Population Density and Biocontrol of Cucurbit fly, *Dacus ciliatus* Loew (Diptera: Tephritidae) in some vegetable plants of Cucurbitaceae

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**Abstract**

The cucurbit fly *Dacus ciliatus* Loew is important pest that infests some plants of Cucurbitaceae. The study was carried out to explain ecology of the insect and management. Field and laboratory experiments were conducted. Population numbers were considered in the field during autumn and spring seasons on some Cucurbitaceae vegetables, and different stick traps (yellow and blue) were applied to attract and observe the insect. Three bio-pesticides, *Bacillus thuriengiensis*, *Beauveria bassiana* and plant extract (*Tagets*) were applied by recommended rates to control this species*.* Three concentrations of each biopesticide (1g/L, 2g/L and 3g/L of *Bacillus thuriengiensis* ; 3g /L, 5g/L and 7g/L of *Beauveria bassiana* and 5ml/L, 10ml/L and 15 ml/L of plant extract (Tagets) were used in laboratory, then mortality was recorded after 1st, 3rd and 5th day. The highest population was recorded on the vegetables and by yellow stick in autumn season. The maximum reduction and mortality were found by the plant extract in the field and laboratory (15 ml/L).

Key word: *Dacus ciliatus*; Tephritidae; Diptera; Bio-pesticide and Plant extract; Population density yellow and blue stick traps

**Introduction**

 Cucurbit fly belong to Diptera order, Tephritoidae superfamily, Tephritidae Family and Dacinae subfamily, and Tephritidae is the largest family of Diptera (Clarke *et al.,* 2002) the family includes about 500 genera and approximately 5000 species (Norrbom, 2010) About 250 species of the family are economically important and widely distributed in temperate, sub-tropical, and tropical regions of the world (Christenson and Foote, 1960). Some species of the family can cause severe losses in vegetable crops particularly in Cucurbitaceae and Solanaceae in the world (Mziray *et al.,* 2010). Fruit flies (Diptera: Tephritidae) are reported to cause both direct and indirect losses. Direct damage is associated with female oviposition punctures in the fruits which might cause entering of diseases; then, the larvae feed on fruit tissue leading to premature ripening and falling of fruits and rotting (Ekesi and Mohamed, 2011). Indirect losses are due to quarantine measures imposed by importing countries to prevent an introduction of the fruit fly into recipient countries (Mugure, 2012) Carroll *et* *al.,* (2004) indicated that 190 species of family are economic impacts around the world. *Dacus ciliatus* is one of the important species of the family, the species is widespread in Africa and Asia. Details on its distribution are available in the EPPO Global Database (EPPO, 2018), and this species could be arranged as highly serious agricultural pests. The fruits of cucumbers are exposed to being infected with six types of flies in autumnal cultivation in central Iraq, and these species are arranged according to their economic importance as *D. ciliatus* (Loew), *D. frontalis* (Becker), *Atherigona orintalis* (Schin.), *Atherigona varia* (Meigen), *Myiopardalis pardalina* (Bigot) and *Ceratitis capitata* (Wiedeman) (Al-Jorany *et al.*, 2015). In addition, temperature is known to be a very important abiotic factor affecting immature development of insects (Wagner *et al.,* 1984). Fruit fly monitoring is essential for determining population dynamics, comparing infestation levels between various sites, and evaluating the success of a treatment technique (Eliopoulos, 2007). Fruit flies have been successfully managed using different techniques including cultural, legislative control, biological controls and chemical methods. Entomopathagenic fungi is microbes that act as parasites of insects by killing or disabling them, and the method is also satisfied to controls the fruit flies (Mar and Lumyong, 2012). Ohba and Aizawa (1986) noticed that the *Bacillus thuringiensis* produces proteinaceous crystalline structures (delta endotoxins) which are poisonous to a number of insects during the sporulation process. Ekesi *et al.,* (2007) mention that affirmed that the populations of fruit flies might be diminished and they reduce fecundity and fertility of the adults via entomopathogenic fungi such as *Metarhizium anisophilae* and *Beauveria bassiana*. In addition, plant extracts, which contain a high concentration of bioactive chemicals, may provide an alternative to synthetic insecticides for the controlling of plant pests and diseases (Pino *et al.,* 2013). Akhtar *et al.,* (2004) studied several plant extracts, such as *Azadirachta indica, A. Juss*, *Acorus calamus* L., *Curcuma longa* L., *Peganum harmala* L., *Taget minuta*, *Saussurea lappa* (Decne.) C. B. Clarke, and *Valeriana jatamansi* Jones and found growth prevention of *B. zonata*. Thus, plant extracts are considered one of non-chemical control methods due to their availability, lower human and mammalian toxicity, and environmental friendliness (Aqil *et al.,* 2010). The main aims of this study are to apply biopesticides [*Bacillus thuringiensis*, *Beauveria bassiana*, and plant extract (*Tagets*)] to control insect pests in laboratory and fields.

**Material and methods**

 **Field experiments**

Field experiments were conducted in autumn and spring seasons in 2021 and 2022 respectively at Grdarasha research station of the Agriculture Engineering Science / Salahaddin University – Erbil. After soil management, the land was designed according to randomized complete block design (RCBD) with three replicates to cultivate crops in Cucurbitaceae family consisting of (melon, cucumber, cucumis, pumpkin and snake cucumber). The experimental unit was 12 m2 (6\*2 m). The population abundance of cucurbit fly *Dacus ciliates* was detected and monitored on the vegetable crops. Effect of biotic factors such as temperature and relative humidity on cucurbit fly and vegetable crops were also recorded. The Temperature and relative humidity data were obtained for environmental study from the Planning Departments / General Directorate of Agriculture in Erbil governorate Table (1) show as below:

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Month/Year 2021 | Average of Temperature ◦C | Average of Relative humidity % | Month/Year 2022 | Average of Temperature ◦C | Average of Relative humidity % |
| April | 22.35 | 48.9 | April | 21.00 | 50 |
| May | 29.05 | 39.1 | May | 32.50 | 31 |
| Jun | 31.80 | 31.9 | Jun | 32.50 | 31 |
| July | 35.70 | 33 | July | 34.30 | 30.3 |
| August | 35.10 | 29.5 |  |  |  |
| September | 28.95 | 36.5 |  |  |  |
| October | 23.90 | 48.1 |  |  |  |

**Field Data collection:**

 Hand picking, Aspirator, Vials (Different size), Forceps, Yellow sticky and Blue sticky traps are common facilities used to find population density, monitoring of many insect pests; as well as, using light traps. two color (Blue and Yellow) trap were applied to collect cucurbit fly *D.* *ciliatus* in the cucurbitaceae crops. Yellow traps are well known to attract fruit flies as well as other insect species that are pollinators and predators as reported by Brévault and Quilici (2017).

Yellow and blue sticky traps were used to measure population and appearance period of cucurbit fly in open field. The stickies were measured 25cm length and 12cm width. They were placed on woody stands and used during 8/August to 29/October 2021 and 20/April to 2/July 2022 in the first and second season respectively. All the sticks were placed in five directions. Sticky traps are frequently used to manage pest populations or monitor pest prevalence in a specific location or region (Iaea, 2003).

 **Bio-pesticides:**

 Two types of microbial (*Bacillus thuriengiensis* and *Beauveria bassiana*) and plant extract (Tagets) were applied in the study.

**1- Laboratory experiment**

 Laboratory experiment was conducted according to complete randomized design (CRD).

with three replicates as the following steps:

A- *Bacillus thuringensis*: Three concentration of *B. thuringinsis* (1, 2 and 3g/L.) were applied to treat pieces of cucumber and placed in plastic petri dish. Then, 10 larvae 2nd instar were placed in every petri dish, and the larvae started to eat the cucumber; at that time, daily mortality of the larvae at 54 % relative humidity and 28.8 ◦C was recorded for five days.

B- *Beauveria bassiana*: Three concentrations (3, 5 and 7g /L.) were applied directly by spraying the larvae and cucumber and daily mortality was recorded for five days.

C- Tagets extracts: Tagets plants were collected and washed with water and kept under laboratory condition. Then, all parts plant were ground after drying to fine powder by using simple grinder, after that 10 g of mixed powder (leaves, flower and seeds) of Tagets plant was put in 1L. of distilled water for 24 h. The aqueous mixture was filtered then three concentrations (5ml, 10ml and 15ml /L.) were prepared from the extract, and they were applied directly by spraying in laboratory.

The mortality data were corrected according to Schneider-Orellis formula (1947):

$Corrected \%=\left(\frac{Mortality \% in treated Plot-Mortality \% in Control Plot}{100-Mortality \% in Control Plot }\right)\*100$

**2- Field experiment:**

A concentration of each treatment (2 g/L, 5 g/L and 10 ml/L) of *B. thuriengiensis*, *B. bassiana* and plant extract (*Tagets*); respectively, was sprayed according to randomized complete block design (RCBD) with three replicates on vegetable crops. The mortality was recorded in the 1st, 3rd and 5th days.

Mortality data was converted using the Abbott formula into percent corrected mortality (Abbott, 1925).

$Corrected \%=\left(1-\frac{n in T after treatment}{n in Co after treatment}\right)\*100 $

Where: n = Insect population, T = treated, Co = control

**Data analysis**

The statistical data processing was conducted using the statistical software program – IBM SPSS version 26.0 (Crop, 2019), The Duncan test was used to identify the differences between treatment means

**Result and Discussion**

The results of autumn season are shown in (Fig. 1). The highest population was recorded on cucumber and snake cucumber at the fourth week of Augast/2021 and the lowest observed at 12th week October and 7th weeks September/2021, respectively. However, the highest density of the insect was found in the first week Augast/2021 on cucurbit and pumpkin while the lowest was at 11th weeks October and 4th weeks Augast/2021, respectively. In addition, the population of *D. ciliates* was the highest in the second week Augast and lowest at week twelve October/2021 on melon crop. The variation of population numbers is influenced by abiotic factors such as temperature, relative humidity, rainfall and light and biotic factor (host plant). Masood *et al.,* (2009) observed in field and laboratory works that the neem and its components can reduce the population of the melon fruit fly, *Bactrocera cucurbitae*.

Fig (1): population density of cucurbit flies on some plants of cucurbitaceous vegetable during autumn seasons, 2021

The results of spring season are shown in (Fig. 2) showed that the highest population of cucurbit fly was recorded at 6th week May, 7th week May, 8th weeks Jun, 9th week Jun and 12th weeks July/2022 and the lowest was at 5th weeks May, 4th weeks May, 11th weeks Jun, 11th weeks July and 7th weeks May/2022 on cucumber, cucurbit, snake cucumber, melon and pumpkin, respectively.

Fig (2) population density of Cucurbit fly on some plants of cucurbitaceous vegetable during spring season ,2022

Sticky traps:

The result of yellow and blue stick traps were applied during the two seasons (Autumn and Spring) to attract *D. cilliatus* then observe the population number, and data are shown in (Fig. 3)

The maximum number of populations was observed at 9th week; while, the minimum was recorded at 2nd week for yellow stick trap. However, the maximum and minimum of population were recorded at 2nd and 11th weeks; respectively, for blue stick. Furthermore, the data of spring season of both colors were very low as the insect appeared rarely between 9-6-2022 and 2-7-2022. Sikandar *et al.,* (2017) found that the most favored color of fruit flies is yellow and transparent in an orange orchard in Pakistan; nevertheless, (Said *et al.,* 2017) observed that the most preferred fruit flies color is yellow and white in a chili garden.

Fig (3): population of cucurbit fly during autumn season, 2021 by yellow and blue stick traps

**Effect of the three Bio-pesticide (Bt., Bb. and *Taget* extract) with different concentrations on 2nd larval instar *Dacus* *ciliatus* in Laboratory:**

In addition, the three bio-pesticide (B. th., B.b and Taget) were tested under laboratory condition (54±2 % relative humidity and 28±2 oC) on 2nd larval instar of *D. ciliatus* and showed a positive mortality number. During the first day no mortality was recorded. However, the maximum mortalities were recorded in the second day by *B. bassiana.* (7g/L) and by the plant extract (15 ml/L) in the third, fourth and fifth days (33.33±6.66, 55.18±9.12, 72.42±6.89 and 82.76±3.44), respectively (Table-2). According to Aemprapa (2007), the two fungal entomopathogens *Metarhizium ansiopliae* (met.) and *Beauveria bassiana* (Bals.) reduced the population of *D. ciliatus* by 50%.

Table (2): Effect of bio-pesticide on mortality percentage of 2nd larval instar *Dacus ciliatus* under Lab. condition

|  |  |
| --- | --- |
| Bio-pesticide | mortality percentage of 2nd larval instar/ day (Mean ± SE) |
| Con. | Day1 | Day2 | Day3 | Day4 | Day5 |
| *B. thuriengiensis* | 1gm | 0.00±0.00 l | 16.66±8.81 j-l | 44.84±6.89 d-h | 50.01±3.84 c-g | 65.39±6.65 a-d |
| 2gm | 0.00±0.00 l | 20.00±5.77 i-l | 51.74±9.12 b-f | 57.71±7.69 b-e | 69.24±13.86 a-c |
| 3gm | 0.00±0.00 l | 30.00±5.77 g-k | 55.18±3.44 b-e | 69.24±3.84 a-c | 80.77±10.17 a |
| *B. bassiana* | 3gm | 0.00±0.00 l | 0.00± 0.00 l | 20.71±3.44 i-l | 38.48±10.17 e-i | 53.86±6.65 b-f |
| 5gm | 0.00±0.00 l | 10.00±5.77 k-l | 27.61±5.96 h-k | 38.48±7.68 e-i | 50.02±13.86 c-g |
| 7gm | 0.00±0.00 l | 33.33±6.66 f-j | 48.29±5.96 c-g | 57.70±3.84 b-e | 65.39±6.65 a-d |
| Tagets | 5ml | 0.00±0.00 l | 23.33±12.01 i-k | 51.73±6.89 b-f | 68.97±5.97 a-c | 72.42±3.44 a-b |
| 10ml | 0.00±0.00 l | 10.00±5.77 k-l | 24.16±3.44 i-k | 62.08±6.89 a-d | 82.76±3.44 a |
| 15ml | 0.00±0.00 l | 23.33±3.33 i-k | 55.18±9.12 b-e | 72.42±6.89 a-b | 82.76±3.44 a |

Means with the same letter are not significantly different at P < 0.05/Duncan test.

**Effect of the three Bio-pesticide (Bt., Bb. and *Taget* extract) on population number of *Dacus* *ciliates* in the field:**

Applying *Bacillus thriengiensis*, *Beauvera bassiana* and *Taget* extract reduced population number of *Dacus* *ciliates* in the field. The maximum reduction (38.46±7.17) was occurred by plant extract and minimum (15.51± 4.54) by *B. thurieingiensis* in the 1st day of application. By the third day, the maximum diminish was observed by the plant extract (67.05 ± 3.84) and the minimum by *B. bassiana* (39.12 ± 7.1). The maximum reduction also was recorded by the plant extract (83.53±1.92) and the minimum by *B. bassiana* (67.05±3.84) in the fifth day (Table-3). The results show that plant extract was more significant to reduce population numbers than *Bacillus thriengiensis* and *Beauvera bassiana.* Khan *et al.,* (2016) observed that *Tagetes minuta*, had a 73% mortality rate in male fruit flies. Hanawi *et al.,* (2016) indicated that *Beauveria bassiana* and *Metarhizium anisophilae* were effective in suppressing *Dacus ciliates*, and *B. bassiana* had higher controlling than *M. anisophilae*.

Table (3): Effect bio-pesticide reduction percentage on adult stage *Dacus ciliatus* in Field

|  |  |
| --- | --- |
| Bio-Pesticide | reduction percentage of adult/ day (Mean ± SE) |
| Day 1 | Day3 | Day5 |
| *B. thuriengiensis* | 15.51±4.54 d | 48.90±5.13 c | 79.22±3.82 a-b |
| *B. bassiana* | 17.58±4.41 d | 39.12±7.10 c | 67.05±3.84 b |
| Tagets | 38.46±7.17 c | 67.05±3.84 b | 83.53±1.92 a |

Means with the same letter are not significantly different at P < 0.05/Duncan test.

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