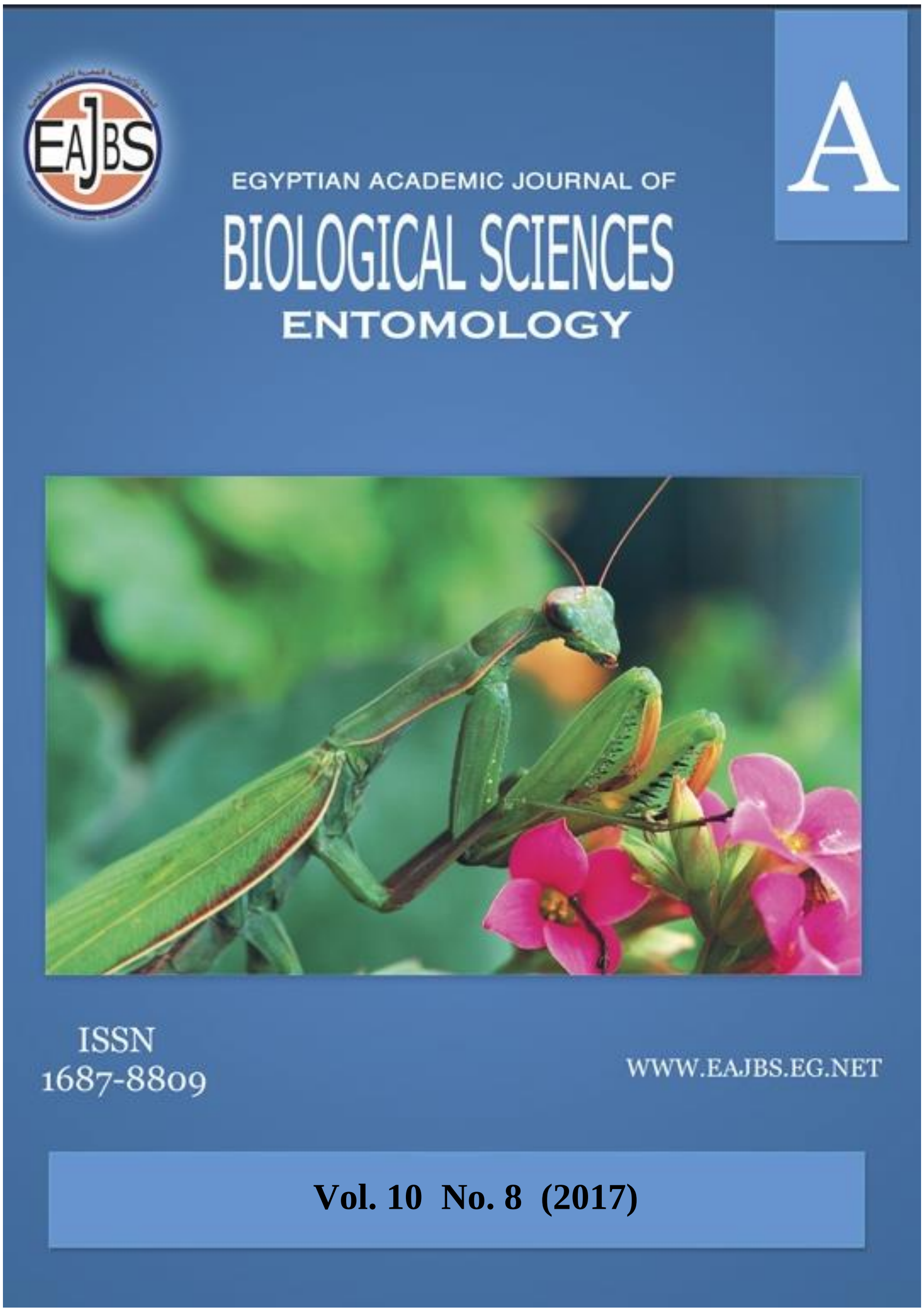

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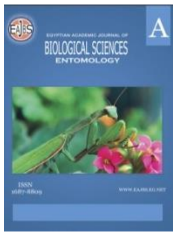

Effect of Chlorophyllin Compound (Photosensitizer) on Main Metabolites Level of *Spodoptera littoralis*, Total Carbohydrates, Total Proteins, and Total Lipid

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ABSTRACT

Photosensitizers are promising and expected to give new technical in agriculture sectors in the future. In this paper, our discussion is focused on photosensitizer and its effects on the main metabolites, total carbohydrates, total proteins, and total lipid were determined in the total body homogenates of *Spodoptera littoralis*. In the present study, the results showed the highest decrease in the total carbohydrate and total lipid. On the other hand, total protein showed lower reduction in most treatments. The photosensitizer copper chlorophyllin was more effective on enzyme activity level of *Spodoptera littoralis* and lightly played an active role in enzyme activity.

INTRODUCTION

Spodoptera littoralis is one of the most serious agricultural pests which is common in Egypt. It is responsible for significant yield losses in many regions of the world. Cotton is infested during the period between May-July following migration from previous host plant clover *Trifolium alexandrinum*. Ishaaya and Klein (1990) found that *Spodoptera littoralis* larvae collected from a cotton field that was heavily sprayed with conventional insecticides showed strong resistance to Organophosphorous and Pyrethroid. The intensive use of broad-spectrum insecticides against *S. littoralis* has led to the development of resistance to many registered pesticides for its control (Aydin and Gu¨ rkan, 2006) including IGRs (Temerak, 2002). Wu *et al.*, (2007) reported that insecticides often show undesirable side effects. For instance, significant toxicity was observed to non-target organisms, such as beneficial insects, fishes or mammals. Human health is also related to wide application of conventional insecticides, because their residues are found in water and different kinds of food and might induce various illnesses. Additionally, in recent years, growers have complained about the failure of control by using these insecticides. Photosensitivity insecticide has attracted increasing attention as a new type of highly efficient and environment friendly pesticide to be used to control the pest due to its rapid photodegradation in the visible light, nontoxic photodegradation products, and phototoxic effects in a variety of biological systems (Ben Amor and Jori 2000). Photosensitization involving light, a photo-sensitizer, and oxygen is a potentially damaging event in biological systems. This generates several reactive oxygen species (ROS) such as singlet oxygen, hydrogen peroxide, superoxide, and hydroperoxyl or hydroxyl radicals that are capable of damaging various sub-cellular structures and molecules (Paillous & Fery-Forgues, 1994; Peiette, 1991; Sies, 1993). Among these, singlet oxygen has been identified as one of the major species responsible for biological damage caused by photosensitization (Weishaput, Goomer, & Dougherty, 1976). The generation of free radicals in skin by solar ultraviolet light (UV) accelerates skin cancer and photo-aging (Witt, Motchink, & Packer, 1993). Formulations of photoactive dyes and baits have been proposed as alternatives for Malathion insecticide for control of major tephritid fruit pests that infest or threaten the United States (Heitz 1995). The major difference between application of photoactive dye and malathion (or other organophosphate) pesticides for fruit flies is that the toxic action of dye formulations functions only if the formulation is consumed by the adult flies, whereas the organophosphates are contact as well as stomach poisons. The concentrations and exposure times of erythrosin B relative to mortality rates of apple maggot, *Rhagoletis pomonella* (Walsh), were modeled in Krasnoff *et al.,* (1994). The appearance of efficient photoinsecticidal agents as possible alternatives to traditional organophosphate and carbamate pesticides may present a new tool to control the population of several types of insects (Rebeiz, *et al.,* 1987; Heitz, 1997; Ben Amor *et al.*, 1998). The main classes of photodynamic sensitizers that have been used as photoinsecticides are the xanthenes, porphyrins, phenol-thiazines, furanocoumarins, thiophenes, and acridines. These agents become active against pest insects in sunlight or artificial light while other pesticides are quickly inactivated by irradiation (Fields *et al.,* 1991; Ben Amor and Jori, 2000). Xanthene dyes and phloxin B have been most extensively studied as pesticides (Ben Amor and Jori, 2000; Heitz 1987). Recently, there has been a major effort to replace soil and foliar insecticides with phloxine B-cucurbitacin bait and reduced doses of toxins for control of corn rootworms in the United States (Schroder *et al.,* 2001). The use of a photochemical process as a new tool to control the population of different types of insects has been frequently examined in both laboratory experiments (Heitz, 1987; Yoho *et al.*, 1976) and field studies (Lenke *et al.*, 1987). Many of these chemicals are considered safe for human contact and ingestion and are registered as additives for drugs and cosmetics (Federal Register 1982, Lipman 1995). Photosensitization involves activation of light-sensitive compounds, producing chemical reactions that damage or destroy cells; in some cases the excited photosensitizer is converted into a toxic photoproduct (Spikes, 1985). Most investigations have been performed using photoactivatable polycyclic dyes that absorb near UV light wavelengths, such as thiophene, furanocoumarins, and quinines (Cunat *et al.*, 1990; Berenbaum and Feeny, 1981), although the use of xanthene derivatives absorbing in selected intervals of visible light has also been proposed (Sakurai and Heitz,1982; Fondren and Heitz, 1978). In many cases, the phototoxic action of the chemical compound is fully developed only in the presence of oxygen; hence the photoinsecticidal actions appear to be of the photodynamic type (Heitz, 1987). In recent years, an increasing attention has been focused on the photosensitizing properties of photosensitizer as insecticides to control several types of insects. Several insects including *Ceratitis capitata, Bactrocera oleae, Stomoxys calcitrans*, and protozoan parasite *Colpoda inflata* have been revealed as sensitive to porphyrin-type compounds when exposed to light (Lukšiené *et al.*, 2007). 5-Aminolevulinic acid (ALA), is a precursor substance of protoporphyrin IX synthesis. The excess ALA through a feedback mechanism inhibits the final step of a biosynthetic pathway. This leads to the accumulation of protoporphyrin IX a well-known photodynamic agent (Rebeiz *et al.*, 1988, 1990), which eventually induces cells to become photosensitive and produce large quantities of $^1O_2$ (Ben Amor and Jori, 2000). $^1O_2$ has high cytotoxicity and can attack the membrane to cause unsaturated lipids, steroids, as well as amino acid residues of protein oxidation and destroy the structure and function of the membranes. In insects, the fat body is the main organ responsible for energetic metabolism. This is also the organ of conversion and storage of fat, carbohydrates, and proteins (Arrese and Soulages, 2010). The fat body is responsible for metabolism of carbohydrates and is especially enlarged in insect larvae chlorophyll is most popularly used as a coloring pigment in the food industry. Its dark green pigment is used to give a range of green shades to the food products. It is used as a commercial green dye in a number of industries like the cosmetics industry, washing soaps, detergents industry, tooth care industry, and the food industry obviously. Chlorophyll has an anti-bacterial property that makes it useful in surgeries, acute rhinitis, ear infections, inflammations, and other problems. Besides, it also has a capability to nullify the various odors, and is therefore, used in pharmaceutical products like ointments etc. It is also used in a lot of chemical processes and analysis. It is consumed as a dietary supplement for its beneficial properties and traits.

MATERIALS AND METHODS

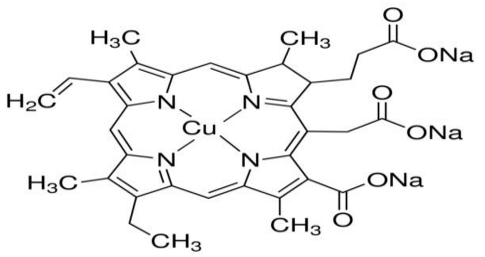
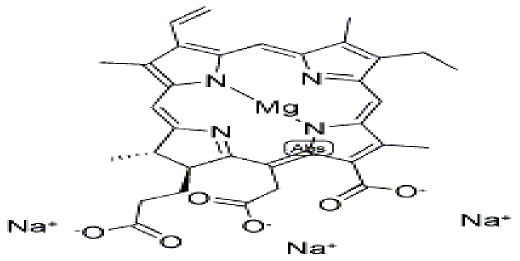
Biological Study:

Insects:

A laboratory strain of the cotton leafworm *Spodoptera littoralis* was obtained from the Central Agricultural of Pesticide Laboratory that is established under constant conditions of 25°C ± 1 and 70 ± 5 % R.H. and out of any contamination with chemicals during the time of study. The strain was reared in the laboratory as described by El-Defrawi (1964) under the previous optimum condition during the experiment.

To establish the toxicity lines of all tested compounds, a castor-bean leaves were dipped for 15 seconds in each concentration of each tested compound then the treated leaves were left for one hour in room temperature to dry. Newly molted 2nd instar larvae were fed on the treated leaves in glass jars (1 lb.) covered with muslin for 24 hrs. After feeding, the treated larvae *Spodoptera littoralis* were exposed to sunlight for 60 minutes and 120 minutes.

Compounds Structure:

Sodium Copper Chlorophyllin		Sodium Magnesium Chlorophyllin	
Chemical Formula	$C_{34}H_{31}CuN_4Na_3O_6$	Chemical Formula	$C_{34}H_{31}MgN_4Na_3O_6$
Molecular Weight	724.15	Molecular Weight	684.91
			

Experimental Design:

Bioassay:

For the detection of the median lethal concentration (LC_{50}) values of photosensitizer (copper chlorophyllin or magnesium chlorophyllin,), a castor-bean leaves were dipped for 15 seconds in each aqueous concentration of the tested compound then were left to dry. The treated leaves were offered to newly molted 2nd instars larvae for 24 hours in dark and exposed to sunlight for different durations (30 minutes, 60 minutes, and 120 minutes). The average of mortality percentage was corrected using Abbott's formula (1925). The corrected mortality percentage of each compound was statistically computed according to Finney (1971). Resistance ratio (RR) is calculated by dividing each LC_{50} values of each treated on that strain with the lower LC_{50} value for each insecticide.

Direct Sunlight:

The treated larvae were exposed to the sunlight for 30 minutes, 60 minutes, and 120 minutes. The fluency rate measured by the dosimeter was taken as the average of intensities during exposure time.

Dark Experiment:

The larvae treated with copper chlorophyllin, magnesium chlorophyllin were left in the dark until the end of larval life.

Biochemical Study:

Preparation of Samples for Biochemical Studies:

The biochemical assay was done after *Spodoptera littoralis* were treated and exposed to different interval to sunlight. The larvae homogenates and after centrifugation the supernatant were used directly for enzyme assay.

Determination of the Main Components:

The main metabolites (total proteins, total lipids, and total carbohydrates) were determined in the total body homogenates.

A-Determination of Total Proteins:

Total proteins were determined by the method of Bradford (1976).

Procedure: Sample solutions 50 µl were pipetted into a test tube and the volume was adjusted to 0.1 ml with phosphate buffer (pH 6.6). 5 ml of protein reagent were added to the test tube and the contents were mixed (inversion or vortexing). The absorbance at 595 nm was measured after 2 min. and before 1 hr against blank prepared from 0.1 ml of phosphate buffer (pH 6.6) and 5 ml of protein reagent. The weight of protein was plotted against the corresponding absorbance resulting in standard curve used to determine the protein in unknown samples

B-Total Carbohydrates:

Total carbohydrates were determined by the method described by Singh and Sinha (1977).

Procedure: Sample solution 100µl was diluted to one ml with H_2O, then 5 ml anthron reagent. A blank containing 1.1ml of H_2O and 5 ml of anthron reagent was placed. All tubes were placed in a boiling water bath for 10 min. then were left to cool for 15 min. at room temperature.

C-Total Soluble Lipids (TSL):

Total lipids were estimated according to Knight *et al.,* (1972) using phosphovanillin reagent**.**

Procedure: Sample solution 250 µl was added to concentrated sulfuric acid (5 ml) in a test tube and heated in a boiling water bath for 10 min. After cooling to room temperature, the digest (500 µl) was added to hosphovanillin reagent (6.0 ml). After 45 min., the developed colour was measured at 525 nm against reagent blank prepared from 500 µl distilled water and 6.0 ml phosphovanillin reagent. The result is expressed as mg lipid/insect.

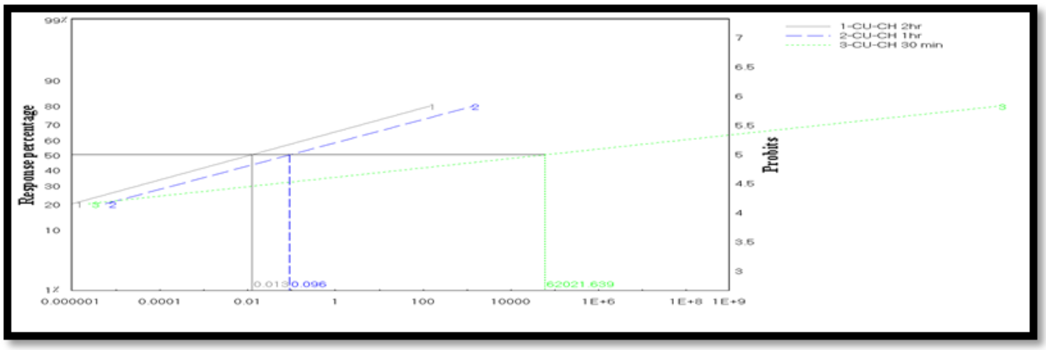
RESULTS AND DISCUSSION

Table (1) and Figure (1) indicated that the tested laboratory strain to the photosensitizer (copper chlorophyllin) exposed to sunlight for two hour was the most susceptible (LC_{50} = 0.013) compared to the other treated and has resistance ratio (RR = 1), and on the other the treated *Spodoptera littoralis* exposed to sunlight for one hour show (LC_{50} = 0.096) and resistance ratio (RR = 7.385) and the treated *Spodoptera littoralis* exposed to sunlight for 30 min. show (LC_{50} = 62021.64) and resistance ratio (RR = 4.77E+06). From these information, we conclude that there is a direct correlation with the exposure time, whenever the exposure time was increased there was an increase in the proportion of death and there is an inverse relationship with the concentration and the exposure time. Fondern and Heitz ,1978 studied the light intensity as a critical parameter in the photodynamic toxicity of rose Bengal to the adult housefly, and showed that the accumulated number of photons needed to kill 50% of population decreased as the intensity increased. This would indicate that there is a regenerative capacity within the insect that is more efficiently overcome by photodynamic action as the light intensity increase. Photosensitization involving light, a photo-sensitizer, and oxygen is a potentially damaging event in biological systems. This generates several reactive oxygen species (ROS) such as singlet oxygen, hydrogen peroxide, superoxide, and hydroperoxyl or hydroxyl radicals that are capable of damaging various sub-cellular structures and molecules (Paillous and Fery-Forgues, 1994).

Table (1): Toxicity data and resistance ratios of copper chlorophyllin against the 2nd larval instars *Spodoptera littoralis* after treated with different exposure time of sunlight (355 w/m^2).

No	Line Name	LC_{50}	LC_{90}	Slope	RR	Index
1	CU-CH 2hr	0.013	21485.29	0.206	1	100
2	CU-CH 1hr	0.096	2.43E+05	0.2	7.385	13.542
3	CU-CH 30 min	62021.64	4.39E+20	0.081	4.77E+06	0.00002

* Copper chlorophyllin = CUCH   * Resistance Ratio (RR) compared with lower LC_{50}



Index compared with CU-CH 2hr   Resistance Ratio (RR) compared with CU-CH 2hr

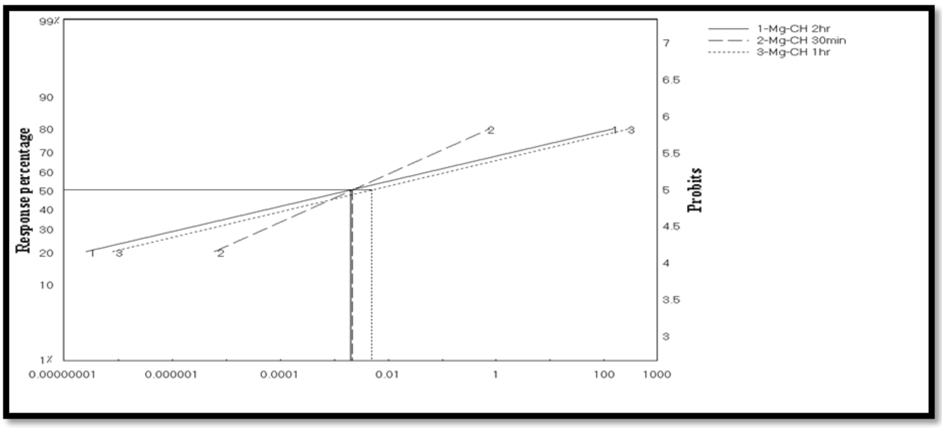
Fig. (1): Log-probit concentration lines of copper chlorophyllin on the 2nd larval instar *S,littoralis* of laboratory strains after treated with different exposure time of sunlight.

Table (2) and Figure (2) indicated that the tested laboratory strain to the photosensitizer (magnesium chlorophyllin) exposed to sunlight for two hour was the most susceptible (LC_{50} = 0.0021) compared to the other treated with different exposure times and has resistance ratio (RR = 1), the other the treated *Spodoptera littoralis* exposed to sunlight for one hour show (LC_{50} = 0.0051) and resistance ratio (RR = 2.429) and the treated *Spodoptera littoralis* exposed to sunlight for 30 min. show (LC_{50} = .0023) and resistance ratio (RR = 1.095).

Table (2): Toxicity data and resistance ratios of magnesium chlorophyllin against the 2^{nd} instars larvae *Spodoptera littoralis* after treated with different exposure time of sunlight (355 w/m^2).

No	Line Name	LC_{50}	LC_{90}	Slope	RR	Index
1	Mg-CH 2hr	0.0021	57787.49	0.172	1	100
2	Mg-CH 30min	0.0023	17.45	0.33	1.095	91.304
3	Mg-CH 1hr	0.0051	1.02E+05	0.176	2.429	41.176

* magnesiumchlorophyllin = MgCH * Resistance Ratio (RR) compared with lower LC_{50}



Index compared with Mg-CH 2hr Resistance Ratio (RR) compared with Mg-CH 2hr

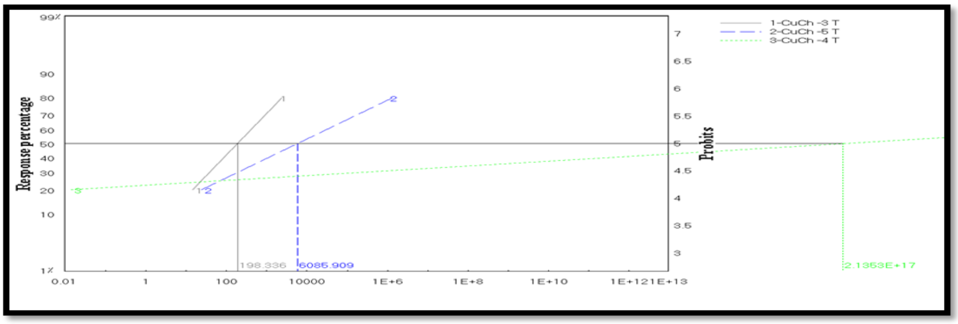
Fig. (2):Log-probit concentration lines of magnesium chlorophyllin on the 2^{nd} larval instars of *S. littoralis* of laboratory strains after treated with different exposure time of sunlight

The time toxicity from Table (3) and Figure (3) indicated that the tested laboratory strain *Spodoptera littoralis* to the copper chlorophyllin exposed to sunlight for two hour show that the concentration 10^{-3} was more susceptible on other treated with concentration 10^{-4} or 10^{-5}, the most susceptible (LT_{50} = 198.336 min.) and has resistance ratio (RR = 1), and on the other concentrations 10^{-4} or 10^{-5} LT_{50} (2.14E+17 min, 6085.909 min), respectively, and resistance ratio RR (1.08E+15, 30.685) respectively. There is a direct correlation with the concentration and mortality in the same exposure time to sunlight. There an inverse relationship with the concentration and the exposure time when increased the concentration this led to decrease exposure time to sunlight. Ben Amor and Jori (2000) reported that several photosensitizing agents, which are activated by illumination with sunlight or artificial light sources, have been shown to be accumulated in significant amounts by a variety of insects when they are administered in association with suitable baits. The subsequent exposure of such insects to UV/visible light leads to a significant drop in survival.

Table (3): Time toxicity data after treated with different concentrations of copper chlorophyllin on the 2nd instars larvae *Spodoptera littoralis* and exposed for two hour of sunlight.

No	Line Name	LT_{50}	LT_{90}	Slope	RR	Index
1	CUCh $X10^{-3}$ T	198.336 min	9912.813 min	0.754	1	100
2	CUCh $X10^{-5}$T	6085.909 min	2.48E+07 min	0.355	30.685	3.259
3	CUCh $X10^{-4}$ T	2.14E+17 min	3.20E+46 min	0.044	1.08E+15	9.29E-14

Index compared with CuCh $X10^{-3}$ T Resistance Ratio (RR) compared with CuCh $X10^{-3}$ T



Index compared with CuCh $X10^{-3}$ T Resistance Ratio (RR) compared with CuCh $X10^{-3}$ T

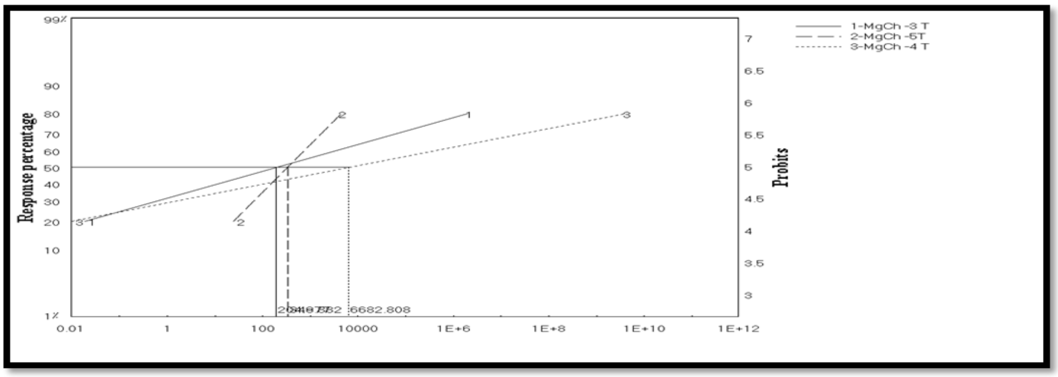
Fig. (3): Log-probit different concentration lines of copper chlorophyllin on the 2nd larvae instar *Spodoptera littoralis* and exposed for two hour to sunlight.

The Time toxicity from Table (4) and Figure (4) indicated that the tested laboratory strain *Spodoptera littoralis* to the magnesium chlorophyllin exposed to sunlight for two hour show that the concentration 10^{-3} more susceptible on other treated with concentration 10^{-4} or 10^{-5}. The concentration 10^{-3} was more susceptible, where (LT_{50} = 204.977 min.) and has resistance ratio (RR = 1), on the other concentration 10^{-4} and 10^{-5} LT_{50} (6682.808min, 346.882 min), respectively. And resistance ratio RR ($4.66E^{+12}$ min., 18485.64 min.), respectively. From these information we obtain that, direct correlation with the concentration and mortality in the same exposure time. Abd El-Naby (2002) reported that the efficiency of photodynamic sensitizers as insecticidal agents is affected by a variety of experimental parameters; and the first factor being photosensitizer concentration. The photoinsecticidal effect steadily increased with increasing the concentration of the photosensitizing. The second factor was the duration of the post treatment light exposure period. There an inverse relationship with the concentration and the exposure time when increased the concentration this led to decrease exposure time to sunlight. The rate of the photosensitized killing of insects appeared to increase with prolongation of post treatment exposure to light, light intensity, an important factor in the mode of action of halogenated xanthene dyes (Fondren and Heitz 1978).

Table (4): Time toxicity data after treated with different concentrations of magnesium chlorophyllin on the 2nd instars larvae *Spodoptera littoralis* and exposed for two hour of sunlight.

No	Line Name	LT50	LT90	Slope	RR	Index
1	MgCh $X10^{-3}$ T	204.977 min	2.71E+08 min	0.209	1	100
2	MgCh $X10^{-5}$ T	346.882 min	18485.64 min	0.742	1.692	59.091
3	MgCh $X10^{-4}$ T	6682.808 min	4.66E+12 min	0.145	32.603	3.067

Index compared with MgCh $X10^{-3}$ T Resistance Ratio (RR) compared with MgCh $X10^{-3}$ T



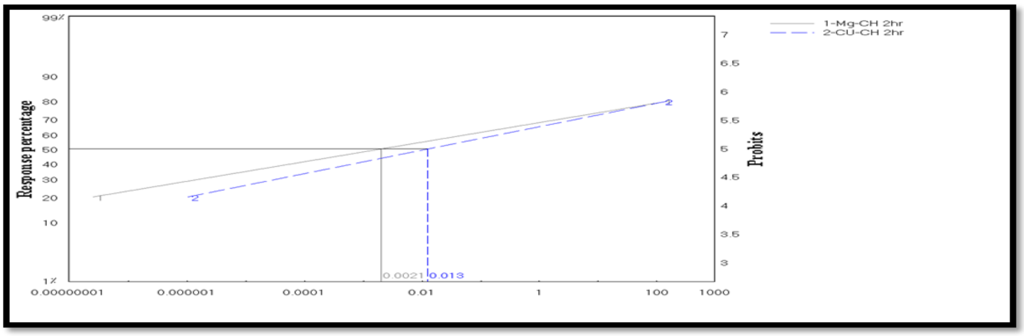
Index compared with MgCh X10^{-3} T   Resistance Ratio (RR) compared with MgCh X10^{-3} T

Fig. (4): Log-probit different concentration lines of magnesium chlorophyllin on the 2nd larvae instars *Spodoptera littoralis* and exposed for two hour to sunlight.

The toxicity data from Table (5) and Figure (5) indicated that the tested laboratory of the 2nd strain *Spodoptera littoralis* treated by copper chlorophyllin or magnesium chlorophyllin exposed for two hour of sunlight, show that the LC_{50} = 0.0021 for magnesium chlorophyllin more effective than LC_{50} = 0.013 for copper chlorophyllin on the same condition. The resistance ratio (RR). (1, 6.19), respectively. That means the magnesium chlorophyllin has more toxicity than copper chlorophyllin.

Table (5): Toxicity data and resistance ratios after treated with LC_{50} of copper chlorophyllin or magnesium chlorophyllin against the 2nd instars larvae *Spodoptera littoralis* and exposed for two hour of sunlight

No	Line Name	LC_{50}	LC_{90}	Slope	RR	Index
1	Mg -CH	0.0021	57787.486	0.172	1	100
2	CU-CH	0.013	21485.293	0.206	6.19	16.154



Index compared with Mg-CH 2hr   Resistance Ratio (RR) compared with Mg-CH 2hr

Fig. (5): Log-probit concentration lines of copper chlorophyllin or magnesium chlorophyllin on the 2nd instars *Spodoptera littoralis* and exposed for two hour to sunlight.

Biochemical Study:

The data summarized in Table (6) represent the changes in total Carbohydrate in the homogenate of the 2nd instars *Spodoptera littoralis*, tested by compound copper chlorophyllin (Cu 10^{-3}) or magnesium chlorophyllin (Mg 10^{-3}) showed reduction in total carbohydrates to most treated *S. littoralis*, the data in table (6) represented, after the 2nd instars *Spodoptera littoralis* exposed to sunlight to one hour after feeding to compound copper chlorophyllin or magnesium Chlorophyllin showed reduction (67.11%, 70.48%) respectively, In total carbohydrates. Also after the 2nd instars

Spodoptera littoralis exposed to sunlight to two hour after treated by compound copper chlorophyllin or magnesium Chlorophyllin showed higher reduction (47.21%, 68.02%) respectively, In total carbohydrates. On the other hand the dark experimental show low reduction in total carbohydrates (85%) in treated with magnesium chlorophyllin and no change in total carbohydrates in treated with copper Chlorophyllin in dark experimental. from this result there is an inverse relationship the decrease in concentration of total carbohydrates occurs when increase the exposure time to sunlight.

Table (6): Percentage of total carbohydrates on *Spodoptera littoralis* homogenate after treated with LC_{50} of photosensitizer (copper chlorophyllin-magnesium chlorophyllin) and exposed to sunlight for different times.

Total Carbohydrates (mg glucose/g b.wt.)						
	Compound	2nd Instars larvae	%	Compound	2nd Instars larvae	%
1	Cu 1h	22.01 ± 0.76	67.11	Mg 1h	23.12 ± 0.63	70.48
2	Cu 2h	15.48 ± 0.80	47.21	Mg 2h	22.31 ± 0.80	68.02
3	Cu Dark	32.95 ± 0.62	100.46	Mg Dark	27.88 ± 0.79	85.01
4	Control	32.80 ± 0.95	100.00	Control	32.80 ± 0.95	100.00

* Copper Chlorophyllin *Magnesium Chlorophyllin * % = percentage relative to control

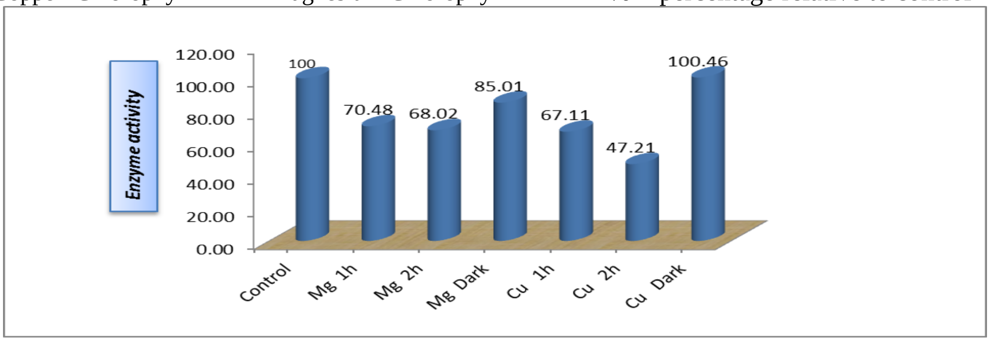


Fig. (6): Percentage of total carbohydrates on spodoptera littoralis treated by photosensitizer (copper chlorophyllin - magnesium chlorophyllin) and exposed to sunlight for different intervals times

In Table (7) the data indicated that, after the 2nd instars *Spodoptera littoralis* after treated with LC_{50} of copper chlorophyllin compound or magnesium chlorophyllin compound and exposed to sunlight for 2hr showed lower reduction in percentage of total protein.

Table (7): Percentage of total protein on *Spodoptera littoralis* homogenate after treated with LC_{50} of photosensitizer (copper chlorophyllin- magnesium chlorophyllin) and exposed to sunlight for different times.

Total Protein (mg protein/ g b.wt)						
	Compound	2nd Instar larvae	%	Compound	2nd Instar larvae	%
1	Cu 1h	46.56 ± 0.83	109.24	Mg 1h	48.55 ± 0.94	113.89
2	Cu 2h	34.84 ± 0.79	81.72	Mg 2h	38.43 ± 0.54	90.15
3	Cu Dark	40.01 ± 0.83	93.85	Mg Dark	46.99 ± 1.19	110.24
4	Control	42.63 ± 1.00	100.00	Control	42.63 ± 1.00	100.00

* Copper Chlorophyllin *Magnesium Chlorophyllin * % = percentage relative to control

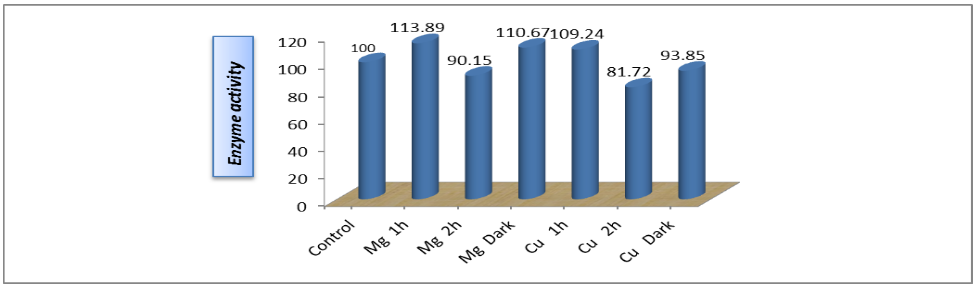


Fig. (7): Percentage of total protein on *Spodoptera littoralis* treated by photosensitizer (copper chlorophyllin - magnesium chlorophyllin) and exposed to sunlight for different intervals times.

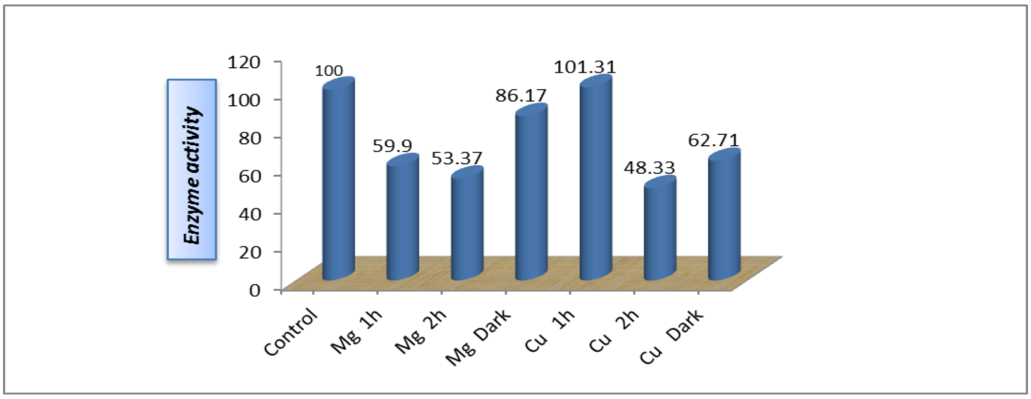
The data summarized in Table (8) represented the changes in total Lipid in the homogenate of the 2nd instars *Spodoptera littoralis*, After the 2nd instars *Spodoptera littoralis* exposed to sunlight for two hour after feeding to compound copper chlorophyllin or magnesium chlorophyllin showed high reduction (48.33%, 53.37%) respectively, In total Lipid. On the other hand, after the 2nd instars *Spodoptera littoralis* exposed to sunlight to one hour after treated by photosensitizer magnesium chlorophyllin showed higher reduction (59.90%) in percentage total Lipid. Photosensitizerare compounds that produce reactive oxygen species (ROS) upon irradiation, (ROS) can in turn cause cell death by oxidizing biomolecules such as proteins, nucleic acids, and lipids (Vatansever *et al.*, 2013).

Table (8): Percentage of total Lipid on *Spodoptera littoralis* homogenate after treated with LC50 of photosensitizer (copper chlorophyllin- magnesium chlorophyllin) and exposed to sunlight for different times.

Total Lipid (mg / g b.wt)						
	Compound	2nd Instar larvae	%	Compound	2nd Instar larvae	%
1	Cu 1h	31.23 ± 0.63	101.31	Mg 1h	18.47 ± 0.71	59.90
2	Cu 2h	14.90 ± 0.79	48.33	Mg 2h	16.45 ± 0.80	53.37
3	Cu Dark	19.33 ± 0.83	62.71	Mg Dark	26.57 ± 0.79	86.17
4	Control	30.83 ± 1.67	100.00	Control	30.83 ± 1.67	100.00

* Copper Chlorophyllin *Magnesium Chlorophyllin ***** % = percentage relative to control

The result from bioassays showed that the highest decrease in the total carbohydrate, total lipid and on the other hand total Protein show lower reduction in most treatments. The efficacy of photoactive compounds as pesticides depends on insect's feeding intensity and ingestion of the dye (Ben Amor and Jori, 2000). The quantitative changes of the total proteins, total carbohydrates, and total lipid. Fat body is the main organ responsible for energetic metabolism. This is also the organ of conversion and storage of fat, carbohydrates, and proteins (Arrese and Soulages 2010).



Fig, (8): Percentage of total Lipid on *Spodoptera littoralis* treated by photosensitizer (copper chlorophyllin - magnesium chlorophyllin) and exposed to sunlight for different intervals times.

A significant reduction in the total lipid levels of the pests treatment by sodium magnesium chlorophyllin or sodium copper chlorophyllin, a significant reduction in the total carbohydrates levels of the target insects and on the other hand low increase in the total proteins level, (Broome *et al.*, 1976) reported that photosensitized insects show large differences from controls as regards weight and levels of protein mass, suggesting the occurrence of a lethal energy stress in the insect. The copper chlorophyllin or magnesium Chlorophyllin is effective in enzyme activity level of *Spodoptera littoralis.* From this result there is an inverse relation, the decrease in concentration of enzyme activity level (total carbohydrates) occurs when increase the exposure time to sunlight. Exposure time to direct sunlight play an active role in enzyme activity of the 2^{nd} instars of *Spodoptera littoralis* larvae to complete photosensitization reaction, the increase of exposure time to sunlight more significant in enzyme activity. It is of great importance to develop new, ecologically safe technologies to control insect pest populations. Photoactive compounds usually used for photosensitization might be effective as pesticide agents, with low impact on the environment, being non-toxic and not mutagenic. Phosensitizer accumulates within the insect body and, following exposure to visible light, induces lethal photochemical reactions and death.

CONCLUSIONS

In this study, we have examined the systemic effects of chlorophyllin compound (photosensitizer) on the second instar larvae of *Spodoptera Littoralis*. Consequently, these studies could open the way for the definition of phototreatment protocols tailored to specific insects and environmental conditions. In this paper we pointed out that, *Spodoptera Littoralis* is sensitive to chlorophyllin compound based photosensitization. It looks like that sodium magnesium chlorophyllin (Mg-CH) is very potential photopesticide against *spodoptera littoralis*, more than sodium copper chlorophyllin (CU-CH) and activated by illumination with sunlight sources, the mortality increased when increasing concentration of photosensitizer and light intensity. The efficacy of photoactive compounds as pesticides depends on insect's feeding intensity and ingestion of the dye. The quantitative changes of the total proteins, total carbohydrates, and total lipid. Fat body is the main organ responsible for energetic metabolism. This is also the organ of conversion and storage of fat, carbohydrates, and proteins. A significant reduction in the total lipid total and carbohydrates levels of the pests treatment by sodium magnesium chlorophyllin or sodium copper. On the other hand, low increase in the total proteins level. The results may be enough to conclude that this chlorophyllin compound can be recommended as photoinsecticide for field studies to control cotton worm *(Spodoptera littoralis)* and this may introduce a possibility to replace the currently used chemical insecticides by this class of compounds. Also the information provided as a consequence of the results obtained in this brief paper can be considered as opening new pioneering avenues for both research and applications.

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ARABIC SUMMARY

تأثير مركبات الكلوروفيلين (الحساسة للضوء) على مستوى التمثيل الكلى للكربوهيدرات، البروتينات الكلية و الدهون الكلية لدودة ورق القطن سبودوبتيرا ليتوراليس

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المركبات ذات الحساسية الضوئية هى مركبات واعدة ومن المتوقع أن تعطي تقنية جديدة في قطاعات الزراعة في المستقبل. في هذا البحث تتركز مناقشتنا على الحس الضوئي والتأثير على التمثيل الغذائى ، الكربوهيدرات الكلية ، البروتينات الكلية، والدهون الكلية .حيث تم دراستها فى دودة ورق القطن (سبودوبتيرا ليتوراليس). في هذه الدراسة لوحظ أن النتائج أظهرت انخفاض مرتفع فى المستوى الكلى للكربوهيدرات والمستوى الكلي للدهون ، ومن ناحية أخرى اظهر مستوى البروتين الكلى انخفاض نسبيا في معظم المعاملات بالكلوروفيلين أكثر فعالية على مستوى نشاط الانزيم من سبودوبتيرا ليتوراليس والضوء تلعب دورا نشطا في نشاط الانزيم