



## Toxic and biological impact of vegetative parts extract, *Euphorbia tirucalli* L. towards *Ceratitis capitata* (Wiedemann)

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### Abstract

The Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann) is a mainliner pest of fruit orchards worldwide. To improve the control methods depended on natural materials, a laboratory study was carried out to valuation the toxicity and biological parameters of *C. capitata*, adult that applicate to mature stem and leaves of *Euphorbia tirucalli* L. extracted with some solvents such as, acetone, ethanol, petroleum ether, and water. The *Euphorbia tirucalli* extracted with acetone was the more toxicity in comparison with anther solvent, and the LC<sub>50</sub> and LC<sub>90</sub> values were (2.04& 4.09) (1.56 &3.25), and (1.07&3.14) µg/ml post 24,48, and 72h. of treatment. The extract with acetone led to a decline in the oviposition period, hatchability percentage and, adult longevity of males and females of *C. capitata* at LC<sub>50</sub>. The insertion of *E. tirucalli* extract or its components into food could be beneficial in medfly manageable strategies.

**Keywords:** Mediterranean fruit fly, *Euphorbia tirucalli*, leaf extract, *Biological aspects*

### Introduction:

The Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), commonly named medfly, is one of the world's most ruinous pests enclosing both wild and cultivated plants, and it is marked to be the most diffusive of all the Tephritidae (**Zucchi, 2001**). *C. capitata* is multivoltine and highly polyphagous, being able to strafe more than 350 various species of fruits (**McQuate and Liquido, 2017**), consequently triggering quantitative and qualitative wastages to numerous crops. The high reproductive potential and acclimation of *C. capitata* assembled with the low influence of natural enemies, and its wide host range causes considerable disturbance to growers (**Castillo et al., 2000**). To decrease the medfly populations beneath injury threshold, chemical control using insecticide baited sprays is a common method used in most countries (**Urbaneja et al., 2009**). However, the large-scale of chemical products has resulted in insect resistance, and control failures have been reported in field conditions (**Couso-Ferrer et al., 2011**). The excess



omniscience about the disadvantaged outcome derived from the randomized use of insecticides has heartening studies appertained to novel tactics in pest manageable, betwixt them the use of natural products, for their competency and degradability. More than 2000 plant species are known to have potential activity against insects (Souza *et al.*, 2017), through numerous mechanisms including contact toxicity, anti-feed ant, growth inhibitor, suppression of reproductive behavior, and reduction of fecundity and fertility (Tak *et al.*, 2017). With regard, *Euphorbia tirucalli* L. as plant extracted and their components have been attained for their insecticidal activity towards *C. capitata*, adults. This species was chosen for secretes a toxic substance (milk latex) that researchers have proven to be highly toxic to humans and many organisms. Some solvents were tested in the laboratory to evaluate their toxicity and physiological changes on *C. capitata* adults to assess.

## Materials and Methods:

### a. Insect rearing:

The culture of *Ceratitis capitata* was initiated from the pupal stage which obtained from the Plant Protection Research Institute, Agricultural Research Center at Giza, Egypt, and the mass rearing was completed in the Plant Protection Research Laboratory's Zagazig branch- Sharqia, Egypt. The relative humidity (RH) was kept at  $65.5\% \pm 3$  while the temperature of fruit fly colonies was kept constant at  $25.2\% \pm 3^{\circ}\text{C}$ .

The adult flies were reared on an artificial diet described by (Tanaka *et al.*, 1969) was composed of 1 part of protein hydrolysate: 3 parts of sugar and a source of water. The cage size was (30×30×30 cm) and made from plastic with several tiny pores placed within the cage to valeting as oviposition pots contained water inside to keep moisture for eggs. Eggs were collected 2-3 times per week and reposed on an artificial diet in plastic trays covered with cloth lids and left for hatching and larval development, the artificial diet for larval include wheat bran, brewer's yeast, sodium benzoate, HCL, and water, for jumping larvae to pupate, the diet trays were put in a sizable wooden box filled with sand at the bottom (Shehata *et al.*, 2006). To muster pupae, the sand was sifted and relayed to adult rearing cage was made of a wooden frame coated with wire screens from different sides except one side which had a sleeve opening, and the cage floor was made of wooden sheet until the emergence of flies.

### b. Extraction of *Euphorbia tirucalli*:

According to (Su and Horvat, 1981) Vegetative total of *Euphorbia tirucalli* L. were collected and minced into small pieces, and put in an oven for drying at  $50^{\circ}\text{C}$  after that, it was quashed and transferred into plant powder by using small grinder. The plant powder is weighted and steeped in acetone, ethanol, petroleum ether, and water solvents. The extract was filtered and evaporated



at 50°C by the vacuum rotary evaporator separately, then saved in a refrigerator at 18°C until used.

### c. Toxicity indicators:

Plant extracts assayed towards the newly adult of *C. capitata* under laboratory requisites. The different concentrations (0.5, 1, 1.5 and 2%) of each solvent were prepared. Small glass jars were used which each contained ten adult male and ten adult female flies kept separately, and they starved before treatment. Cotton pieces were submerged in a series of five concentrations, with five replicates for each concentration, and five untreated replicates were also put up as a control. Tested flies were examined daily, the dead flies were excluded and the mortality percentage was recorded after 24, 48, and 72 hours.

### d. Biological parameters:

Some biological parameters were observed to evaluate the impact of *E. tirucalli* L. at LC<sub>50</sub> under laboratory conditions. For experiments three replicates were prepared the newly emerged adult flies were starved for 24 h., then the diet was mixed with LC<sub>50</sub> of *E. tirucalli*. Twenty-five pairs of adults (50 males and 50 females) were restricted in each cage. Adults were equipped with water in small plastic (perform the plastic fruits) with small pores as a vessels for oviposition the recorded biological attributes were: egg incubation period (the period from the beginning of the first laid egg to the first egg hatching), fecundity %, hatchability %, larval and pupal duration according to (Murtaza *et al.*, 2021), emergence and, adult longevity.

### e. Statistical analysis:

LC<sub>50</sub> and LC<sub>90</sub> values were calculated using, **Biostat 2007, (professional Build 3200)**, and the toxicity index and relative potency were determined using the formula of **Sun (1950)** and **Zidan and Abdel-Maged (1988)**. **Costat statically computer program (2005)** was used to statistical analysis by one-way ANOVA and the least significant difference (LSD) at (P>0.05).

## Results and Discussions:

### 1- Toxicity impact:

The percentages of mortalities of *Ceratitis capitata* (Wiedemann) were determined after 24, 48, and 72h. of treatments with different solvents such as acetone, ethanol, petroleum ether, at different concentrations of *Euphorbia tirucalli* L.. Results in Table (1) evaluated the lethal concentrations LC<sub>50</sub> & LC<sub>90</sub> towards *C. capitata*, adults treated with *E. tirucalli* by different solvents. The values were (2.04&4.09), (2.60& 5.41), (2.38&4.99), and (3.40&6.21) µg/ml, comparing the toxicity action of the four tested solvents based on the toxicity index acetone solvent was considered as the standard solvent and registering 100%, while, toxicity index for ethanol, petroleum ether and water were (78.46&75.60), (85.71&81.96), and (60.00 & 65.85)%.



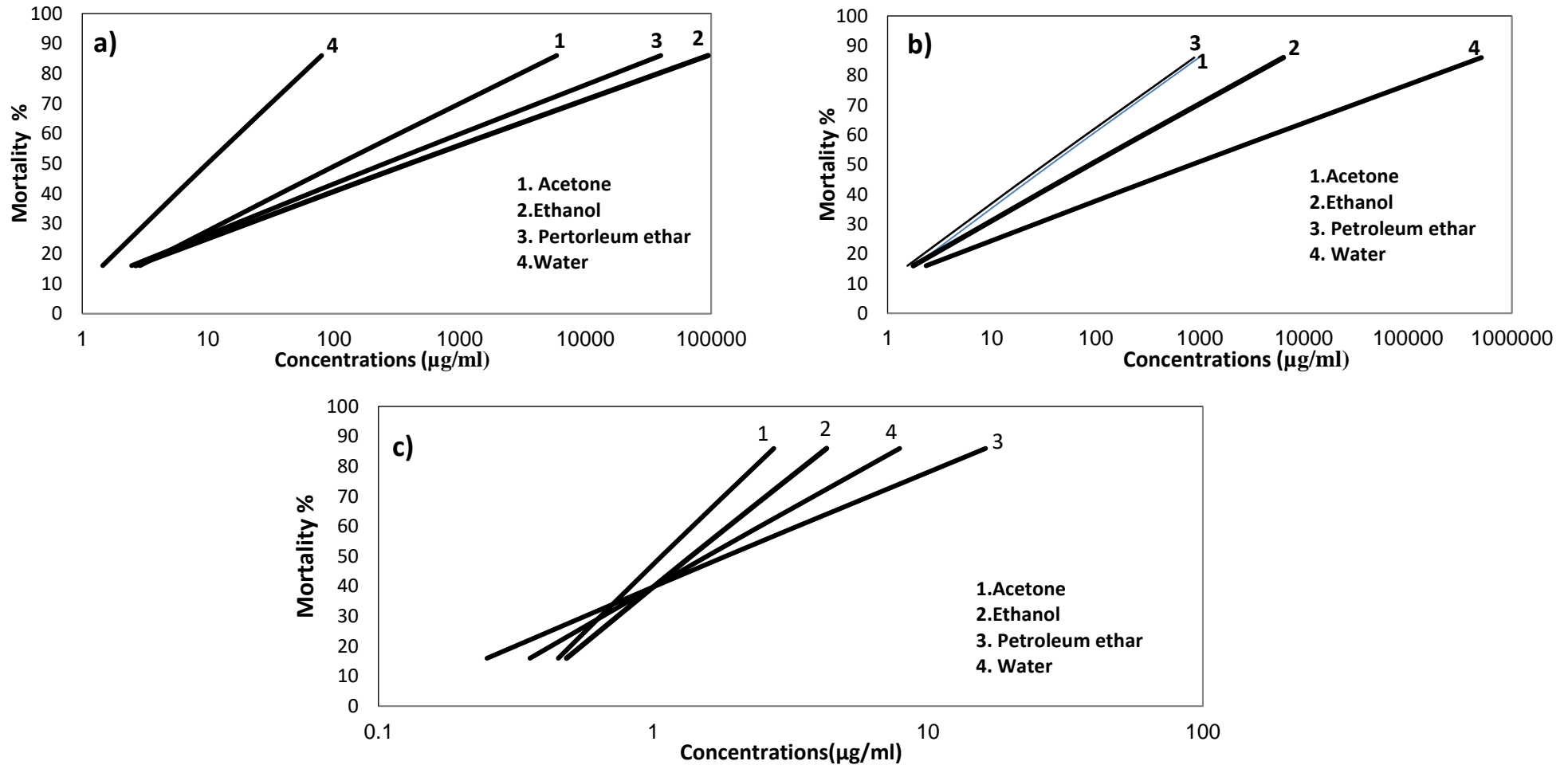
Also, relative potency level can be used as the easiest method in comparing the degree of the toxicity of different materials to any pest. The relative potency levels at  $LC_{50}$  and  $LC_{90}$  of the tested materials are manifested as the number of folds, at the required toxicity levels, compared with the least effective solvent included in the evaluation against the same tested insect. The number of folds idealizing the relative potency level Table (1), where potency level was obtained by dividing the  $LC_{50}$  and  $LC_{90}$  of water, which is considered as the standard compound at  $LC_{50}$  and  $LC_{90}$  levels, by the corresponding figures of each tested solvent. At the  $LC_{50}$  level the relative potency levels expressed as the number of folds indicate that acetone, ethanol, and petroleum ether were effective against the adults of *C. capitata* which recorded (1.66&1.51), (1.30&1.14), (1.42&1.24) fold. Whereas slope values were ( $1.52 \pm 0.30$ ,  $1.48 \pm 0.44$ ,  $0.49 \pm 0.14$ , and  $1.53 \pm 0.68$ ) for acetone, ethanol, petroleum ether, and water, consecutive post 24h of application, Fig. (1a).

Also, results in Table (1) noted that the  $LC_{50}$  and  $LC_{90}$  were relatively close for acetone and petroleum ether after 48h. and the values were (1.56&1.60), and (3.25& 3.30)  $\mu\text{g/ml}$ , consecutive, while ethanol was observed with (1.94& 4.15)  $\mu\text{g/ml}$  (Fig.1b). In contrast, water solvent recorded with the lowest values (3.42&6.72)  $\mu\text{g/ml}$ . with regards, the values of toxicity index at  $LC_{50}$  and  $LC_{90}$  were (80.41&78.31), (97.50&98.48), (45.61&48.36) %, as for relative toxicity, the tested solvents showed these values with (2.19& 2.06), (1.76& 1.61), (2.13&2.03) fold for ethanol, petroleum ether, and water, consecutive. Among the results in the same table, the slope values were showed with ( $0.58 \pm 0.12$ ,  $0.53 \pm 0.37$ ,  $0.75 \pm 0.09$ , and  $1.18 \pm 0.65$ ) for acetone, ethanol, petroleum ether, and water, Fig., (1b). On the country, post 72h. of treatment, the values of  $LC_{50}$  &  $LC_{90}$  for acetone, ethanol, petroleum ether, and water were (1.07&3.14), (1.37&5.28), (1.19&3.27), and (2.70&9.85)  $\mu\text{g/ml}$ . Also, results explained both of toxicity index and relative toxicity which appeared with (78.10& 59.46), (89.91&96.02), and (38.35&31.87) % and (2.60& 3.13), (2.03&1.86), and (2.34&3.01) fold, (Tab.1). In Fig. (1c) illustrated the values of slope with ( $0.92 \pm 0.77$ ,  $0.44 \pm 0.22$ ,  $0.56 \pm 0.62$ , and  $0.97 \pm 0.33$ ). **Priya and Rao (2011)** reported that *Euphorbia tirucalli* has a different medicinal properties: antibacterial, antioxidant, insecticidal, larvicidal, molluscicide, anti-mutagenic, proteolytic, anti-arthritis, anti-herpetic, antifungal activities and hepato-protective effects.



**Table (1): Comparative toxicity of *Euphorbia tirucalli* extracts from different solvents against *Ceratites capitata***

<i>Euphorbia tirucalli</i>	LC <sub>50</sub>	LC <sub>90</sub>	Toxicity index		Relative toxicity		Slope± SE
			LC <sub>50</sub>	LC <sub>90</sub>	LC <sub>50</sub>	LC <sub>90</sub>	
Post 24 hours of treatment							
Acetone	2.04	4.09	100	100	1.66	1.51	1.52±0.31
Ethanol	2.60	5.41	78.46	75.60	1.30	1.14	1.48±0.44
Petroleum ether	2.38	4.99	85.71	81.96	1.42	1.24	0.49±0.14
Water	3.40	6.21	60.00	65.86	1.00	1.00	1.53±0.68
Post 48 hours of treatment							
Acetone	1.56	3.25	100	100	2.19	2.06	0.58±0.12
Ethanol	1.94	4.15	80.41	78.31	1.76	1.61	0.53±0.37
Petroleum ether	1.60	3.30	97.50	98.48	2.13	2.03	0.75±0.09
Water	3.42	6.72	45.61	48.36	1.00	1.00	1.18±0.65
Post 72 hours of treatment							
Acetone	1.07	3.14	100	100	2.60	3.13	0.92±0.77
Ethanol	1.37	5.28	78.10	59.46	2.03	1.86	0.44±0.22
Petroleum ether	1.19	3.27	89.91	96.02	2.34	3.01	0.56±0.62
Water	2.79	9.85	38.35	31.87	1.00	1.00	0.97±0.33



**Fig.(1): Toxicity regression line of *Euphorbia tirucalli* extracts from different solvents against *Ceratites capitata* after, a), 24h.; b), 48h., and c), 72h.post applied.**



## 2. Biological parameters:

Based on  $LC_{50}$  of *E. tirucalli*, extracted with acetone which caused a more toxic impact compared with other solvents, some biological parameters were studied, whereas in Table (2) data indicated that *E. tirucalli*, extract decreased the oviposition periods of females and the average values were  $(25.33 \pm 0.89)$  days compared with the control which given  $54.33 \pm 0.19$  days for adult moth at  $LC_{50}$ , **Thakur and Gupta (2012)** who observed a significantly reduced in delayed and oviposition of fruit fly, *Bactrocera tau* (Walker) post-treatment with ethanol extract of *M. azedarach*, *L. camara*, *A. sativum*, *C. longa* and *neem* formulation by feeding technique at 300ppm compared with control. For the mean number of eggs deposited female moths resulted from treated by *E. tirucalli* led to a decline in fecundity compared with control and the values were  $(212 \pm 1.67$  and  $331 \pm 2.03)$  egg/female, consecutively. On the other hand, data mentioned that the incubation period has a non-significant impact in comparison with the control, whilst, the plant extract has the ability to detract the hatchability percentage with  $(38.05 \pm 1.90)\%$  in contrast with control  $(94.51 \pm 0.91)\%$ , and also led to slightly raised in the larval duration with  $(9.03 \pm 0.67)$  day, at variance with the pupal duration which monitored with non-impact with the control. Else, data monitored that the plant extract detracted in the male and female longevity with  $(33.00 \pm 1.38$  &  $34.31 \pm 3.66)$  counter to the control which noticed with  $(75.42 \pm 2.5$  &  $79.21 \pm 1.83)$  day, consecutively. **Moustafa et al. (2018)** showed that the leaves and stems of Hydroethanolic extracts of *Nerium oleander* caused a prolonged larval period towards the 1<sup>st</sup> instar larvae of *Pectinophora gossypiella* (Suanders). Data also, stated that *E. tirucalli*, extracted caused detracted in the male and female longevity with  $(33.00 \pm 1.38$  &  $34.31 \pm 3.66)$  day, in contrast with the control which noticed  $(75.42 \pm 2.5$  &  $79.21 \pm 1.83)$  day, consecutively. The prolongation in adult longevity may be result of the latent effect of the reduction in food consumption, considerable decrease in growth rate, and obvious reduction in the efficiency of converting ingested and digested food to body tissues which may lead to refuse feeding and died of starvation. **Silva et al. (2013)** observed that the extract of *Azadirachta indica* on *C. capitata* significantly affected the longevity. **Mwin and Van Damme (2011)** assessed that the *E. tirucalli* fresh latex has a toxicity impact on *Anopheles mosquitoes* in the field and established nematode host status of *E. tirucalli* to find out if it is nematocidal or the nematode victim, when investigated the evaluation of pesticide properties of *E. tirucalli* against several pest species.

### Conclusion:

Overall, the research recommend that *Euphorbia tirucalli*, extract as an effective natural material (pesticide) for controlling *Ceratitis capitata* under laboratory conditions.





Parameters  Tested materials	Ovi-position period (day)	Fecundity (egg no./female)	Incubation period (days)	Hatchability %	Larval duration (days)	Pupal duration (days)	Emergence (%)	Longevity	
								Male	Female
Euphorbia tirucalli	25.33 <sup>C</sup> ±0.89	212 <sup>bc</sup> ±1.67	3.10 <sup>b</sup> ±0.47	38.05 <sup>d</sup> ±1.90	9.03 <sup>a</sup> ± 0.67	8.22 <sup>b</sup> ±0.94	92.00 <sup>ab</sup> ±3.03	33.00 <sup>d</sup> ±1.38	34.31 <sup>d</sup> ±3.66
Control	54.33 <sup>a</sup> ±0.19	331 <sup>a</sup> ±2.03	2.45 <sup>b</sup> ±0.33	94.51 <sup>a</sup> ±0.91	7.47 <sup>ab</sup> ±0.43	8.60 <sup>b</sup> ±1.33	100 <sup>a</sup>	75.42 <sup>a</sup> ±2.5	79.21 <sup>a</sup> ±1.83
P	***	**	NS	***	**	NS	*	***	***

Table (2): Impact of extract, Euphorbia tirucalli on some biological parameters against Ceratitis capitata

-Newly emergence moth of a laboratory colony of C. capitata were treated using LC<sub>50</sub>.

- \*\* :highly significant at p<0.01 and \*\*\*:very high significant at p<0.001.





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