

Efficiency of Xanthene Dyes and Their Phototoxicity Against Physiology of Mediterranean Fruit Fly, *Ceratitis capitata* (Wiedemann), (Diptera: Tephritidae)

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Abstract: *Background:* *Ceratitis capitata* (Wiedemann) (medfly), is considered to be one of the most destructive fruit pests because of its high capability to damage the production, its global distribution and its wide range of hosts. *Methods:* In this study stock solution of erythrosine -B was prepared and diluted by distilled water to the following concentrations (0.001 %), (0.01 %), (0.05%) and (0.1%). Groups of ~ 20 laboratory strain survived flies were exposed to white fluorescent neon lamp for 4-h at 10 W/m² fluence rate. *Results:* The percentages of adult mortality were 1.60, 21.10, 58.22 and 78.22% at the concentrations 0.001, 0.01, 0.05 and 0.1%, respectively. Erythrosine-B showed phototoxic effects on *C. capitata* adult flies at 24 h post-exposure to white fluorescent neon lamp for 4- hours at 10 W/m² fluence rate. The mortality percentage of first generation of *C. capitata* adults was 48.47% as compared with 49.34 % in the case of control group (susceptible strain of parent generation).

Key words: Efficiency • Xanthene Dyes • Phototoxicity • *Ceratitis capitata*

INTRODUCTION

Ceratitis capitata (Wiedemann) (medfly), is considered to be one of the most destructive fruit pests because of its high capability to damage the production, its global distribution and its wide range of hosts. Several specific control methods have been developed and applied successfully in many countries against *C. capitata* [1]. Development of chemical insecticides' resistance in pest and vector populations, the damage caused to non-target organisms and the realization of other environmental hazards of these chemicals have led to an increasing interest in biological control measures [2]. The use of photochemical processes as a tool to control the population of several types of insects has been repeatedly examined in both laboratory experiments [3, 4] and field studies [5, 6]. Most investigations have been performed by using photoactive table polycyclic aromatic

dyes that absorb near-UV light wavelengths, including thiophenes, furocoumarines and quinones [7, 8]. The use of xanthene derivatives such as eosin and its analogues absorbing selected light intervals in the visible spectral range has also been proposed [9-11]. All of these dyes require the presence of molecular oxygen to express their phototoxic action; hence the overall photoinsecticidal process appears to be of the photodynamic type [12]. In addition, furocoumarines, upon photoexcitation, can generate various types of addition products with DNA bases, which often results in genotoxic effects [13]., Rebeiz and co-workers proposed the use of porphyrins as photoinsecticides [14]. In particular, these authors tested protoporphyrin IX and its Zn(II) derivative, which appear to be especially promising photoinsecticidal agents since these compounds absorb essentially all the UV-visible wavelengths, that is to say these molecules can be efficiently excited by natural sunlight. Along the same

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line, demonstrated that haematoporphyrin is an efficient phototoxin to several insects [15]. Subsequently, we extended our investigations to some meso-substituted porphyrins [16] in order to identify possible relationships between the chemical structure and the photoinsecticidal activity of this class of compounds. *Ceratitis capitata*, a Mediterranean fruit fly, was selected as an experimental model.

The aim of this study is studying the possible selection of fruit fly for resistance to proposed photoactive dye.

MATERIALS AND METHODS

Rearing Technique of Susceptible Strain *Ceratitis capitata*

Medflies laboratory strain (susceptible strain) was reared at laboratory conditions ($26 \pm 2^\circ\text{C}$, $72 \pm 5\%$ R.H.) in pests physiology laboratory, Plant Protection Institute, Agricultural Research Center, Dokki, Cairo, Egypt for many generations. Newly emerged adults were placed into a wooden cage ($9 \times 9 \times 10 \text{ cm}^3$) with four sides; two sides were covered with screen wire, front side covered with muslin to allow the females to lay eggs. The other opposite side from each cage was provided with a plug to provide the cage with food and water. Pans full of water to 1.5 cm height were surrounding the cage to receive the deposited eggs. Adult flies were provided with a food consisting of sugar and brewer's yeast (3:1) diet in Petri dish. Eggs were collected daily and scattered on the surface of rectangular plastic trays containing larval artificial medium. The larval medium used in the present study consisted of 100 g wheat bran, 25 g brewer's yeast, 30 g sucrose, 0.5 g sodium benzoate and 80 ml tap water; the larval containers were covered with white muslin cloth and tight with a rubber band to ensure maintenance suitable humidity. Infestation of the *Drosophila* spp. and other foreign flies controlled by distributing bottles containing baits consists of agar, yeast, corn (or wheat), molasses and water. This bait was prepared according to the method mentioned [17]. The trays were placed in a wooden cage with sand at the bottom to allow the jumping larvae to pupate [18]. Rearing technique of wild strain *Ceratitis capitata*: A wild Medfly, *C. capitata* flies, was initiated from infested all citrus varieties samples that collected from two Governorates, El-Beheyra (Noubarya zone), El-Giza, during 2020 – 2021, the collected fruits were kept in plastic containers covered with a thin con layer of the fine

sand and kept inside an environmental chamber at about ($26 \pm 2^\circ\text{C}$, $72 \pm 5\%$ R.H), the mature larvae will be jumped out from the infected fruits to the fine sand for pupation. Pupae of the healthy larvae were collected by sieving sands and transferred to the adult screened cages ($9 \times 9 \times 10 \text{ cm}^3$) Emerging Flies were supplied with sucrose and brewer's yeast (3:1) diet in Petri dish with adequate fresh water on sponge for drinking.

Preparations of the Stock Solution of Xanthene Dyes:

Stock solutions of xanthene dyes (Oxford Laboratory, Mumbai, India) were prepared by dissolving a known weight of each erythrosine-B and phloxine-B in a known volume of distilled water. The xanthene dyes concentrations in the final solution was determined by absorption spectrophotometry, using Σ (molar extinction coefficient) which $\Sigma = 4.6 \times 10^4 \text{ dm}^3 \text{ mol}^{-1} \text{ cm}^{-1}$ at 527 nm and $\Sigma = 4.29 \times 10^4 \text{ dm}^3 \text{ mol}^{-1} \text{ cm}^{-1}$ at 537 nm for erythrosine- B and phloxine-B respectively. The solution of xanthene dyes was stable when kept in the dark at 4°C . According to Beer's Lambert law, the measured absorbance is correlated to the concentration of the solution.

The Susceptibility of Laboratory Strain *Ceratitis capitata* to Xanthenes Dyes:

Application in the present study was carried out on 3-7 days. Day before treatment, flies were separated by aspirating and placing them inside cubic (30 cm per side) screened (10 mesh per centimeter) cages. Isolated flies were given only water for 24 h. In adjuvant experiments bait contains various concentrations of Tween-60 without or with photosensitizers were prepared: Four concentrations were prepared for each photosensitizer from (0.001 %), (0.01 %), (0.05 %) and (0.1 %). For each concentration, three cages containing a group of 50 starved flies were prepared. Test bait was left in the presence of flies at dark. The incubation room was kept at $26 \pm 2^\circ\text{C}$, $72 \pm 5\%$ R.H. After 24 h incubation at dark, number of died flies was recorded to determine the dark effect of the selected photosensitizers. Groups of 20 survived flies were exposed to white florescent neon lamp for 4-hours at 10 W/m^2 fluence rate and temperature of $26 \pm 2^\circ\text{C}$. The exposure cages were surrounded by a cooling icebox, to avoid the high temperature of the lamp. The dimensions of the exposure box are (7x 10 x5 cm). The light spot was adjusted to provide the highest degree of light intensity distribution inside the cage [19].

Another set of experiments was conducted using baits contained one concentration equal to the determined LC_{50} of the studied photosensitizers.

Groups of flies fed on the prepared baits were exposed to white fluorescent neon lamp 4 hours, different exposure time intervals.

"Susceptibility determination was started 3 hours and 24 hours post exposure of laboratory strain *C. capitata* adult flies for erythrosine-B and phloxine-B respectively, the alive insects of each cage were returned back to the rearing cage and the dead insects were removed and counted. The mortalities in the control were also estimated. Mortality percentages were corrected for the natural mortality according to Abbott's formula [20].

$$\text{Corrected mortality} = 100(T - C) / 100 - C$$

T = treated mortality

C = control mortality

Another set of experiments treated by four different aforementioned concentrations of selective dye, Mortality results were calculated using the correction of Abbott [20] and the LC_{50} , values of the various generations used to indicate resistance development within successive generations. The increase in resistance was calculated as the ratio between the LC_{50} values of selected strain and that of the parent strain, i.e. $RR = \text{The } LC_{50} \text{ of the successive generation} / \text{The } LC_{50} \text{ of parental strain}$ Where RR = resistance ratio.

Wild Strain: The adult populations of wild *C. capitata* emerged from citrus fruits collected from aforementioned five Governorates treated by previous LC_{50} of selective dye to indicate the susceptibility after 24 hours from exposure to same previous parameter of light intensity fluence rate.

Statistics: All toxicity data were corrected for control mortalities according to Abbott's equation [20]. The LC_{50} and LC_{90} , the fiducial limits at $P = 0.05$ level and the slope of concentration/mortality regression were estimated by probit analysis [21] using a software package "LD-Pline",

Data of the other experiments were evaluated statistically using ANOVA and means compared using T-Test and F-Test at $P < 0.05$). The relationship between the mortality of *Ceratitis capitata* and both different concentrations and different successive 16 generations. All such statistical analyses were done using the software package (SAS, 1992). Results are recorded as mean \pm standard deviation (SD).

RESULTS

The Susceptibility of Laboratory Strain *C. capitata* Adults to Xanthene Dyes

Dark Toxicity: Both xanthene dyes (erythrosine- B and phloxine -B) had no effects on the mortalities *C. capitata* adults in the absence of light.

Light Toxicity:

The Effect of Different Concentrations of Erythrosine- B on the *C. capitata* Adults: Four different concentrations of erythrosine-B were used. In this experiment stock solution of erythrosine -B was prepared and diluted by distilled water to the following concentrations (0.001 %), (0.01 %), (0.05%) and (0.1%). Groups of ~ 20 laboratory strain survived flies were exposed to white fluorescent neon lamp for 4-hours at 10 W/m² fluence rate. Data presented in Table (1). For instance, the percentages of adult mortality were 1.60, 21.10, 58.22 and 78.22% at the concentrations 0.001, 0.01, 0.05 and 0.1%, respectively.

Erythrosine-B showed phototoxic effects on *C. capitata* adult flies at 24 hours post- exposure to white fluorescent neon lamp for 4- hours at 10 W/m² fluence rate. Table 1).

As previously found (in a lab study) the result of this experiment reveals that the phototoxic efficacy of erythrosine- B increases with an increasing in the applied concentrations.

Table 1: Susceptibility of laboratory of strain, *C. Capitata* adults fed on erythrosine- B

Concentration %	Mortality % (Mean \pm SD)	
	Hours Post Exposure	
	3	24
Control	0.0	0.0
0.001	1.60 \pm 2.42 ^a	5.00 \pm 5.01 ^a
0.01	21.10 \pm 2.43 ^b	43.22 \pm 5.537 ^b
0.05	58.22 \pm 7.32 ^c	74.00 \pm 7.34 ^c
0.1	78.22 \pm 7.33 ^d	85.65 \pm 2.59 ^d

Table 2: Susceptibility of laboratory of strain, *C. Capitata* adults fed on phloxine-B

Concentration %	Mortality % Mean	
	Hours Post Exposure	
	3	24
Control	0.0	5.0 \pm 00
0.001	5.20 \pm 5.00 ^a	10.20 \pm 5.35 ^a
0.01	20.20 \pm 10.00 ^b	50.23 \pm 10.53 ^b
0.05	88.48 \pm 5.27 ^c	93.53 \pm 5.25 ^c
0.1	90.20 \pm 5.00 ^d	95.22 \pm 5.10 ^d

Table 3: Lethal doses of erythrosine-B and phloxine- B on *C. capitata*

Lc _s	Concentration of erythrosine-B		Concentration of phloxine-B	
	3 h	24 h	3h	24 h
LC ₅₀	0.0333 (0.0293-0.0378) ^a	0.0149(0.0137-0.0171) ^a	0.018(0.0086-0.027) ^b	0.0096(0.004-0.016) ^b
LC ₉₀	0.2419 (0.1907-0.3223) ^a	0.1329(0.1267-0.1715) ^a	0.092(0.053-0.247) ^b	0.052(0.028-0.125) ^b
LC ₉₅	0.4247(0.3189-0.6015) ^a	0.2475 (0.29-0.338) ^a	0.147(0.077-0.507) ^b	0.083 (0.042-0.248) ^b
Slope	1.4858±0.0820	1.3447±0.0667	1.761±0.346	1.757±0.323

The Effect of Different Concentrations of Phloxine – B on the *C. capitata* Adults: In order to evaluate the phototoxic effect of phloxine-B on *C. capitata* adults, the adults were exposed to white florescent neon lamp for 4-hours at 10 W/m² fluence rate after fed these adults on different concentrations of phloxineB. The mortality was recorded after 3 hours and 24 hours post exposure. Data presented in Table (2); was declared the ascending increasing in percentage of adults mortality at 3 hours post exposure to white florescent neon lamp for 4-hours at 10 W/m² fluence rate. For instance, the percentages of adults mortality were 5.20, 20.20, 88.48 and 90.20 % at the concentrations of (0.001 %), (0.01 %), (0.05 %) and (0.1 %), respectively.

Effect of *C. capitata* Adults to Phloxine-B at LC₅₀: The effect of different exposure time and post-exposure time factors was studied under constant values of other factors using concentration of phloxineB equal to the calculated LC₅₀ (0.0095%). Data tabulated in Table (3) show trends of mortality rate among *C. capitata* flies fed on phloxine-B after 24 hours post exposure to fluence rate of white florescent neon lamp equal to 10 W/m² for 30, 60, 90, 120, 150, 180, 210 and 240 min at 26 ± 1°C. Mortality rate after 24 hours post exposure increased with an increasing of exposure period, reached 50 ± 1.00 % among *C. capitata* exposed for 240 minutes. On other hand, it is quite clear from Table (3) that the mortality of *C. capitata* adults fed on phloxine-B at 0, 24 and 48 hours was non significantly increased at (L.S.D = 2.9561, 3.0598, 3.4073, Respectively) during the exposure period between 30-150 min.

However, there is significant increase in the mean percentage of mortality at exposure time of 180 min and at the same time this value was non significant increased at the time post exposure (0, 24 and 48 hours).

Susceptibility of *C. capitata* Adults Fed on LC₅₀ of Phloxine -B: The effect of selected phloxine -B which consider one of xanthene dyes on the mortality of

C. capitata adults were tested for 16 laboratory strain successive generations of *C. capitata* flies fed on LC₅₀ of phloxine-B after 24 hours post-exposure to fluence rate of white florescent neon lamp equal to 10 W/m² for 4 hours at 26 ± 1°C.

Results obtained in Table (5) the mortality percentage of first generation of *C. capitata* adults was 48.47% as compared with 49.34 % in the case of control group (susceptible strain of parent generation).

The same trend in the percentage of adult mortality was recorded for 13th, 14th and 16th of successive treatment generations as compared with 49.42, 50.93 and 50.77, respectively, in other hand the mortality percentage of 2nd, 4th & 9th successive treatment generations were 50.45, 50.68 and 50.37%, respectively, compared with 49.67, 50.84 and 51.84 %, respectively, for parent susceptible strain generations.

The non-significant decrease in the mean percentage of adult mortality of 1st generation under treatment was 48.47 ± 1.62 as compared to 49.47 ± 1.29 in parent susceptible strain.

Susceptibility of successive generations of laboratory strains of *C. capitata* adults fed on different concentrations of phloxine -B for 24 hours and then exposed to white florescent neon lamp at fluence rate of 10 W/m² for 4 hours after 24 hours post exposure time.

Adult populations of *C. capitata* were fed in the laboratory For 16 generations to different concentrations of phloxine-B [(0.001%), (0.01%), (0.05 %) and (0.1%)] for 24 hours and then exposed to white florescent neon lamp at fluence rate of 10 W/m² for 4 hours after 24 hours post exposure time, in order to determine its susceptibility and select strains resistant to Phloxine-B. The obtained results dealing with this effect are shown in Tables (6-7).

The lethal concentrations of Phloxine -B obtained from successive laboratory strains of *C. capitates* adults which fed on different concentrations of phloxine -B for 24 hours and then exposed to white florescent neon lamp at fluence rate of 10 W/m² for 4 hours after 24 hours post exposure time.

Table 4: Effect of exposure to white florescent neon on phototoxicity of phloxine-B at LC₅₀ on *C. capitata* adults

Exposure time (min.)	Mortality % Mean \pm SD			
	Time Post Exposure			
	Control	0	24	48
30	0.0	01.68 \pm 1.53 ^a	1.68 \pm 1.55 ^a	1.68 \pm 1.54 ^a
60	0.0	01.68 \pm 1.53 ^a	1.68 \pm 1.55 ^a	1.68 \pm 1.54 ^a
90	0.0	2.01 \pm 2.23 ^a	2.01 \pm 2.02 ^a	2.01 \pm 2.01 ^a
120	0.0	3.01 \pm 1.73 ^a	3.34 \pm 2.34 ^a	3.34 \pm 2.32 ^a
150	0.0	3.68 \pm 2.08 ^a	3.68 \pm 2.084 ^a	3.68 \pm 2.082 ^a
180	0.0	10 \pm 1.01 ^b	10.68 \pm 1.161 ^b	11.01 \pm 1.01 ^b
210	0.0	17 \pm 1.01 ^c	25 \pm 2.19 ^c	26.34 \pm 2.32 ^c
240	0.0	25 \pm 2.02 ^d	50 \pm 1.02 ^d	50.34 \pm 2.53 ^d

Table 5: Susceptibility of *C. capitata* adults fed on LC₅₀ of phloxine-B for 24 hours

Generations	% Mortality of parent S.S generations (Mean \pm SD)	% Mortality of Successive treatment generations (Mean \pm SD)
1 st generation	49.47 \pm 1.29 ^a	48.47 \pm 1.62 ^a
2 nd generation	49.67 \pm 3.24 ^a	50.45 \pm 3.51 ^a
3 rd generation	46.68 \pm 2.88 ^a	50.37 \pm 2.45 ^a
4 th generation	50.84 \pm 1.53 ^a	50.68 \pm 2.08 ^a
5 th generation	51.35 \pm 4.06 ^a	49.69 \pm 4.90 ^a
6 th generation	50.30 \pm 2.00 ^a	49.63 \pm 3.37 ^a
7 th generation	49.56 \pm 2.53 ^a	48.96 \pm 0.94 ^a
8 th generation	48.35 \pm 2.08 ^a	47.85 \pm 2.53 ^a
9 th generation	51.78 \pm 2.08 ^a	50.37 \pm 2.577 ^a
10 th generation	48.20 \pm 3.50 ^a	47.82 \pm 2.77 ^a
11 th generation	46.77 \pm 0.58 ^a	46.93 \pm 1.66 ^a
12 th generation	47.87 \pm 5.13 ^a	46.76 \pm 1.53 ^a
13 th generation	49.42 \pm 1.53 ^a	48.99 \pm 1.53 ^a
14 th generation	50.93 \pm 1.15 ^a	48.99 \pm 1.53 ^a
15 th generation	48.99 \pm 0.58 ^a	46.77 \pm 1.53 ^a
16 th generation	50.77 \pm 1.53 ^a	48.45 \pm 2.52 ^a

Table 6: Susceptibility of *C. capitata* adults fed on different concentrations of Phloxine-B

Conc. (%) Generations	% Mortality of Mean \pm SD			
	0.001	0.01	0.05	0.1
Percent	4.40 \pm 0.54	49.06 \pm 2.50	93.71 \pm 1.96	95.91 \pm 1.96
1 st generation	4.58 \pm 0.72	18.75 \pm 2.50	93.75 \pm 1.25	95 \pm 4.51
2 nd generation	3.48 \pm 2.40	50.50 \pm 6.43	49.28 \pm 4.11	95.27 \pm 1.14
3 rd generation	3.67 \pm 0.58	48.00 \pm 2.00	94.67 \pm 1.53	97.67 \pm 2.52
4 th generation	3.74 \pm 2.57	49.67 \pm 9.79	93.53 \pm 2.57	95.58 \pm 2.36
5 th generation	3.77 \pm 1.33	50.15 \pm 4.38	91.88 \pm 3.05	95.65 \pm 2.30
6 th generation	3.67 \pm 0.58	5.00 \pm 2.00	93.33 \pm 1.53	95.33 \pm 2.08
7 th generation	3.33 \pm 3.33	48.89 \pm 8.39	94.44 \pm 3.85	95.56 \pm 1.92
8 th generation	5.56 \pm 1.92	50.00 \pm 3.30	92.22 \pm 5.09	93.33 \pm 3.33
9 th generation	4.17 \pm 1.44	45.83 \pm 3.82	94.17 \pm 3.82	96.67 \pm 1.44
10 th generation	4.67 \pm 3.06	47.33 \pm 3.06	91.33 \pm 5.03	96.67 \pm 4.16
11 th generation	3.33 \pm 2.89	45.00 \pm 5.00	93.33 \pm 2.89	98.33 \pm 2.89
12 th generation	2.22 \pm 1.92	43.33 \pm 3.33	91.11 \pm 5.09	94.44 \pm 1.93
13 th generation	2.22 \pm 1.92	43.33 \pm 3.33	82.22 \pm 13.87	83.33 \pm 12.01
14 th generation	1.11 \pm 1.29	42.22 \pm 5.09	78.89 \pm 10.18	82.22 \pm 13.87
15 th generation	2.00 \pm 2.00	39.33 \pm 9.02	80.00 \pm 8.72	81.33 \pm 13.32
16 th generation	2.00 \pm 2.00	38.67 \pm 8.08	78.67 \pm 9.45	82.00 \pm 5.29

Table 7: The LC₅₀, LC₉₀ and LC₉₅, of Phloxine-B of *C. capitata* adults which fed on different concentrations of Phloxine-B

Generations	LC ₅₀	LC ₉₀	LC ₉₅	Slope ±SE	χ ²	R
Percent (SS)	0.0094(0.0082-0.0106)	0.0472(0.0405-0.0559)	0.0745(0.0625-0.912)	1.8285±0.0814	5.9673	0.9956
1 st generation	0.0095(0.0047-0.01062)	0.0472(0.0405-0.0559)	0.0792(0.0513-0.2297)	1.7846±0.0912	6.4558	0.9933
2 st generation	0.0095(0.0049-0.0158)	0.0496(0.0314-0.1235)	0.0726(0.05-0.1903)	0.8592±0.0739	10.4390	0.9923
3 st generation	0.0095(0.0052-0.0156)	0.0463(0.0305-0.1075)	0.0693(0.0467-0.1738)	1.9187±0.0891	7.1938	0.9945
4 st generation	0.0096(0.0084-0.1090)	0.0448(0.0295-0.0993)	0.0747(0.0622-0.0921)	1.8445±0.0857	5.1841	0.9956
5 st generation	0.0097(0.0086-0.0109)	0.0474(0.0406-0.0566)	0.0789(0.0665-0.0960)	1.8068±0.0773	2.4073	0.9983
6 st generation	0.0095(0.0084-0.0109)	0.0497(0.0429-0.0585)	0.0759(0.0633-0.0935)	1.8322±0.0842	5.3922	0.9955
7 st generation	0.0097(0.0076-0.0122)	0.0481(0.0412-0.0573)	0.0719(0.0528-0.107)	1.8929±0.1601	2.3672	0.9935
8 st generation	0.00970.0074-0.0125()	0.0462(0.0362-0.0662)	0.1082(0.0745-0.177)	1.5724±0.1361	5.2083	0.9931
9 st generation	0.0098(0.0079-0.0119)	0.0636(0.0463-0.0953)	0.0719(0.0549-0.1005)	1.9416±0.1432	1.8710	0.9966
10 st generation	0.0098(0.0081-0.0117)	(0.0463(0.0367-0.0613)	0.0802(0.0623-0.1092)	1.8017±0.1170	13935	0.9986
11 st generation	0.0101(0.0075-0.0132)	0.0504(0.0406-0.065)	0.0657(0.0462-0.1067)	2.0275±0.2156	1.1995	0.9972
12 st generation	0.0118(0.0092-0.0147)	0.0561(0.0430-0.0783)	0.0873(0.0642-0.1305)	1.8908±0.1627	1.3780	0.9969
13 st generation	0.0152(0.0116-0.0195)	0.1137(0.0806-0.1785)	0.2011(0.1334-0.3495)	1.4665±0.1303	4.8497	0.9883
14 st generation	0.017(0.0131-0.0218)	0.125(0.0887-0.1991)	0.2215(0.1462-0.3893)	1.4761±0.1333	5.2614	0.9860
15 st generation	0.0173(0.0056-0.0415)	0.1321(0.09-1.1701)	0.2349(0.1787-3.3316)	1.4523±0.1021	7.3196	0.9896
16 st generation	0.0177(0.0144-0.0214)	0.1335(0.1007-0.1898)	0.2369(0.1689-0.3641)	1.4590±0.1028	5.2496	0.992

Results indicated, that the lethal concentrations values recorded for 50% adult mortality were 0.0095, 0.0095, 0.0096, 0.0096, 0.0097, 0.0096, 0.0097, 0.0097, 0.0098, 0.0098, 0.0101, 0.0118, 0.0152, 0.017, 0.0173 and 0.0177 % for successive generations of laboratory strains (1st, 2nd, 3rd, 4th, 5th, 6th, 7th, 8th, 14th, 9th, 10th, 11th, 12th, 13th, 14th, 15th and 16th) generations, respectively, whereas this value was 0.0094 in parent generation (laboratory susceptible strain).

The same tendency was recorded for at LC₉₀ and LC₉₅ of the tested generations including parent generations as shown in Table (7).

The resistance ratio (RR) at LC₅₀ and LC₉₀ levels of laboratory strain generations of *C. capitata* adults fed on different concentrations of phloxine-B and then exposed to white florescent neon lamp at fluence rate of 10 W/m².

The resistance ratio (RR) and lethal responses for various generations (RR) were calculated from obtained results of both treated successive generations and parent susceptible strain which exposed to white florescent neon lamp at fluence rate of 10 W/m² (Table 7).

DISCUSSION

The photosensitizers investigated in this work have visible absorption properties. Aqueous erythrosine-B and phloxine -B absorbs strongly at 527 nm and 537nm respectively. Different concentrations of both xanthenes dyes which is proved erythrosine- B and phloxine were tested either in dark or under the influence of white florescent lamp for 4- hours at 10 W/m² against the *C. capitata* to determine the best effective concentration of either erythrosine- B or phloxine- B for the photodynamic

killing control of the *C. capitata*. The both xanthenes dyes had no detectable effects on mortality of target insect in dark conditions.

However, our results agree with those obtained by Berni *et al.* [22] in larvae of *C. capitata* which treated up to 7Mm phloxine- B and such observation was also recorded in *Bactrocera zonata* [23] for both xanthenes dyes, also same trend obtained [24] to control population of polyphagous plant pest *Liriomyza bryoniae* fed on xanthenes dyes on the other hand, all the results that will be argued here in signify the mortality percentage of the *C. capitata* on the laboratory strains after 3 h and 24 h from the treatment by either erythrosine- B or phloxine- B under the light condition and a dose response relation was observed between the concentration of these two compounds and fly motility. The two xanthenes dyes evaluated in this study have close values of quantum. Yield (Qc) which were reported 0.6 for erythrosine- B [25] and 0.59 for phloxine- B [26]. Such values of quantum yield are high, enough to exceed the difference in the number of excited photosensitizer molecules and revealed that erythrosine-B and phloxine-B could be an efficient photoinsecticides against *C. capitata* from the photochemical point of view. *C. capitata* were more affected by the highest concentration of either erythrosine- B (78.33 % and 86% mortality respectively, 3 h and 24 h post-exposure to white florescent neon lamp) or phloxine-B (90% and 94.73 % mortality respectively, 3 h and 24 h post-exposure to white florescent neon lamp) as depicted in fig. S. However the calculated LC₅₀ for phloxine- B on *C. capitata* under white neon fluorescent lamp (0.015 % and 0.0095 %) respectively, is much less

than reported for phloxine-B on Mexican fruit fly, *Anastrepha ludens* (0.04%) [27] and more than reported for phloxine-B on peach fruit fly *Bactrocera zonata* (0.0039% and 0.0024%) [19]. Generally, absorption of light by the phototoxic chemicals may induce mortality [28]. According to statistical analysis, the high significant difference toxicity against *C. capitata*. As indicated by their calculated LC_{50s} , the phloxine-B was high significantly toxic to *C. capitata* than erythrosine-B this may be due to the higher concentration. Concentration had a larger number of molecules that accumulated in the insect tissues and sensitize the formation of greater amount of singlet oxygen, that kill the living cells by oxidation of their cellular membranes, than the lower concentration wherever the integument of *C. capitata* is sufficiently thin and transparent to allow passage of visible light to sites containing phototoxic amounts of accumulated photosensitizer [16].

From above discussed results it is deduced that the best photoinsecticidal activity against *C. capitata* as reflected by its LC_{50} s of xanthene dyes was phloxine-B which it was lower than erythrosine-B, so phloxine-B was chosen for further investigations to determine the resistance ratio of laboratory strain successive generations and susceptibility of laboratory strain successive generations based on LC_{50} of phloxine-B. Susceptibility of wild strains from different governorates to LC_{50} of previous chosen xanthenes dye.

Exposure time and post exposure time play an important factor of phototoxicity level of phloxine-B on *C. capitata* using concentration of phloxine-B equal to the calculated LC_{50} (0.0095 %), the exposure time represent the key of extent oxidative stress which depend on the continuous of photosensitization process [29].

In experiments of post-exposure time it was observed that insects continued to die in the dark for further 24 hours following exposure to light, suggesting the existence of a lag in the activity of photoactivated phloxine-B and residual effect of the light exposure. This may reflect the continued action of a photoactivated substance produced during the exposure period or the time required for photosensitized phloxine-B to be translocated and exert its influence at specific site of action. Broome [30], Abdel-Raheem [31] and Salem [32] reported similar continuation of mortality after return of photosensitized insects to darkness following exposure to light. The results of this study revealed that the appropriate point after which mortality assess of phloxine-B efficacy in controlling *C. capitata* is not the end of exposure period to light but at least after 24 hours post exposure.

CONCLUSION

Both xanthenes dyes (erythrosine-B and Phloxine-B) had no detectable effects on mortalities *C. capitata* adults in the absence of light.

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