## EFFECT OF FOOD PROCESSES ON SOME INSECTICIDE RESIDUES IN MOLOUKHIA LEAVES

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#### Abstract

The effect of washing and blanching on the behavior and persistence of chlorpyrifos-methyl, diazinon and phenthoate in Moloukhia was investigated. The effect of the insecticides on carbohydrates, chlorophyll and soluble protein contents as well as the activity of peroxidase and polyphenol oxidase were also studied.

The initial deposit (zero time) the residues on moloukhia were 62.70, 115.74 and 160.86ppm for chlorpyrifos-methyl, diazinon and phenthoate respectively. The residues half life values (RL $_{50}$ ) for the same pesticides were 14.65, 13.26 and 13.85 hrs respectively. Washing with tap water after one hour of application removed 54.22% of chlorpyrifos-methyl, 68.98% of diazinon and 71.98 % of phenthoate. The corresponding values after blanching were 79.74, 76.02 and 98.31 % respectively.

The results revealed that all tested insecticides treatment did not show any change in the total content of carbohydrates, total soluble sugars, reducing sugars and non-reducing sugar contents, chlorophyll a, b and total chlorophyll contents, total soluble protein contents and peroxidase activity.

phenthoate treatment significantly increased in total chlorophyll content. Diazinon significantly decreased in the chlorophyll a, b and total chlorophyll contents. Chlorpyrifosmethyl treatment caused a significant decreased in total soluble protein contents in moloukhia leaves.

#### INTRODUCTION

Moloukhia *Chorchorus olitorius* is important crop in Egypt and is widely used by consumers.

Organophosphours insecticides namely chloryrifos-methyl, diazinon and phenthoate are widely used in Egypt to control the economic pests i.e. aphids (*Aphids gossypii*), white fly (*Bemisia tabaci*), and cotton leaf worm (*Spodopetralittoralis*).

The purpose of the present work is to study the behavior and persistence of the insecticides chlorpyrifos-methyl, diazinon and phenthoate in moloukhia under field conditions. The effect of washing and blanching on the amount of residues remaining in the plant after one hour from application with the chemicals was investigated.

The effect of the insecticides on the chemical constituents of moloukhia was further studied.

#### MATERIALS AND METHODS

#### Insecticides used:

#### 1.Chlorpyrifos-methyl (Reldan E.C. 50%):

O, O dimethyl O- (3, 5, 6- tri chloro-2 pyridyl) phosphorothioate.

#### 2. Diazinon (Basudin E.C. 60%):

O,OdiethylO-2 isopropyl-6- methyl pyrimidin-4-yl phosphorthioate.

#### 3. Phenthoate (Cidial E.C.50%):

5- a. ethoxy carbonyl benzyl O, O- dimethyl phosphorodithoate.

Moloukhia *Chorchorus olitorius* was planted in El-Dair village, Aga center, Dakahlia Governorate, Egypt on  $10^{th}$  March 2001 under normal field conditions. The soil in this experiment was sandy clay loam .

The experimental area was divided according to the complete randomized block design including three replicates. The plot was 1/100 feddan. The northern plots were left as control. Moloukhia was sprayed on May 12<sup>th</sup>, 2001. with the three tested insecticides.

The insecticides formulation were diluted with water and sprayed by using a knapsack sprayer equipped with one nozzle. The candidate insecticides were used with the recommended rates 1L/fed.

#### Sampling:

Plant samples were taken at random from each experimental plot. Sub sampling was done at the laboratory, four replicates were taken 50gm for each one. The sample intervals were one hour after application (zero time), 1, 3, 7, 10 and 15 days after application. Clean new polyethylene bags were used for the collected samples and stored at  $20\,^{\circ}\mathrm{C}$  in a deep freezer until analysis.

#### Analytical procedures:

#### Extraction:

Moloukhia leaves were placed in the blender cup (50g) with 50 g sodium sulfate anhydrous and extracted with 200 ml distilled ethyl acetate according to Storherr and Watts (1968). Then blended for three minutes at high speed. The extract was decanted through funnel with a pad of cotton and 30 g sodium sulfate anhydrous into graduated cylinder. A known volume from filtrated (100ml) was taken and then evaporated to dryness on rotary evaporator at  $40\,^{\circ}\mathrm{C}$ .

#### Clean up procedure:

The florisil column clean up procedure of Mills et al. (1972) was used. A18 mm (i.d.)  $\times$  40 cm glass column chromatography was filled with 6 gm of activated florisil (60-100 mesh) and topped with anhydrous sodium sulfate and compact thoroughly.

The column was pre washed using 50 ml n- hexane. The sample extract was dissolved in 10 ml of the same solvent and transferred to the column and then eluted with 200 ml eluant (50 % dichloromethane: 48.5 % n-hexane: 1.5% acetonitrill) at rate of 5 ml /min. The eluant was evaporated to dryness by rotary evaporator at 40  $^{\circ}$ C and the residues were ready for chromatographic determination.

#### Gas liquid chromatography determination:

HP6890 gas chromatograph equipped with flame photometric detector operated in the phosphorus mode (526 nm) filter was used for determination of chlorpyrifos – methyl, diazinon and phenthoate. The column was PAS-1701 (25m x 0.32 mm x 0.52 m $\mu$ ). Injector temperature was 250 °C, detector temperature was 250 °C and Column temperatures were 230, 210 and 230 °C for chlorpyrifos-methyl, diazinon and phenthoate, respectively.

Gases flow rate were 60, 30 and 30 ml/min. for nitrogen, hydrogen and air, respectively. Retention times for chlorpyrifos –methyl, diazinon and phenthoate under these conditions were 1.99, 1.99 and 3.14, respectively.

Rates of recoveries of the insecticides on moloukhia leaves were determined at the level of 1 ppm for the three insecticides the average rate of recovery for the three insecticides were 92, 64 and 92% respectively.

The effect of washing and blanching on removing chlorpyrifos-methyl, phenthoate and diazinon residues from moloukhia leaves after application with recommended rates. Samples were collected after one hour and one day from application and were prepared as follows:

#### Washing with tap water:

Moloukhia leaves samples were rinsed for three minutes with tap water. Then drained on a clean paper for 30 minutes at room temperature until drying. Samples were kept in polyethylene bags under deep freezing until analysis.

#### Blanching of moloukhia leaves:

Samples were placed in a jar filled with boiling water for 2-3 minutes then drained on a clean paper and left until reached to room temperature. Then samples were kept in polyethylene bags under deep freezing until analysis.

#### Determination of carbohydrates fraction contents:

The effect of chlorpyrifos-methyl, diazinon and phenthoate residues on chemical constituents of moloukhia leaves was studied.

Sample consisting of 5 grams plant material were subjected to acid hydrolysis by adding 20 ml of hydrochloric acid 2N in a tube was sealed and put in electrical oven over night at 90°C, after then samples were left to reach to room temperature and filtered on filter paper. The filtrate made to a known volume by distilled water. The

total hydrosable carbohydrates were determined with phenol-sulfuric acid method as described by Hegazy *et al.* (2004).

Total Soluble sugars were extracted from the samples (2 g) in 80 % ethanol (20ml) over night. A known volume was evaporated and dissolved in a known volume of distilled water. Soluble sugars were determined by phenol sulfuric acid as described by Hegazy et al. (2004).

Reducing sugars were determined by picric acid methods mentioned by Hegazy et al. (2004).

The non- reducing sugar contents were calculated by the following equation:

Non-reducing sugar = total soluble sugars - reducing sugar

### Determination of chlorophyll contents:

The fresh samples (5 g) were ground in a porcelain mortar with acetone in presence of little amount of calcium carbonate and acid washed sand. The extract was filtered throughout filter paper, then the mortar residues were washed with acetone several times until the washing liquid was colorless. The extract and washing filtrate were made up to a known volume (50ml). The absorbance was measured at 665 and 649 nm for chlorophyll a and chlorophyll b, respectively, by using Pye Unicam SP1800 spectrophotometer. The chlorophyll was calculated from the following equation as indicated by Vernon (1960)

Chlorophyll 
$$a(mg/g) = \frac{11.63(A(665)-2.18A(649)s)t}{10000 kW}$$
  
Chlorophyll  $b(mg/g) = \frac{20.11(A(649)-5.18(A(665)s)t}{10000 kW}$ 

 $Total\ chlorophyll\ (mg\ /\ g) = Chlorophyll\ a(mg\ /\ g) + Chlorophyll\ b(mg\ /\ g).$ 

#### Where:

V: is the final dilution volume ml. W: weight of sample g.

A: absorbance at 665 and 649 nm for chlorophyll a and b respectively.

### Extraction of enzyme fractions:

Samples were extracted with sodium phosphate buffer (o.2 M, PH 7.9) at  $\overset{\circ}{4}$  C for one hour at the ratio of 1:10 (w/v) as described by Hegazy et al. (2004). The homogenate was filtered through a triplicate layer of chess cloth, and then the supernatant was used for enzyme assay.

Soluble protein fraction was estimated by using the comassie brilliant blue G-250 according to Bradford (1976).

Peroxides were assayed by photochemical method as Hegazy et al. (2004) that adapted to be suitable for determination.

The increasing in absorbance at 430 nm was recorded against blank (with phosphate buffer instead of enzyme extract). One unit of enzyme activity was defined as the amount of enzyme, which cause change in the optical density at 430 nm per minute at 25 °C under standard assay condition. Specific activity was expressed in units

by dividing it on mg protein. The value of  $\Sigma$  at 430 nm for the reaction product of pyrogallol is 2.47 m M<sup>-1</sup> CM<sup>-1</sup>.

mg (Protein) $x\Sigma x0.1$ 

Where AAb: the change in absorbance at 430 nm.

Specific activity =

V: extract volume

Σ: Constant equal 2.47 mM-1CM-1

#### Statistical analysis:

Data were analyzed statistically using the complete randomized block design mentioned by Sendecor and Chachran (1967).

#### **RESLUTS AND DISCUSSION**

The insecticides chlorpyrifos-methyl, diazinon and phenthoate were sprayed on molukhia leaves under normal field conditions at the rate of 1L/fed. The active ingredient used per feddan was 500gramsfor chlorpyrifos-methyl and phenthoate and 600gramsfor diazinon .

The samples were taken at different intervals, 1 hour after application (zero time),1, 3, 7, 10 and 15 days post treatment. The samples were analyzed for insecticide residues. The effect of the insecticides on some chemical constituents and some enzymes activity in addition to the effect of washing and blanching which was also studied.

## 1. Persistence of tested insecticides on and in moloukhia leaves:

Residues of chlorpyrifos-methyl, diazinon and phenthoate on and in moloukhia leaves at different intervals after insecticide application are shown in Table (1). The data show that the concentrations of the initial deposits were 62.70, 115.74 and 160.86ppm after one hour from application, respectively. These amounts decreased to 12.60, 6.91 and 22.43ppm after one day from insecticides application, respectively.

The residues were dissipated to different rates with the elapse of time after 3, 7 and 10 days from application to reach 0.55, 0.07 and 0.008 ppm for chlorpyrifosmethyl, 3.44, 0.57 and 0.17ppm for diazinon and 4.89, 1.09 and 0.20ppm for phenthoate, respectively. No detectable amounts were found from chlorpyrifos-methyl and phenthoate residues at 15 days from application. The data indicated the continuous loss of residues with the elapse of time. The percent loss rate were amounted to 79.90, 99.12, 99.88 and 99.98 % for chlorpyrifos-methyl, 94.03, 97.03, 99.50, 99.86 and 99.97 % for diazinon and 86.05, 96.96, 99.32 and 99.87 % for phenthoate after 1, 3, 7, 10, 15 days from insecticide treatment respectively. The residue half-life values were 14.65, 13.26 and 13.85 hours for the same insecticides on and in moloukhia leaves. These results are in agreement with Hegazy et al. (1997)

a) who found that the RL  $_{50}$  of triazophos on moloukhia leaves was 1.55 days. Shokr (1997) found the RL  $_{50}$  of malathion and actellic were 11.6 and 11.3 hours on moloukhia leaves. Hegazy *et al.* (1999) found the RL  $_{50}$  of fenitrothion was 10.6 hours on moloukhia leaves. Hegazy and Nasr (2003) determined the RL $_{50}$  of diniconazole, fenitrothion, malathion and profenofos were 62.0, 15, 18 and 18 hours in moloukhia leaves.

Table1.Residues of chlorpyrifos-methyl, diazinon and phenthoate on and in moloukhia leaves.

Time after application (days)	Chlorpyri	fos-methyl	Diaz	zinon	Phenthoate		
	ppm	Loss%	ppm	Loss%	ppm	Loss%	
Initial#	62.70 12.60 0.55 0.07	0.00 79.90	115.74	0.00	160.86	0.00	
1			6.91	94.03	22,43		
3		99.12	3.44	97.03	4.89	96.96	
7		99.88	0.57	99.50	1.09	99.32	
10	0.008	99.98	0.17	99.86	0.20	99.87	
15	ND	ND	0.03	99.97	ND	ND	
RL <sub>50</sub> hrs	13.		13.85				

#: One hour after application.ND: No Detected.

Each value is the average of three replicates.

According to Codex Alimentarius Commission (1997) the maximum residue levels (MRL'S) were 0.5, 0.1 ppm for chlorpyrifos-methyl, and diazinon on moloukhia leaves, respectively. The pre-harvest intervals (Safety period for consumption) were 5 and 10days for chlorpyrifos-methyl and diazinon on moloukhia leaves. The safe period of harvesting vegetables treated with the organophosphorus insecticides was ranged between 1 day and 12 days post treatment, which depending on the chemistry of tested insecticide and the kind of crops (Hegazy et al. 1999, Shokr 1997, Shady et al. 2000 and Hegazy and Nasr 2003).

According to Bates (1979) data of pesticide residues in treated crops are required for premarket registration of pesticides and for setting maximum residue limits (toxicologically acceptable level) to protect the consumer against the possible health hazards of exposure to pesticides.

## 2. Removal of tested insecticide residues from treated leaves by some processes:

The frequent use of insecticides to control pests, has led to pollute the environment with toxic residues. The need for removing these residues becomes very important.

In Egypt, consumers usually use water to wash vegetables and fruits before eating.

The efficiency of washing with tap water and blanching process for removing chorpyrifos-methyl, diazinon and phenthoate residues from moloukhia leaves after one hour and one day from treatment was evaluated.

# 2.1. The effect of washing with tap water on pesticide residues in moloukhia leaves:

The results in Table (2) showed the levels in ppm and the percentage of residue removal of insecticides after washing at one hour and one day after application.

Chlorpyrifos-methyl, diazinon and phenthoate residues on and in moloukhia leaves were 62.70, 115.74, 160.86ppm after one hour of insecticides application, respectively. These amounts were reduced to 28.70, 35.90 and 45.21ppm respectively by the washing process. With corresponding percentages of removal were 54.22, 68.98 and 71.89 % respectively after one hour of application. The residues were 12.60, 6.91 and 22.43ppm for chlorpyrifos-methyl, diazinon and phenthoate after one day of application respectively. These amounts were reduced to 2.70, 3.15 and 9.20ppm after washing for three minutes with tap water. Corresponding percentages of removal were 78.57, 54.52 and 58.98 % respectively. Washing procedure used in processing fruits and vegetables either on a commercial or home scale removed 85 to 92 percent of DDT on tomatoes Farrow et al. (1968) and Hegazy et al. (1997b) found that the washing process removed 31.41 % from triazophos residues. Shady et al. (2000) found that the washing process removed 84.34 % from malathion residues on moloukhia leaves.

# 2.2. The effect of blanching process on certain tested insecticide residues in moloukhia leaves:

The results in Table (2) showed the residues of chlorpyrifos-methyl, diazinon and phenthoate on and in moloukhia leaves were 62.70, 115.14 and 160.86 ppm after one hour of application, respectively. These amounts were reduced to 12.70, 24.64 and 2.71ppm after blanching process after one hour from application, the corresponding percentages of removal were 79.74, 78.71 and 98.31 %, after one hour of application. The residues were 12.60, 6.91 and 22.43ppm for chlorpyrifos-methyl, diazinon and phenthoate after one day of insecticides application, respectively. These amounts were reduced to1.55, 2.08 and 6.01ppm by blanching process after one day from application, with the percentages of removal 87.69, 69.91 and 73.20 % respectively. Hegazy et al. (1997 a) reported that blanching process removed about 99.96 and 100 % of triazophos residues on moloukhia leaves and okra fruits, respectively. Shokr (1997) found that the blanching process removed 90.24, 80.07 and 86.28 % from pirimiphos-methyl, fenitrothion and malathion residues on moloukhia

leaves, respectively. Hegazy et al. (1999) showed that boiling process was very effective in eliminated dimethoate residues on and in grape leaves which reached to 98 % loss. Hegazy and Nasr (2003) found that blanching process removed 75.40, 89.29, 73.66 and 75.68 % from diniconazole, fenitrothion, malathion and profenofos residue on moloukhia leaves, respectively. Blanching with hot water or steam of fruits and vegetables may decreased residues depending on temperature used, insecticide involved and whether cooking is done in contact with metal or glass containers.

Table 2. Effect of some different process on certain tested insecticide residues in and on moloukhia leaves, after one hour and one day of spraying.

Time after application		Chlorpyri	fos -methyl	Diaz	tinon	Phenthoate Residues							
	Process	Res	sidues	Resi	dues								
		ppm	%Loss	ppm	%Loss	ppm	%Loss						
Malaukhia	Unprocessed	62.70	0.00	115.74	0.00	160.86	0.00						
Moloukhia leaves after one hour	Washed	28.70	54.22	35.90	68.98	45.21	71.89						
	Blanched	12.70	79.74	24.64	78.71	2.71	98.31						
Moloukhia	Unprocessed	12.60	0.00	6.91	0.00	22.43	0.00						
leaves after one day	Washed	2.70	78.57	3.15	54.52	9.20	58.98						
	Blanched	1.55	87.69	2.08	69.91	6.01	73.20						

## Effect of insecticides on the chemical composition of moloukhia leaves:

## 3.1. Effect on carbohydrate contents in moloukhia leaves:

The results of chlorpyrifos-methyl, diazinon and phenthoate effect on total carbohydrate contents in moloukhia leaves are presented in Table (3). The means of total carbohydrate in moloukhia leaves were 135.53, 156.99, 141 and 133.24 mg/g fresh weight for control, chlorpyrifos-methyl diazinon and phenthoate insecticide treatments, respectively. The results showed no significant effect on total carbohydrate content in moloukhia leaves for chlorpyrifos-methyl, diazinon and phenthoate compared with control at all intervals of study.

Table 3. Effect of tested insecticides on total carbohydrate contents mg/g fresh weight on moloukhia leaves.

Time after application(days)	Control	Chlorpyrifos- methyl	Diazinon	Phenthoate		
Initial <sup># (a)</sup>	92.15	120.93	108.15	134.60		
1	124.77	99.10	115.26	118.16		
3	172.39	87.89	129.78	132.41		
7	157.32	91.42	147.81	150.90		
10	131.01	116.69	187.81	134.21		
15	135.53	156.99	157.51	129.20		
Means±SD	135.53±27.75	156.99±25.67	141±29.57	133.24±10.57		
T Value (calculated)0.05		1.25	- 0.39	0.21		

<sup>\*</sup> One hour after application.

Each value is the average of three replicates.

(-Value): significant increased

## 3.2. Effect on total soluble, reducing and non reducing sugars:

Data in Table (4) reveal that chlorpyrifos-methyl, diazinon and phenthoate did not show any significant effect on soluble sugar contents in moloukhia leaves. The data indicate that no significant decrease in total soluble sugars content in moloukhia leaves for the three insecticides treatment compared with control. The means of total soluble sugars content in moloukhia leaves were 19.56, 13.46, 13.49 and 13.85 mg/g fresh weight for control, chlorpyrifos-methyl, diazinon and phenthoate, respectively. Data showed that the chlorpyrifos methyl treatment had no significant effect on reducing sugars content. On the other hand both of diazinon and phenthoate had no significant increase on reducing sugar contents. Data in Table (4) show that the chlorpyrifos-methyl, diazinon and phenthoate treatments have no significant difference on non-reducing sugars content on moloukhia leaves.

## 3.3. Effect on chlorophyll contents:

As show in table (5) chlorpyrifos-methyl treatment had no significant decrease in the chlorophyll a , b and total chlorophyll compared with the control. On the other hand diazinon significantly decreased the chlorophyll a, b and total chlorophyll.

Table 4. Effect of tested insecticides on total soluble sugars, reducing sugars and non-reducing sugars content on moloukhia leaves (mg/g fresh weight).

	T	T-	1	Т-				Т-	
	Non- reducing	5.81	4.38	5.92	7.94	6.18	7.59	6.30±1.29	1.79
Phenthoate	Reducing	7.54	6.82	7.40	7.84	8.31	7.37	7.55±0.49	- 0.35
	Total	13.35	11.20	13.32	15.78	14.49	14.96	13.85±1.60	1.57
	Non- reducing	4.35	7.17	6.15	7.85	6.79	4.18	6.08±1.51	1.62
Diazinon	Reducing	7.15	7.58	7.92	8.82	5.65	6.87	7.33±1.06	- 0.08
n	Total	11.50	14.75	14.43	16.76	12.44	11.05	13.49±2.19	1.42
hyl	Non- reducing	5.98	5.98	4.14	7.14	6.74	7.60	6.26±1.22	1.75
Chlorpyrifos-methyl	Reducing	5.63	4.64	6.63	10.45	8.49	6.83	7.11±2.08	0.14
ਓ	Total soluble	11.61	10.62	10.77	17.59	15.73	. 14.94	13.46±2.89	1.48
	Non- reducing	25.15	4.36	15.02	3.31	10.92	15.30	12.34±8.09	
Control	Reducing	9.91	6.41	5.11	9.08	5.73	7.35	7.27±1.89	
er do	Total soluble	35.06	10.77	20.13	12.39	16.65	22.38	19.56±8.78	
Time after	application(days)	Initial*	1	8	7	10	15	Means±SD	T Value (calculated)0.05

\* One hour after application. Each value is the average of three replicates. (-Value): significant increased

-	d)	Total	28.64	21.48	25.18	23.26	22.81	20.32	23.62± 2.96	3.84*
	Phenthoate	q	29.67	7.37	11.19	7.41	7.84	6.60	8.35± 1.73	2.31
rt).		а	18,97	14.11	13.99	15.85	14.97	13.72	15.27± 1.97	2.09
fresh weigh		Total	23.39	27.49	22.13	19.88	23.59	19.72	22.70± 2.87	3.99*
ves (mg/g	Diazinon	q	8.34	10.14	8.01	8.98	8.05	6.81	8.39± 1.11	3.49*
loukhia lea		а	15.05	17.35	14.12	10.88	15,54	12.91	14.31± 2.23	3.8*
gnt on mo	ethyl	Total	27.64	21.46	27.37	17.20	30.55	23.34	24.60± 4.86	1.46
) Tresn Wie	Chlorpyrifos-methyl	þ	9.18	7.66	8.83	6.22	10.41	7.43	8.29± 1.48	2.18
ents (mg/g	Chlor	а	18.46	13.80	18.90	10.98	20.14	15.91	16.37± 3.48	0.80
opriyii cong		Total	29.66	28.54	29.14	26.53	24.78	27.70	27.73± 1.81	
es oil cilo	Control	p.	10.24	9.82	10.11	11.80	9.32	10.26	10.26± 0.83	
בת ווואברוורונ		D	19.42	18.72	19.03	14.73	15.46	17.47	17.47± 1.96	
dove 5. Lifect or tested ilisecticides of childrophyli contents (mg/g) fresh wieght on moloukhia leaves (mg/g fresh weight).	Time after application(days)		Initial#	T		7	10	15	Mean±SD	T Value (calculated)0.05

One hour after application, Each value is the average of three replicates. (-Value): significant increased

Table 6. Effect of tested insecticides on specific activity of peroxidase and soluble protein content (mg/g fresh weight) on moloukhia leaves.

	-															
hoate		Soluble protein	6.77	7 33	3	7.33		7.62	8.97		09.9		7.44±	0.84		2.58
Phenthoate		reroxidase	179.41	220.94		55.23		53.14	112.83		276.04	140 00	149.00±	91.13		- 1.03
Diazinon Phenthoat		Soluble protein	6.81	6.34		8.23		8.14	8.93		7.34	7.634	T.00.7	0.97		1.56
Dia	Peroxidace	activity	148.62	128.24		123.47		74.60	68.01		110.31	108 88+	1 00 10	31.58		- 0.44
s-methyl	Soluble protein		Soluble protein 6.92			7.53		7.20	6.44		6.83	6.93±	0,70	01.0	* * * * * * * * * * * * * * * * * * * *	3.31
Chlorpyrifos-methyl	Peroxidase	activity	117.01	91.74		187.67		112.45	157.15		207.46	145.58±	45 93	200	- 7 31	10.3
Control		Soluble protein	8.79	8.08		7.33		9.50	9.00		7.45	8.36±	0.87			
O	Peroxidae	activity	115.14	75.16		175.61	0	82.23	86.98	5	70'10	102.12±	34.05			
Time after application(days)			Initial#	1		m			10,	î	Ç.	Moan+CD		T Value		(calculated)0.05

\* One hour after application. Each value is the average of three replicates. (-Value): significant increased

Data in table (5) showed that the phenthoate have no significant decrease in chlorophyll a, b but the total chlorophyll was significantly decreased by treated with phenthoate The chlorophyll a, b and total chlorophyll contents of moloukhia leaves were 17.47, 10.26 and 27.73 for control, 16.37, 8.29 and 24.65 for chlorpyrifosmethyl, 14. 31, 8.39, and 22.70 for diazinon treatment and 15.27, 8.35and 23.62 mg/g fresh weight for phenthoate treatment, respectively.

## 3.4. Effect on peroxidase activities and soluble protein contents:

Data in Table (6) indicate the chlorpyrifos-methyl, diazinon and phenthoate had no significant increases on peroxidase activity in moloukhia leaves compared with control, while diazinon and phenthoate had no significant decrease on soluble protein contents in moloukhia leaves, but the chlorpyrifos-methyl had a significant increases in soluble protein contents. The peroxidase activities were 102.12, 145.58, 108.88, 149.60 for control, chlorpyrifos-methyl, diazinon and phenthoate treatment, respectively. The soluble protein contents were 8.36, 6.93, 7.63, 7.44 mg/g fresh weight for control, chlorpyrifos-methyl, diazinon and phenthoate treatment, respectively.

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## تأثير عمليات التصنيع على بعض متبقيات المبيدات في أوراق الملوخية

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أجريت هذه الدراسة بهدف دراسة بقاء متبقيات ثلاثة مبيدات فسفورية و هي الكلوربيرفوس-ميثيل و الديازينون و الفينثيويت على أوراق نبات الملوخية و ذلك تحت الظروف الحقلية و تم اخـــذ العينات بعد ساعة من المعاملة، ١، ٣٠، ١٠، ١٠، ١٥، يوم من المعاملة

## و يمكن تلخيص النتائج المتحصل عليها كالأتى :-

٧- أوضحت الدراسة أن عملية الغسيل بماء الصنبور الأوراق الملوخية آدت إلى الإزالية المعاملة على التوالي و الفينثيويت بعد ساعة من المعاملة على التوالي و إزالة. ٧٨,٥٧ ، ٥٤,٥٧ ، ٥٠,٥٧ ، ٥٨,٩٨ ، ٥٨,٩٨ ، ٥٨,٩٨ ، ٥٠,٥٧ ، من متبقيات هذه المبيدات بعيد يوم واحد من المعاملة على التوالي . كما أدت عملية السلق الأوراق الملوخية إلى إزالية ٧٩,٧٧ ، ٧٩,٧١ ، ٨٠,٧١ ، من متبقيات الكلوربيرفوس مثيل و الديازينون و الفينثيويت بعيد سساعة من المعاملة على التوالي. ٩٨,٦١ ، ٩٩,٩١ ، ٧٣,٧٠ من هذه المتبقيات بعد يوم واحد من المعاملة على التوالي. ٩٨,٦٩ ، ٩٩,٩١ ، ٧٣,٧٠ من هذه المتبقيات بعد يوم واحد من المعاملة على التوالي.

٣- أوضحت النتائج إن كل من المبيدات المستخدمة ليست ذات تأثيرات معنوية على محتوى أوراق الملوخية من الكربوهيدرات الكلية و السكرات الكلية الذاتية و المختزلة و المسكرات الغيسر المختزلة وكلوروفيل أ و ب و الكلى و لوحظ ان استخدام مبيد الديازينون أدى لحدوث زيادة معنويــة في محتوى أوراق الملوخية من الكلوروفيل أ و ب و الكلى بينما مبيد الفينثيويت أدى السي حدوث نقص معنوى في محتوى الأوراق من الكلوروفيل الكلى. وأوضحت النتائج إن كـل مــن المبيدات المستخدمة ليست ذات تأثيرات معنوية على محتوى أوراق الملوخية من البروتين الكلــى الــذائب باستثناء المعاملة بمبيد الكلوربيرفوس مثيل أدت الى حدوث نقص في محتوى أوراق الملوخيــة. لــم يكن لهذه المبيدات أي تأثيرات على نشاط إنزيم البروكسيديز.