°C for the further studies.

## 3. Treatments

### 3.1. Toxicity and determination the LC values

To evaluate the possible toxicity of these PEs in comparison with pyriproxyfen, the LC values, slope, toxicity index and relative potency level were recorded after 72 h from treatments (Finney, 1971). Larval mortality was noted, and if the larva did not react to touch stimulation, it was regarded as

dead (Osman and Mahmoud, 2009). The mortality% corrected by Abbott's formula as follow:

$$\text{Corrected mortality \% (Abbott's, 1925)} = \frac{\% \text{ of observed mortality} - \% \text{ of control mortality}}{100 - \text{control mortality \%}} \times 100.$$

The third larval instars of BCW were used in this experiment ( $25.0 \pm 5$  mg for each). Different concentrations (from each treatment) were used to calculate the LC values. The leaves of plant extracts were tested and pyriproxyfen in serial concentrations. Leaves of *R. communis* were dipped into each concentration (after washed with sterile distilled water three times) and dried at room temperature, then the larvae and leaves were weighed before the start and placed into plastic cups (5 cm in diameter and 5 cm in high). The third larval instar was placed in each cup individually as mentioned above and were starved first for two hours to bring them in the same of nutrition (Shaurub et al., 2018).

### 3.2. Effect of treatments on biological parameters

On larval stage after the determination of the LC<sub>50</sub> values, the effect of treatment (at LC<sub>50</sub> values) was evaluated. The castor bean leaves were dipped in LC<sub>50</sub> concentration for 15 seconds, then the effect of treatments on larval stage was noted (from each treatment) and in the same manner as previously reported. The feces were discarding. After accounting for the natural loss of moisture, the weight of the fresh and uneaten leaves was weighed every day. Two controls were used; control (1) is larvae with untreated leaves and control (2) is leaves only without larvae.

The consumed fresh leaves were recorded by the following formula of Ghanema (2002) at the end of larval stage.

$$\text{Corrected weight of the consumed leaves} = C_b / C_a \times T_a,$$

Where:

C<sub>b</sub> = Initial weight of leaves before larval exposure.

C<sub>a</sub> = Final weight (after exposure to natural dryness for 24 h) of leaves without larvae.

T<sub>a</sub> = Final weight of treated leaves after feeding the larvae for 24 h.

Daily weight (fresh basis) of consumed treated leaves<sup>-1</sup> larva =  $A - B / C$ .

Where:

A = Initial fresh weight of treated leaves before feeding the larvae.

B = Corrected fresh weight of treated leaves after feeding the larvae.

C = Number of survival larvae.

Also, the following data were calculated as follows:

- Mean weight of consumed leaves (gm larva<sup>-1</sup>).
- Feeding ratio (F.R.) =  $b / a \times 100$  (Wada and Munakata, 1968).

Where:

a = Amount of fresh weight of consumed leaves in untreated.

b = Amount of fresh weight of consumed leaves in treatment.

- Feeding inhibition%.

- Antifeedant index (Pavela et al., 2008).

$$A.F.I. = [(C-T)/(C+T)] \times 100.$$

Where:

C= Weight of consumed leaves untreated.

T= Weight of consumed leaves in treatment.

The larval duration (days), larval weight (mg) and larval mortality % were recorded from 3<sup>rd</sup> larval instar until pupation.

### 3.3. Joint action of treatments

The experiments were designed on the mixtures of LC<sub>25</sub> from the most effective compound with other LC<sub>25</sub> from each treatment based on LC<sub>50</sub> values. The joint action of the different mixtures was expressed at the Co-toxicity factors, calculated and recorded according to Mansour et al. (1966) as follows:

Co-Toxicity factor (CTF%) =

$$\text{Observed\% mortality} - \text{expected\% mortality} / \text{expected\% mortality} \times 100$$

This Co-toxicity factor was used to categorize the results into three groups as follow:  $\geq +20$  mean, potentiation or synergism;  $< -20$  means antagonism; and between  $-20$  and  $+20$  means additive action. The larvae were tested as mentioned above previously reported and 100 BCW 3<sup>rd</sup> larval instar was used and the mortality % were recorded at 72 h after treatment.

### 4. Statistical Analysis

The results were analyzed using one-way ANOVA through the SAS (2004) program and significant difference between treatments were calculated by using Duncan's Multiple Range Test (Duncan, 1955).

## RESULTS

The data illustrated in Table (2) explain the LC<sub>25</sub>, LC<sub>50</sub>, LC<sub>90</sub>, slope values, toxicity index (TI) and relative potency (RP) values for the tested materials on the 3<sup>rd</sup> larval instar of the black cutworm (BCW) *Agrotis ipsilon* using the leaf-dipping technique after 3 days of treatments. It is clearly that pyriproxyfen was the most efficient (LC<sub>50</sub> = 275.40 ppm) as compared with Damsissa leaf extract (DAMLE), Cafoor leaf extract (CAFLE) and Datoora leaf extract (DATLE). Damsissa was the most effective than other extracts. The LC<sub>50</sub> values were 3850.36, 4230.15 and 4375.22 ppm for DAMLE, CAFLE and DATLE, respectively. The corresponding toxicity index (TI) values of DAMLE, CAFLE and DATLE were 7.16, 6.51 and 6.29%, respectively based on LC<sub>50</sub> values of pyriproxyfen (100%). Results from slope values showed that the insect population was relatively homogeneous in the susceptibility toward tested materials (slope values in the table were less than 1.0 for all the tested materials).

**Table (2).** Toxicity values of pyriproxyfen and plant extracts after three days on 3<sup>rd</sup> larval instar of black cutworm *Agrotis ipsilon* under laboratory conditions.

| Treatments              | Lethal concentrations (ppm) |                                       |                         | Slope $\pm$ SE*  | Toxicity index (T.I.)** | Relative potency (R.P.)*** |
|-------------------------|-----------------------------|---------------------------------------|-------------------------|------------------|-------------------------|----------------------------|
|                         | LC <sub>25</sub> values     | LC <sub>50</sub> values (lower-upper) | LC <sub>90</sub> values |                  |                         |                            |
| Cafoor                  | 2150.51                     | 4230.15<br>(3950.11 - 4420.75)        | 8462.25                 | 00.88 $\pm$ 0.35 | 06.51                   | 15.36                      |
| Damsissa                | 1516.11                     | 3850.36<br>(3640.16 - 4120.25)        | 6540.66                 | 00.92 $\pm$ 0.23 | 07.16                   | 13.98                      |
| Datoora                 | 2120.26                     | 4375.22<br>(4211.26 - 4605.43)        | 8290.51                 | 00.97 $\pm$ 0.39 | 06.29                   | 15.89                      |
| Pyriproxyfen (Standard) | 103.25                      | 275.40<br>(195.61 - 361.19)           | 440.23                  | 00.89 $\pm$ 0.61 | 100.00                  | 01.00                      |

\*SE = Standard error. \*\*T.I. = the toxicity index was calculated according to (Sun, 1950) by LC<sub>50</sub> of most effective compound/ LC<sub>50</sub> of tested compound X100.

\*\*\*R.P. = the relative potency level was calculated according to (El-Sheikh and Alamir, 2011) by LC<sub>50</sub> of the tested compound/ LC<sub>50</sub> of the most effective compound.

Table (3) shows the impact of investigated materials on food consumption included weight of consumed leaves (mg larva<sup>-1</sup>), feeding inhibition (%) and antifeedant index (%) from the 3<sup>rd</sup> larval instar until pupation. The consumed leaves mean weight showed that DATLE was the least on consumption followed by DAMLE and CAFLE, respectively. The most significant material was pyriproxyfen ( $P=0.05$ ) compared with other materials and untreated control. All treatments reduced feeding ratio %, feeding inhibition % and Antifeeding index values compared with untreated control. Values of feeding ratio were 59.4, 64.32, 68.25 and 74.34% for pyriproxyfen, DAMLE, CAFLE and DATLE, respectively. Similarly, feeding inhibition values were 40.60, 35.68, 31.76 and 25.66% for pyriproxyfen, DAMLE, CAFLE and DATLE, respectively. The same trend showed by antifeeding index of 26.20% for pyriproxyfen, 21.90% for DAMLE, 19.18% for DAMLE and 14.92% for DATLE, respectively.

Impact of all treatments on larval weight, larval duration and larval mortality% (at the end of larval stage) were recorded and shown in Table (4). The effect on larval weight (mg larva<sup>-1</sup>) showed that pyriproxyfen was the most effective in reducing larval weight and significantly than other treatments followed by DAMLE, CAFLE and DATLE, respectively. The highest reduction % of larval weigh recorded 29.51% with pyriproxyfen followed by 17.23% with DAMLE while the lowest reduction values recorded 6.98% and 5.49% for CAFLE and DATLE, respectively. Impact of tested materials on larval mortality % demonstrated that pyriproxyfen was the most effective treatment. values of larval mortality recorded 81.8, 69.2, 66.2 and

64.8% for pyriproxyfen, DAMLE, DATLE and CAFLE, respectively. Pyriproxyfen and DAMLE gave the highest values than other treatments.

**Table (3).** Effect of pyriproxyfen and plant extracts on food consumption of larval stage of black cutworm *Agrotis ipsilon* from 3<sup>rd</sup> larval instar until pupation under laboratory conditions.

| Treatments            | Food consumption  |                 |                      |                     |
|-----------------------|---|-----------------|----------------------|---------------------|
|                       | Mean weight of consumed leaves (mg larva <sup>-1</sup> ) ± SE | Feeding ratio % | Feeding inhibition % | Antifeedant index % |
| Untreated             | 04.29 ± 0.49 <sup>a</sup>                                     | 00.00           | 00.00                | 00.00               |
| Cafoor leaf extract   | 03.59 ± 0.48 <sup>bc</sup>                                    | 68.25           | 31.76                | 19.18               |
| Damsissa leaf extract | 03.39 ± 0.47 <sup>bc</sup>                                    | 64.32           | 35.68                | 21.90               |
| Datoora leaf extract  | 03.92 ± 0.48 <sup>ab</sup>                                    | 74.34           | 25.66                | 14.92               |
| Pyriproxyfen          | 03.12 ± 0.67 <sup>c</sup>                                     | 59.40           | 40.60                | 26.20               |
| L.S.D. at 0.05        | 00.69   |                 |                      |                     |

• SE = Standard error. • Mean values followed by different letters in the column are significantly different ( $P \leq 0.05$ ).

**Table (4).** Effect of pyriproxyfen and plant extracts on larval weight (mg larva<sup>-1</sup>), larval duration(days) and larval mortality (%) from the 3<sup>rd</sup> larval instar until pupation of black cutworm *Agrotis ipsilon* under laboratory conditions.

| Treatments            | Mean larval weight (mg larva <sup>-1</sup> ) ± SE | Reduction % | Mean larval duration (days) ± SE | Increase than control % | Larval mortality % |
|-----------------------|---|-------------|----------------------------------|-------------------------|--------------------|
| Untreated             | 372.55 ± 21.41 <sup>a</sup>                       | 00.00       | 19.20 ± 1.34 <sup>b</sup>        | 00.00                   | 00.00              |
| Cafoor leaf extract   | 346.53 ± 26.83 <sup>ab</sup>                      | 06.98       | 19.88 ± 1.16 <sup>ab</sup>       | 03.54                   | 64.80              |
| Damsissa leaf extract | 308.35 ± 38.08 <sup>b</sup>                       | 17.23       | 21.31 ± 1.08 <sup>a</sup>        | 10.99                   | 69.20              |
| Datoora leaf extract  | 352.09 ± 24.40 <sup>a</sup>                       | 05.49       | 20.89 ± 1.20 <sup>ab</sup>       | 07.93                   | 66.20              |
| Pyriproxyfen          | 262.59 ± 46.11 <sup>c</sup>                       | 29.51       | 21.82 ± 1.75 <sup>a</sup>        | 13.65                   | 81.80              |
| L.S.D. at 0.05        | 43.16   |             | 01.96                            |                         |                    |

• SE = Standard error. -The reduction and increase% were calculated from treatments and untreated values.

• Mean values followed by different letters in the column are significantly different ( $P \leq 0.05$ ).

The joint action of pyriproxyfen at LC<sub>25</sub> with other treatments is described in Table (5). The results demonstrated that all mixtures increased mortality%, especially when mixing the pyriproxyfen and DAMLE at LC<sub>25</sub>. The mix of pyriproxyfen with DAMLE gave 39.51% of Co-Toxicity Factor (CTF%), while the mix of pyriproxyfen with DATLE recorded 27.87% of CTF%, and these effects are described as a potentiation (synergism), and the mix of pyriproxyfen with CAFLE gave 17.65% of CTF% and is explained as an additive action.



**Table (5).** Joint action of pyriproxyfen and plant extracts on 3<sup>rd</sup> larval instar of black cutworm *Agrotis ipsilon* after three days under laboratory conditions.

| Treatments  | Expected mortality % | Observed mortality % | Co-toxicity factor % • | Description                 |
|---|----------------------|----------------------|------------------------|-----------------------------|
| Pyriproxyfen LC <sub>25</sub> + Cafoor LC <sub>25</sub>   | 42.00                | 51.00                | 17.65                  | Additive                    |
| Pyriproxyfen LC <sub>25</sub> + Damsissa LC <sub>25</sub> | 49.00                | 91.00                | 39.51                  | Potentialiation (Synergism) |
| Pyriproxyfen LC <sub>25</sub> + Datoora LC <sub>25</sub>  | 44.00                | 61.00                | 27.87                  | Potentialiation (Synergism) |

- One hundred third instar larvae were used in each treatment.
- In each treatment the mortality from pyriproxyfen at LC<sub>25</sub> was collected with the same values with each LC<sub>25</sub> from other plant extracts (Total).

## DISCUSSION

One of the most important polyphagous and underground damaging pests in the world is the black cut worm (BCW) *Agrotis ipsilon* (Hufn.) (Lepidoptera: Noctuidae). Because it damages the roots of many plants, this insect pest has caused a high level of economic damage to several kinds of crops, especially at seedling stage (El-Hosary et al., 2013; Wang et al., 2021 and Amein and Mahammad, 2022). The use of conventional chemical insecticides on a large scale caused many problems, such as the appearance of resistance to insecticides, resurgence, residual effects in the environment, and effects on non-target organisms such as natural enemies and beneficial insects, while causing major issues by direct toxicity to people, animals and fish (Ali et al., 2017; Hawkins et al., 2019; Shaurub et al., 2020 and Akbar et al., 2024). Therefore, we need to develop alternative or additional techniques to avoid the problems from use of conventional chemical insecticides.

The usage of PEs and IGRs is one of the best and most promising alternatives. The application of PEs or plant oils (POs), which are natural compounds that have been shown to negatively impact on the target insects, might be problematic since it can pollute the environment with toxic compounds. Use of IGRs impact on insect growth regulator hormones that are specific for insects and not for animals and humans. When compared to traditional chemical pesticides with IGRs, they are probably less hazardous to natural enemies because of their high selectivity for the target insect species. Additionally, IGRs affect specific physiological regulators that are necessary for insect pests to develop normally (Sallam, 2008; Hawkins et al., 2019 and Khursheed et al., 2022).

Accordingly, to verify the existence of new insecticides against *A. ipsilon* insect pest, to avoid the problems from use of conventional chemical insecticides. We studied the toxicity and effect of three PEs and one IGRs and activity of these materials on *A. ipsilon* under laboratory conditions for

possible use as a safe method of alternative approaching the conventional chemical insecticides with an IPM program.

Results showed that the  $LC_{50}$  values were varied between treatments and IGR (pyriproxyfen) was the most effective than other PEs. Also, PEs varied between us. The most effective was Damsissa leaf extract (DAMLE) followed by Cafoor leaf extract (CAFLE) and Datoora leaf extract (DATLE) was the least effective (Table 2). A similar trend was observed by next reports. Jeyasankar (2012) found eucalyptus oil (*E. globules*) and *G. procumbens* oils (POs) has potential to serve as alternative eco-friendly control of *A. ipsilon* insect pest. El-Kholy et al. (2014) found varian in toxicity of the three PEs and they reported that Damsissa LE showed more pronounced toxic than Datoora LE and Cafoor LE on 4<sup>th</sup> larval instar of *S. littoralis* insect pest. Also, Nasr et al., (2021) found that *M. longifolia*, *A. Judaica*, *M. hortensis*, *O. syriacum* and *A. santolina* plant oils differ in toxicity on *A. ipsilon* 4<sup>th</sup> instar larvae. The toxicity of *A. Judaica*, *M. longifolia* and *O. syriacum* were more toxic than *M. hortensis* and *A. santolina*, respectively. Bakr (2021) found that the  $LC_{50}$  values (ppm) of pyriproxyfen were 65.95 ppm on 4<sup>th</sup> instar larvae and 99.90 ppm on 5<sup>th</sup> instar larvae of *A. ipsilon*. In the present study, we determined the  $LC_{50}$  value on third instar larvae and the  $LC_{50}$  value was 275.40 (ppm). This variation may be due to the age of larval stage and colony of this insect. Also, the variation in  $LC_{50}$  between three PEs may be due to differences in components between these PEs. These results are in accordance with those obtained by researchers. Kokila and Jeyabalan (2023) demonstrate that *C. winterianus* was more taxic than *A. cityodera* PEs against *A. ipsilon* insect. Bakr et al. (2021) found that pyriproxyfen exhibited strong acute toxicity activity against larvae of *A. ipsilon* insect. They found that 4<sup>th</sup> instar larvae were the most susceptible than 5<sup>th</sup> instar.

Table (3) explains the effect of these materials on larval stage (food consumption). It was noticeable from the results that pyriproxyfen was the most toxic and three PEs were the least effective. A different variation was found between these PEs. The DAMLE was more effective than CAFLE and DATLE, respectively. All treatments reduced the weight of consumed leaves ( $mg\ larvae^{-1}$ ) significantly ( $p=0.05$ ) compared with the untreated control. They reflected on feeding ratio and the antifeedant index%. The IGR caused 59.40 feeding ratio followed by DAMLE followed by CAFLE and DATLE, respectively. El-Kholy et al. (2014) mentioned that the PEs from leaves of Cafoor, Damsissa and Datoora leaves reduced food consumption and caused reduction in feeding ratio on 4<sup>th</sup> larval instar of *S. littoralis* insect pest and Damsissa was more effective than Datoora and Cafoor. Ramadan (2020) found that the Egyptians conyza, *Conyza aegyptiace* (whole plants) reduced food consumption ( $mg\ larvae^{-1}$ ) on 5<sup>th</sup> instar larvae of *A. ipsilon* insect pest and varied than *V. rosea* and *M. azedarach* leaf extracts. Similar trend of results was recorded by Nasr et al., (2021) they found that the most toxic POs decreased food consumption, causing a significant reduction in relative

consumption rate, and POs varied in this result, some POs were more effective than others.

The effect on larval weight, larval duration and larval mortality% (Table 4) were varied between these treatments. The most effective compound was pyriproxyfen significantly ( $p=0.05$ ) than other PEs. The DAMLE was significantly more effective than DATLE treatment, whereas no significant between CAFLE and untreated control. The decrease in larval weight under the influence of the tested materials is mainly attributed to the decrease in larval food consumption. These results agree with Bakr (2021) who mentioned that larval body weight was remarkably reduced, and the growth was considerably reduced when treated with pyriproxyfen. Thus, the DAMLE as a plant extract exhibited the same way of pyriproxyfen as an IGR in reducing the body weight of the treated larvae.

On the other hand, treatment with these compounds caused increase in the larval duration as mentioned by Sallam (2017) and Dahi et al. (2017) they refer to an imbalance between ecdysone secretion and juvenile hormone (JH) may be the cause of the elongation in larval periods, as the critical concentration of ecdysone is achieved much later than in untreated larvae. Additionally, the epidermis is prepared for a larval molt when the corpora allata secrete a small amount of JH. An important problem in the growth and development of insects can result from any disruption in the normal hormone. Shaurub et al., (2022), they attributed the development of *A. ipsilon* larvae can disrupt by targeting multiple metabolic pathways and the cellular immune system by pyriproxyfen and *M. azadarach* fruit extract. Larval duration was considerably prolonged, on 4<sup>th</sup> and 5<sup>th</sup> instar larvae of *A. ipsilon* when treated with pyriproxyfen. Amein and Mohammad (2022) found the flufenoxuron IGR enhanced the mean larval duration. Kokila and Jeyapalan (2023) found that larval duration was greatly extended after the treatment of *C. winterianus* than *A. citrodera* plant extracts at different concentrations on *A. ipsilon* 4<sup>th</sup> larval instar, and larval mortality was 90.42% and 98.32% for *A. citrodera* and *C. winterianus* at 2% concentration. Farouk et al. (2023) found that the *N. sativa* straw extract increased mortality in the 4<sup>th</sup> larval instar of *A. ipsilon* which reached to 96.67 % at the highest concentration. Akbar et al. (2024) mentioned that the high mortality rates may be caused by the interference of plant extracts with essential metabolic, behavioral, biochemical and physiological processes of insects. Moreover, insects can also cause their high obsession by harming their nerve cells. On the other hand, IGRs interferes with insect growth regulator hormones, as mentioned previously.

Results in Table 5 showed the joint action of pyriproxyfen (the most toxic compound) with PEs materials. The mixture of pyriproxyfen at LC<sub>25</sub> with LC<sub>25</sub> from each PEs showed that the mixture with DAMLE caused synergism (potentiation) with Co-Toxicity Factor 39.51%, followed by the mixture with DATLE, also recorded as synergism with Co-Toxicity Factor 27.87%. Nasr et al. (2024) mentioned that the use of synergists is a useful strategy for managing

insecticide resistance, improving the efficacy of pesticides, delaying or stopping their breakdown, increasing their toxicity to insect pests, and lowering the quantity of pesticides used. On the other hand, the mixture with CAFLE caused additive action with Co-toxicity factor 17.65%. Commercial mixing compounds have recently been made available by numerous agricultural services companies. These substances have the potential to significantly enhance the Insect Pest Management Program (IPM), including the potential impact of lowering the quantities of each agent used, such as lowering costs, reducing pollution of the environment, causing hazards to beneficial organisms, and reducing selective pressure, which can lead to the development of resistance to each agent (Korrat et al., 2012; El-Sheikh, 2015; Khatun et al., 2015; Kandil et al., 2020; Abdel-Kareem et al., 2022 and Nasr et al., 2024).

## CONCLUSION

Generally, from these results, it could be concluded that Pes from plants and IGR (pyriproxyfen) were used as alternative to chemical insecticide, and DAMLE, DATLE and CAFLE have insecticidal activity against *A. ipsilon*. The mixture of plant extracts and pyriproxyfen also gave good results on populations of black cutworm specially between pyriproxyfen with DAMLE and we can be used in integrated pest management. For the future we needed many researches of these PEs under field conditions.

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## التأثيرات الإبادية والبيولوجية لبعض المستخلصات النباتية على الطور اليرقي للدودة القارضة تحت الظروف المعملية

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تنتشر الدودة القارضة *Agrotis ipsilon* في العديد من دول العالم وتسبب خسائر كبيرة في معظم الخضروات والمحاصيل الحقلية. معملياً تم اختبار سمية المستخلصات النباتية (الكافور - الدميسية - الداتورة) مع البيريبروكسيفين كمنظم نمو حشري على الطور اليرقي للدودة القارضة *Agrotis ipsilon*. تمت معاملة يرقات العمر الثالث حديثة الظهور بتركيزات مختلفة (جزء في المليون) من المواد المختبرة. أشارت النتائج بوضوح إلى أن بيريبروكسيفين كان الأكثر فاعلية وسمية من غيره من مستخلصات النباتات حيث كانت قيمة التركيز القاتل لنصف عدد الأفراد ( $LC_{50}$ ) ٢٧٥,٤٠ جزء في المليون. وكانت قيم ( $LC_{50}$ ) التركيز القاتل لنصف عدد الأفراد ٣٨٥٠,٣٦ و ٤٢٣٠,١٥ و ٤٣٧٥,٢٢ جزء في المليون لمستخلصات أوراق الدميسية والكافور والداتورة على التوالي. أدت هذه المعاملات إلى تقليل استهلاك الغذاء ونسبة التغذية ووزن اليرقات، بينما زادت مدة الطور اليرقي ونسبة الموت في اليرقات. كان مركب البيريبروكسيفين الأكثر فاعلية يليه الدميسية ثم الداتورة وكان الأقل تأثيراً مستخلص أوراق الكافور على التوالي. أظهر التأثير المشترك لخلط هذه المواد عند  $LC_{25}$  من البيريبروكسيفين (المركب الأكثر فعالية) مع قيم  $LC_{25}$  من كل معاملة أن الخلط مع مستخلص أوراق الدميسية كان معززاً للتأثير يليه الخلط مع مستخلص أوراق الداتورة وكان الأقل تأثيراً هو الخلط مع مستخلص أوراق الكافور (تأثير إضافي). كانت نسبة معامل السمية المشتركة ٣٩,٥١ و ٢٧,٨٧ و ١٧,٦٥٪ من الخلط مع مستخلصات أوراق الدميسية والداتورة والكافور على التوالي. خلصت هذه الدراسة إلى أن المستخلصات النباتية والبيريبروكسيفين لها نشاط إبادي على يرقات *Agrotis ipsilon* وأن خلط المستخلصات النباتية السابقة مع البيريبروكسيفين كان له نتائج جيدة على اليرقات خاصة مع مستخلصات أوراق الدميسية ويمكن استخدامهم كبديل لمبيدات الحشرات الكيميائية التقليدية.