

**EFFECT OF PROLONGED ORAL ADMINISTRATION OF CARBARYL
ON FERTILITY, LIVER AND KIDNEY FUNCTION OF ALBINO RATS
BY**

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ABSTRACT

Five groups of albino rats were used for studying the toxic effect for prolonged oral administration of carbaryl in dose 1/30, 1/20 and 1/10 LD₅₀ on some biochemical aspects fertility, teratology and haematology. The first group was kept as a control where the other groups were given orally carbaryl dose of 2.45, 3.68 and 7.35 mg/100 g b.wt. The blood serum showed a significant increase in ALT, AST, ALP, bilirubin, creatinine, uric acid, glucose, total lipid and cholesterol but electrolytes, cholinesterase, protein and albumin were significantly decreased. Also, oral administration of carbaryl caused a significant decrease in leucocytic count, erythrocytic count and hemoglobin concentration hematocrite value.

Also, carbaryl caused a significant decrease in testicular weight, concentration of sperm cells, live sperms active motile sperms and abnormal of sperms. Histopathological changes in both liver, kidney and tests appeared to be dose dependant, with damage increase in the high doses.

INTRODUCTION

Pesticides have been widely used in agriculture and their background levels in the environment have increased. Nowadays, pesticides are responsible as a source of many biochemical and physiological disturbance in animals and humans. Numerous *in-vivo* and *in-vitro* studies of possible consequences of treating animal, cell cultures or enzymes systems with these substances have been conducted. In addition pesticides side effect on the environment needed more research.

Kiran *et al.* (1985) demonstrated that a single oral 500 mg/kg dose of carbaryl or seven doses of 71 mg/kg/day increased the activities of acid phosphatase, AST and ALT in the liver and kidneys.

Sobhy (1987) mentioned that giving methyl carbamate insecticide in doses of 2-5 mg/kg b.wt. to rats caused a significant increase in blood glucose

level, serum total lipids and decrease in serum calcium and potassium and increase of serum sodium level.

Ashry (1990) reported that rats treated with the carbamate showed a significant decrease in serum total protein, serum albumin and increase of globulin level due to renal impairment and hepatic insufficiency.

Rahmy (1994) mentioned that toxic effect of repeated exposure to carbamate at different concentrations on rats for 30 days caused a significant increase in serum urea and elevation in serum creatinine level.

Rizk-Alla (1995) found that a significant increase in serum urea and elevation in serum creatinine level caused a toxic effect of carbamate at concentration of 1/10, 1/20 on rats for 90 days.

Dhanapakian and Juliet (1994) reported that exposure of fish to carbaryl for 15 days caused hypertrophy of renal cells, vacuolation and necrosis of renal components. Swilum (1996a) added that high dose of carbaryl caused marked destruction of kidney tissue mainly on proximal convoluted tubules which were partially reversible. Swilum (1996b) reported that the low dose of carbaryl was slight irregularity in the arrangement of the spermatogenic cell layers, however high dose of carbaryl caused marked destruction of spermatogenic cell layers with appearance of vacuoles inside the cytoplasm of the seminiferous tubules of rat testis.

Abad *et al.* (2001) detected the carbaryl residues in fruits and vegetables by monoclonal antibody-based enzyme immunoassay (ELISA) without sample cleanup. The immunoassay is a competitive heterologous ELISA in the antibody-coated format, with an I_{50} value.

The present study was designed to investigate the effect of prolonged oral administration of pesticides (carbaryl) on some serum parameters, fertility and histopathological changes in liver, kidney and testis of rats.

MATERIALS AND METHODS

Materials:

Insecticide used:

Carbaryl (1-naphthyl N-methyl carbamate) or sevin was obtained from Rham opulence Agrachin Company cairo, Egypt.

Animals used:

Albino rats of either sex weighing 120-145 g were fed on normal diets for two weeks for adaptation. Five groups contains ten rats for each one were used for studying the effect of prolonged administration of the insecticides. The first group was kept as a control (untreated), where the other for groups were given orally carbaryl dose of 2.45, 3.68 and 7.35 mg/100 g b.wt. which equal 1/30, 1/20 and

1/10 LD₅₀, respectively for three months. The fifth group (recovery group) received oral treatment with 1/10 LD₅₀ of carbaryl for three months and left another three months without treatment. The carbaryl was given orally using a bulb tipped gastric gavage needle. Each group was biochemically examined every two weeks. At the end of 12th weeks they were sacrificed to detect any histopathological changes of liver, kidney and testis.

Methods:

It is highly indicated to study the toxic effects of the carbaryl as well as to determine the carbaryl LD₅₀ before studying its different pharmacological effects.

- Alanine transaminase (ALT) and aspartate transaminase (AST) were determined by the method of Reitman and Frankel (1957), alkaline phosphatase (Bessey *et al.*, 1964), Bilirubin (Billing *et al.*, 1971).
- Total protein and albumin level (Doumas *et al.*, 1971), serum creatinine (Siest *et al.*, 1985), total cholesterol (Flegg, 1973), blood glucose (Trinder, 1969), serum total lipids (Christopher *et al.*, 1970), blood urea (Patton and Crauch, 1977), serum cholinesterase activity (Ellman *et al.*, 1961), serum sodium and potassium were determined using a flame photometer according to Lucas and Blank (1977). Haemoglobin concentration measurement in whole blood was carried out according to Dacie and Lewis (1986).
- The effect of carbaryl on male rats fertility: The effect of carbaryl on rats fertility was determined at the end of 65 days of oral administration of the tested insecticide. Then the rats were sacrificed for revealing spermatozoal examination, the progressive mobility of sperms and sperm cell concentration were performed according to Bearden and Fluquary (1980).
- Histopathological examination: This examination was carried out at the end of the experiment. Liver, testes and kidney were taken and subjected for microscopic examination as described by Drury and Wallington (1980). Tissue samples were placed in 10% formaline solution according to Harris (1989).
- Statistical analysis: The obtained results were statistically analysed as explained by Snedecor and Cochran (1973).

RESULTS AND DISCUSSION

1. Determination of LD₅₀ of carbaryl:

The toxic effect of carbaryl was studied in rats. A pilot test was carried out on 8 groups of 10 rats for each one. LD₅₀ was determined and the number of animals, doses and mortality in each group are recorded in Table (1). The obtained results indicated that LD₅₀ of carbaryl in rats was 735 mg/kg b.wt.

2. The effect of carbaryl on serum liver enzymes:

The effect of oral administration of carbaryl on serum transaminases activity [alanine transferase (ALT) and aspartate transferase (AST)], alkaline phosphatase (ALP) and bilirubin level are summarized in Table (2). The results showed a significant increase in the ALT, AST, ALP and bilirubin level in carbaryl treated rats after 2, 6, 9 and 12 weeks as compared to control group. The

increase in ALT, AST, ALP and bilirubin were particularly due to tissue damage in liver, kidney and heart (Ray and Podder, 1983; Parafita and Otera, 1984 and Burtis and Ashwood, 1994).

Table (1): Calculation of the LD₅₀ of carbaryl in rats; dose-mortality relationship.

Group No.	Dose In mg/100 gm b.wt.	No. of rats	No of dead rats	Z Mean of dead rats	d Dose difference	Z x d
1	125	10	10	-	15	-
2	110	10	8	9	15	135
3	95	10	8	8	15	120
4	80	10	6	7	15	105
5	65	10	4	5	15	75
6	50	10	2	3	15	45
7	35	10	2	2	15	30
8	20	10	0	1	15	15
						525

$$LD_{50} = Dm - \frac{z \times d}{n} = 125 - \frac{525}{10}$$

$$= 73.5 \text{ mg/100gm b. wt.}$$

$$= 735 \text{ mg/kg b. wt.}$$

3. The effect of carbaryl on kidney function and some serum constituents:

The effect of carbaryl on the levels of serum creatinine, uric acid, electrolytes, glucose, total lipid and cholesterol of male rats are illustrated in Tables (3 & 4). Creatinine is a non-protein nitrogen compound, filtered by the glomerulus of the kidney and used as an indicator of renal function. The results showed a significant uremia during the 9th week of treatment. The elevation of blood urea and creatinine may be attributed to the toxic effect of carbaryl which led to disorders the kidney function, therefore reduced the glomerular filtration rate and consequently retention of urea in the blood (Sonnenwirth and Jarett, 1980 and Saad, 1992).

Serum electrolytes showed significant decrease after oral administration of carbaryl to rats. These results are in agreement with Rahmy (1994) who found that oral administration of thiocarbamate for 4 weeks decreased the serum levels of sodium and potassium.

Serum glucose in rats treated with carbaryl Table (4) indicated a significant increase in serum glucose levels in comparison to control group. These results are in agreement with those obtained by Ray and Podder (1984) who reported that administration of carbaryl to the rats led to hyperglycemia. Wakakura *et al.* (1978) suggested that hyperglycemic in exposure of the rabbits and dogs to carbaryl may be due to increase of sympathoadrenergic activity led to liberation of catecholamines from the adrenal medulla as a result of accumulation of

acetylcholine by the insecticide which affect the hepatic glycogen metabolism. On the other hand Gupta *et al.* (1981) suggested that hyperglycemia may be attributed to the inhibition of glucose metabolism through pancreatitis and decrease insulin activity.

Also Table (4) demonstrates the mean values of serum total lipids and cholesterol. The results showed a significant increase throughout the experiment. These results are in agreement with the results of Rahmy (1994), who suggest that hyperlipidemia and hypercholesterolemia is expected to occur in some diseases of the liver.

4. The effect of carbaryl on serum cholinesterase activity, protein and albumin:

The effect of carbaryl on serum cholinesterase, protein and albumin are summarized in Table (5). The results showed a highly significant decrease in cholinesterase activity. These results are in agreement with those reported by Mount *et al.* (1981) they reported that oral, dermal and inhalation exposure of rats to carbaryl resulted in transient acetyl cholinesterase inhibition.

Regarding the effect of carbaryl on serum protein and albumine (Table, 5). The results showed a significant reduction in total protein and albumin. These results are in agreement with those recorded by Gomes *et al.* (1999).

5. The effect of carbaryl on hematological parameters of rats:

Data in Table (6) indicated that prolonged oral administration of carbaryl in doses of 1/30, 1/20 and 1/10 for, 2, 4, 6, 9 and 12 weeks caused a significant decrease in leucocytic count, erythrocytic count, hemoglobin concentration hematocrite value and blood indices in treated rats. The platelet count showed highly significant decrease in treated rats in comparison to control group. These results are in agreement with those reported by Krug and Berndt (1985).

6. The effect of carbaryl on reproductive parameters of rats:

Male fertility and reproductive system parameter changes are recorded in Table (7). The results showed significant decrease on testicular weight, concentration of sperm cells, live sperms, active motile sperms and abnormal of sperms especially at 1/10 LD₅₀. These results are in agreement with those reported by Makhoul (1993).

7. Histopathological examination of rats:

The effect of carbaryl on regarding the microscopical changes of liver, kidney and testis. The liver showed the cytomegalic changes of the hepatocytes and cytoplasm was granular and vacuolar. Hepatic cell nuclei appeared fragmented chromation. Most of the hepatic blood vessels were dilated. There were also loss of cell architecture and increased degeneration of hepatic cells Fig. (1). These results are in agreement with those obtained by (Baronia *et al.*, 1991) who reported that after 4 weeks of administration of carbaryl on hepatic cells were vacuolated and their nuclei were pyknotic and degenerated. After 5 weeks the degeneration was more pronounced by vacuolated hepatic cells.

Table (2): Effect of carbaryl administration on the serum enzymes and liver functions of the rats (mean±S.D.).

	control	2 weeks			4 weeks			6 weeks			9 weeks			12 weeks			recovery
		1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	
Bilirubin mg/dl	0.32 +0.1	0.48 +0.15	0.82 +0.2	0.63 +0.12	0.53 +0.15	0.79 +0.2	0.96* +0.02	0.69 +0.05	0.89 +0.1	1.03 +0.06	0.89 +0.09	1.05 +0.1	1.1 +0.19	1.09 +0.16	1.23 +0.1	1.45 +0.06	0.75 +0.1
ALT U/L	232.5 +63.5	303.6 +73.5	366.5 +66.8	336.3 +98.4	421.7 +78.0	450 +88.6	465.5 +78.6	280.3 +52.6	317.5 +59.6	328.3 +78.65	317.4 +48.6	371.9* +65.8	403.8 +43.6	461.8 +82.6	503 +55.6	539.2* +44.5	315.6 +73.5
AST U/L	214.1 +65.5	211.3 +78.3	250.0 +56.3	253.2 +45.3	232.4 +35.9	276.2 +35.6	313.5 +65.7	292.5 +45.9	322.1 +63.3	339.6* +58.6	332.6 +48.6	365.3 +35.6	403.5 +66.3	315.5 +53.3	333.5 +58.6	455.3* +90.3	265.6 +56.3
ALP U/L	78.9 +10.2	66.8 +8.6	74.2 +6.8	87.6 +7.9	68.5 +8.6	58.6 +9.5	74.6 +10.3	109.5* +9.8	83.5 +8.6	84.5 +11.3	76.6 +10.5	98.5 +8.6	116.5 +7.66	88.6 +9.6	106 +7.6	136 +12.3	83.6 +9.3

* P = 0.05

** P = 0.01

*** P = 0.001

Where ALT: Alanine amino transferase; AST: Aspartate amino transferase; ALP: Alkaline phosphatase.

Table (3): Effect of carbaryl administration on the kidney function tests and serum electrolytes of the rats (mean±S.D.).

	control	2 weeks			4 weeks			6 weeks			9 weeks			12 weeks			recovery
		1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	
Urea mg/dl	31.6 +1.6	33.6 +5.6	35.6 +2.7	37.9 +4.7	46.9 +6.1	54.6 +3.1	56.2* +2.5	50.4* +6.5	52.6 0.7	58.4 +2.5	53.6 +3.4	57.3 +3.4	62.5 +3.6	55.3* +4.0	60.5 +3.9	65.8* +2.7	48.5 +3.9
Creatinine mg/dl	0.9 +0.1	0.6 +0.15	0.9 +0.1	0.9 +0.09	0.7 +0.1	1.1 +0.06	1.2 +0.12	1.3 +0.15	1.2 +0.099	1.45 +0.35	1.4 +0.4	1.5 +0.06	1.5 +0.4	1.4 +0.08	1.7 +0.05	1.9 +0.06	1.1 +0.2
Uric acid mg/dl	3.8 +0.36	3.0 +0.18	3.0 +0.12	3.1 +0.3	3.2 +0.5	3.5 +0.6	3.7 +0.6	3.8 +1.2	4.3* +0.2	4.5 +0.4	4.0 +0.9	4.2 +1.1	4.8 +1.3	4.3 +0.9	4.5 +1.1	5.2* +0.2	3.9 +0.9
Sodium mEq/l	149.32 +5.3	147.12 +4.68	144.3 +5.33	139.5 +6.1	134.2 +7.33	128.9 +5.33	132.1 +5.03	126.3 +5.33	123.2 +7.22	119.6 +3.44	124.4 +5.12	112.3* +6.33	106.9 +6.22	132 +5.7	105.23* +6.44	102.12* +6.33	124.3 +7.51
Potassium mEq/l	4.8 +0.5	4.7 +0.3	4.5 +0.4	4.2 +0.3	3.9 +0.6	3.5 +0.4	3.2 +0.3	3.4 +0.5	3.2* +0.4	3.0 +0.3	3.9 +0.35	3.7 +0.4	3.7 +0.3	2.9* +0.42	2.75 +0.35	2.6* +0.5	3.9 +0.6

* P = 0.05

** P = 0.01

*** P = 0.001

Table (4) Effect of carbaryl administration on the total lipid, cholesterol and blood glucose of the rats (mean±S.D.).

	control	2 weeks			4 weeks			6 weeks			9 weeks			12 weeks			recovery
		1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	
		Total lipid mg/dl	654.6 ±56.6	538.6 ±45.3	525.36 ±23.6	689 ±19.5	723.6 ±86.5	769.5 ±56.3	700.3 ±12.3	812.3 ±23.5	838.5 ±12.3	802.3 ±53.6	912.3 ±75.3	976.5 ±15.3	865.2 ±86.3	965.3 ±45.3	
Cholesterol mg/dl	143.5 ±24.3	144.5 ±35.2	165.6 ±12.6	179.3 ±14.6	165.3 ±36.3	193.5 ±25.1	212.6 ±43.6	189.3 ±15.6	200.3 ±14.3	253.5 ±10.6	269.3 ±45.6	312.6 ±35.6	235.6 ±15.3	253.9 ±36.5	289.5 ±26.9	200.6 ±43.5	
Glucose mg/dl	143.5 ±7.7	145 ±16.2	139 ±6.6	145.5 ±23.2	178.5 ±21.7	240.6 ±3.1	278.5 ±6.7	222.5 ±5.9	263.5 ±23.5	312.8 ±6.2	255.6 ±13.2	253.5 ±9.5	235.5 ±8.2	289.5 ±15.3	300.2 ±7.5	180.5 ±15.6	

* P = 0.05 ** P = 0.01 *** P = 0.001

Table (5) Effect of carbaryl administration on the plasma proteins and acetyl cholinesterase activity of the rats (mean±S.D.).

	control	2 weeks			4 weeks			6 weeks			9 weeks			12 weeks			recovery
		1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	
		Tot. Protein gm/dl	6.37 ±1.46	6.79 ±1.28	6.77 ±1.21	6.09 ±1.13	6.17 ±1.05	5.93 ±1.09	5.89 ±1.16	5.89 ±0.97	5.74 ±0.94	5.19 ±0.82	5.49 ±0.84	5.12 ±0.72	4.77 ±0.81	5.1 ±0.76	
Albumin gm/dl	4.39 ±1.18	4.26 ±1.12	4.16 ±1.34	3.96 ±1.09	4.03 ±1.168	3.87 ±1.26	3.59 ±1.13	3.47 ±0.85	3.23 ±0.76	3.08 ±0.72	3.21 ±0.73	2.92 ±0.63	2.7 ±0.53	2.7 ±0.57	2.48 ±0.56	2.42 ±0.46	3.37 ±1.09
CHE U/L	569.3 ±112.3	451.3 ±119.6	476.3 ±89.4	431.3 ±99.5	400.5 ±109.6	368.6 ±79.6	334.3 ±125.6	298.3 ±136.8	278.3 ±103.6	255.3 ±98.6	296 ±125.3	256 ±115.6	245 ±96.3	286.6 ±113.9	233.2 ±63.2	212.9 ±76.9	613.9 ±112.6

* P = 0.05 ** P = 0.01 *** P = 0.001

Where CHE : Acetyl cholinesterase

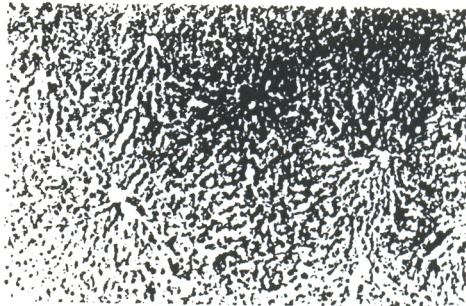
Table (6) Effect of carbaryl administration on hematological parameters of rats (mean±S.D.).

	control			2 weeks			4 weeks			6 weeks			9 weeks			12 weeks			recovery
	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	1/30	1/20	1/10	
Hb mg/dl	16.13 +0.66	15.72 +0.45	14.31 +0.47	13.6 +0.61	11.96 +1.92	11.28 +0.66	10.07 +1.54	10.74 +0.68	9.74 +0.47	7.89 +1.34	7.29 +1.32	6.60 +2.32	6.2 +0.24	6.76 +1.06	6.45 +1.9	6.2 +0.24	6.76 +1.06	6.45 +1.9	13.22 +1.48
RBCs 10 ⁶ cell/mm	5.43 +1.23	4.89 +0.43	5.11 +0.14	5.8 +0.15	4.79 +0.14	3.94 +0.66	3.6 +0.2	4.4 +0.7	3.02 +0.2	3.34 +0.28	2.8 +1.24	2.54 +0.54	2.08 +0.45	2.75 +0.5	2.93 +0.83	2.08 +0.45	2.75 +0.5	2.93 +0.83	4.05 +0.53
Hct %	48 +2.3	46 +2.1	43 +2.3	40 +1.8	40 +1.6	36 +1.9	31 +3.2	30 +1.9	28 +2.32	23 +3.23	21 +2.41	19 +1.6	16 +1.36	16 +1.22	16 +1.2	16 +1.36	16 +1.22	16 +1.2	40 +3.21
MCV fl	89 +5.32	87 +5.44	87 +4.79	83 +3.88	86 +6.9	87 +6.41	81 +2.89	79 +5.32	77 +4.3	74 +5.12	78 +4.98	76 +4.63	83 +4.44	84 +4.26	72 +7.56	83 +4.44	84 +4.26	72 +7.56	69 +4.35
MCH pg	29.7 +2.03	28.3 +1.69	28.03 +2.36	23.35 +2.3	29.2 +1.6	28.36 +1.8	26.6 +1.3	24 +1.7	25.3 +2.1	24.3 +2.2	26.3 +1.6	25 +2.3	29.3 +2.54	27.35 +2.66	22.3* +2.15	29.3 +2.54	27.35 +2.66	22.3* +2.15	27.3 +1.98
WBCs 10 ³ cell/mm	5.99 +0.29	5.38 +0.55	6.6 +0.25	7.05 +0.12	4.7 +0.49	3.09 +0.04	6.38 +0.69	5.75 +0.28	4.55 +0.36	7.2 +0.62	7.33** +0.27	6.04 +0.29	3.13 +0.63	4.5 +0.17	4.32 +0.54	3.13 +0.63	4.5 +0.17	4.32 +0.54	5.2 +1.23
Platelets 10 ³ cell/mm	450.2 +98.33	453.2 +69.0	430.36 +115.0	420.15 +123.0	410.6 +143.1	385.21 +112.6	362.5 +142.6	340.2 +152.3	322.2 +115	300.25 +98.3	280.66* +96.3	262 +99.6	234.2 +75.56	253.1 +113.53	232.22 +132.2	234.2 +75.56	253.1 +113.53	232.22 +132.2	364.2 +100.2

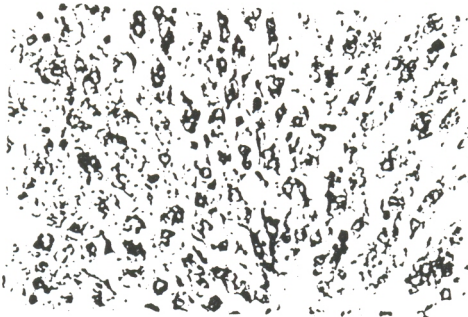
* P = 0.05

** P = 0.01

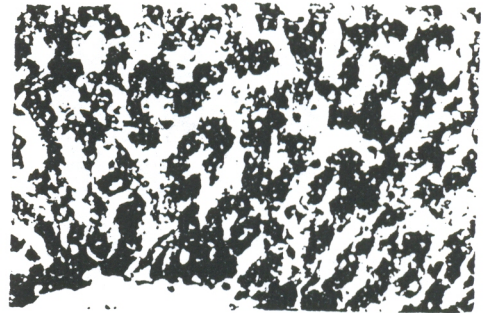
*** P = 0.001



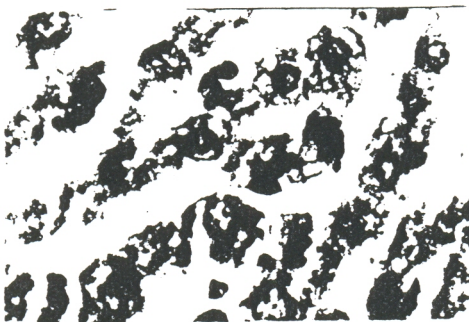
(a) A photomicrograph of normal liver section of adult albino rat of the control group showing Central vein (C) hepatic cord (H), radiating from central vein. (*Haematoxylin & Eosin X 250*)



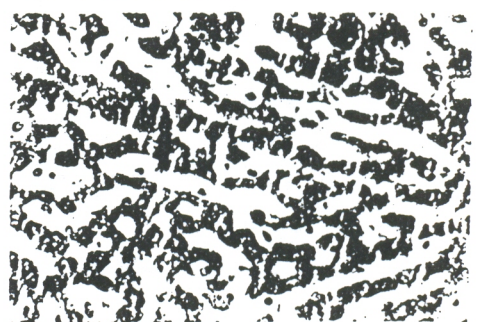
(b) A photomicrograph of liver section of adult albino rat of received 1/30 LD₅₀ carbaryl showing:
a) Intracytoplasmic vacuolation of different sizes
b) Widening blood sinusoids denoting degenerating hepatic cells. (*Haematoxylin & Eosin X 400*)



(c) A photomicrograph of liver section of adult albino rat of received 1/20 LD₅₀ carbaryl showing: Most of hepatic cells were enlarged and the cytoplasm was granular and vacuolar Hepatic cell nuclei appeared fragmented chromatin Hepatic blood sinusoids were dilated (*Haematoxylin & Eosin X 400*)

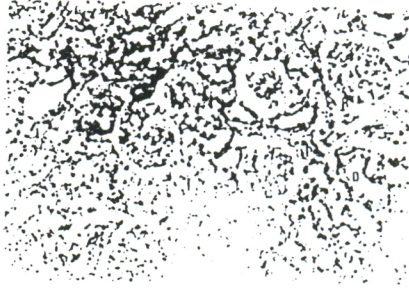


(d) A photomicrograph of liver section of adult albino rat of received 1/10 LD₅₀ carbaryl showing Loss of cell architecture. Increased degeneration of hepatic cells. Some of cells has no nuclei (Necrotic cells). Some of hepatic nuclei has fragmented chromatin and widening of blood sinusoids (*Haematoxylin & Eosin X 1000*)

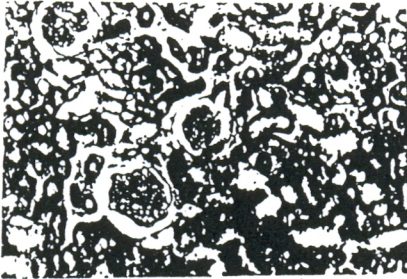


(e) A photomicrograph of liver section of adult albino rat of the recovery group showing Some of cells return to normal cell structure (regenerating cell) The hepatocytes were enlarged and showed granular cytoplasm and other areas showed irregular cell arrangement. (*Haematoxylin & Eosin X 1000*)

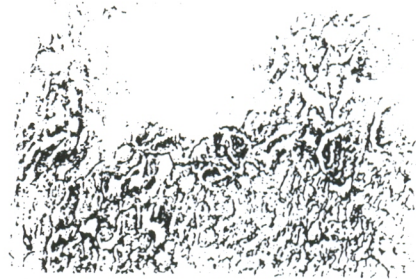
Figure (1) Photomicrographs showing histopathological effects of carbaryl on the liver of rats



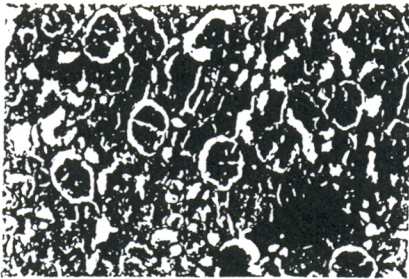
(a) A photomicrograph of normal kidney section of adult albino rat of the control group showing Renal capsule (R), Proximal convoluted tubules (P) and distal convoluted tubules (P) (*Haematoxylin & Eosin X 600*)



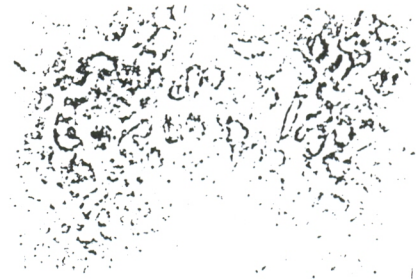
(b) A photomicrograph of kidney section of adult albino rat of received 1/30 LD₅₀ carbaryl showing: Glomerular capillaries (G) appear small in size with partial endothelial vacuolation (→). Convoluted tubules were widely separated and show foci necrosis (*Haematoxylin & Eosin X 600*).



(c) A photomicrograph of kidney section of adult albino rat of received 1/20 LD₅₀ carbaryl showing: The tubules show cloudy swelling and degeneration of separated spaces. The proximal distal, collecting tubules are dilated (*Haematoxylin & Eosin X 600*).

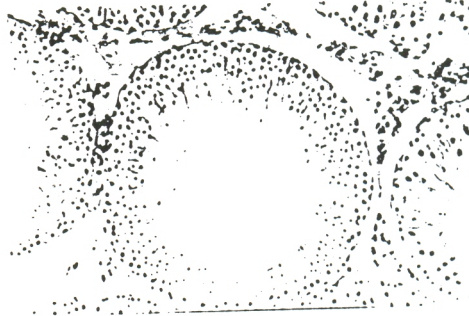


(d) A photomicrograph of kidney section of adult albino rat of received 1/10 LD₅₀ carbaryl showing: Increase in cloudy swelling and degeneration of tubular cells. Distortion of glomeruli with widely separated spaces (*Haematoxylin & Eosin X 1000*).

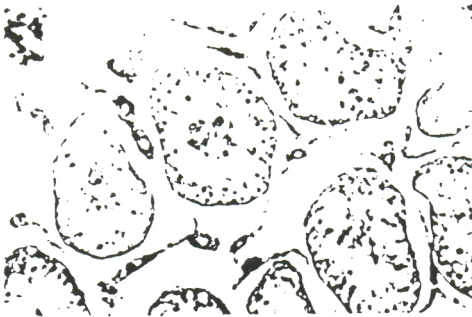


(e) A photomicrograph of kidney section of adult albino rat of the recovery group showing: The tubules show normal epithelial lining and normal lumen. Complete recovery of some glomeruli (*Haematoxylin & Eosin X 600*).

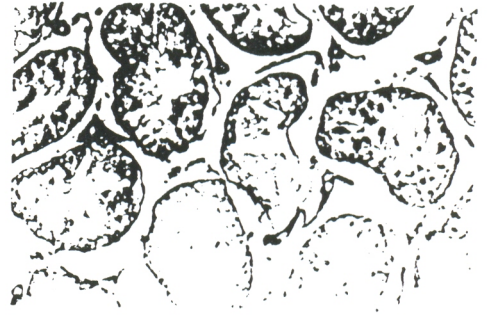
Figure (2) Photomicrographs showing histopathological effects of carbaryl on the kidney of rats



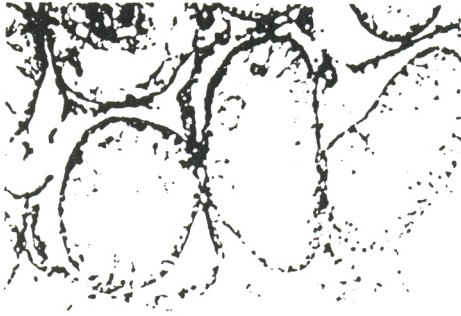
(a) A photomicrograph of normal testis section of adult albino rat the control group showing: Normal histopathological pattern of seminiferous tubules and interstitial cells in between tubules. The lumen of tubules filled with spermatozoa. (*Haematoxylin & Eosin X 250*)



(b) A photomicrograph of testis section of adult albino rat of received 1/30 LD₅₀ carbaryl showing: Mild to moderate degree of degenerative changes in germinal layer of seminiferous tubules in the form of presence of vacuolation and irregular arrangement in the germinal layer. (*Haematoxylin & Eosin X 400*).



(c) A photomicrograph of testis section of adult albino of received 1/20 LD₅₀ carbaryl showing: Irregular in basement membrane. Moderate degree of degenerative changes in spermatogenic cells with beginning of appearance of spermatid giant cells. (*Haematoxylin & Eosin X 400*)



(d) A photomicrograph of testis section of adult albino rat of received 1/10 LD₅₀ carbaryl showing: Severe degree of degenerative changes in the germinal layers of the seminiferous tubules with the presence of giant cell spermatid. Engorged blood vessels between tubules. (*Haematoxylin & Eosin X 400*).



(e) A photomicrograph of testis section of adult albino rat of the recovery group showing: There was an increase in the cell layers within the tubules with the of huge number of immature sperms. Interstitial cells appear normal. (*Haematoxylin & Eosin X 1000*).

Figure (3) Photomicrographs showing histopathological effects of carbaryl on the testes of rats.

Table (7): Effect of carbaryl administration on reproductive parameters of rats (mean±S.D.).

	Control	Treated rats			Recovery
		1/30	1/20	1/10	
Conc. Of sperm cells in (x10 ⁶ sperm/epididymis)	62 ±3.4	34.83 ±4.09	31** ±3.37	29.17* ±2.56	39.5 ±2.656
Conc. of sperm motility in %	62 ±7.9	34.17 ±2.74	27.17* ±2.42	16.5 ±2.5	50.67 ±5.94
Conc. of live sperm in %	86.33 ±3.81	59.83 ±3.93	52.33 ±2.25	39.83* ±4.19	63.33 ±10.95
Conc. of sperm abnormality in %	12.3 ±1.2	15.3 ±1.6	16.9* ±1.5	20.3 ±1.1	16.5 ±2.1

* P = 0.05

** P = 0.01

*** P = 0.001

Kidney showed the partial endothelial vacuolation of the capillary tuft. The changes in renal corpuscles increased with marked distortion, the inter-tubular spaces were marked increased. There were increment in cloudy swellings and degenerations Fig. (2). These resulted are in agreement with (Baronia *et al.*, 1991). They found that after 5 weeks of carbaryl administration, collecting proximal distal tubules showed dilatation and rupture of the glomeruli. Testis showed the degenerative changes in the germinal layers of seminiferous tubules with engorgement of blood vessels, increased number of spermatid giant cell seminiferous tubules (Fig., 3).

REFERENCES

- Abad, A.; Moreno, M.J.; Pelegri, R. and Martinez, M.I., (2001): Monoclonal enzyme immunoassay for analysis of carbaryl in fruits and vegetables without sample clean up. *J. Agric. Food Chem.* 49: 1707-1712.
- Ashry, K.M. (1990): Some toxicological studies on some carbamates commonly used in Egypt. M.Sc Thesis, Fac. Vet. Med., Alex. University.
- Baronia, A.K., Sharma, J.D., and Sahai, Y.N. (1991) : Toxic effects of carbaryl in the liver and kidney of *rattus albino*. *J. Nat. Con.* 3: (2) 127-132.
- Bearden, J.H. and Fuguay, J.W. (1980): "Applied Animal Reproduction" Reston Publishing co., Inc. Reston, Virginia, p. 158-165.
- Bessey, O.A.; Lowry, O.H. and Brock, M.J. (1964): A method for rapid determination of alkaline phosphatase with five cubic millimeters of serum. *J. Biol. Chem.*, 164: 321-329.
- Billing, B.H.; Haslam, R. and Wald, N. (1971): A colorimetric technique for determination of serum bilirubin. *Ann. Clin. Biochem*; 8:21

- Burtis, C.A. and Ashwood E.R. (1994): "Tietz textbook of clinical chemistry" 2nd ed. W.B. Saunders Co. Philadelphia, London, Toronto, Mexico City, Tokyo.
- Christopher, S.; Frings, P.D. and Ralph. M.I. (1970): Colorimetric method for determination of total serum lipid based on Sulphosphovalanine reaction. *Am. J. Clin. Path.*, 53: 89.
- Dacie, S.J.V. and Lewis, S.M. (1986): "Basic Haematological techniques" In *Practical Haematology International Student 6th Ed.* Churchill Livingstone. Edinburgh, London. Melbourne and New York. p 22-50.
- Dhanpakian, P. and Juliet, P. (1994): Histopathological changes in the kidney of cyprinus carpio exposed to malathion and sevin. *J. Environ. Biol.*, 15(4): 283-291.
- Doumas, B.T.; Watson, W.A. and Biggs. H.G. (1971): "Determination of serum albumin." *Clinica. Chem. Acta.* 31: 87.
- Drury, R.A.B. and Wallington, E.A. (1980): "Carleton's Histology Technique" 5th ed. Oxford Univer. Press. New York Toronto. 131-135.
- Ellman, G.L.; Courtney, K.D.; Andres, V.Jr. and Featherstone, R.M. (1961): A new and rapid colorimetric determination of acetyl cholinesterase activity. *Biochem. Pharmacol.* 7: 88-95.
- Flegg, H.M. (1973): Enzymatic determination of cholesterol. *Ann. Clin. Biochem.* 10: 79-84.
- Gomes, J.; Dawodu, A.U. and Anilal, S.V. (1999): Hepatic injury and disturbed amino acid metabolism in mice following prolonged exposure to organophosphorus pesticide. *Human and Exp. Toxic.* 18: 33-37.
- Gupta, R.C.; Ingls, N.E.; Paul B.S. and Kwatra, M.S. (1981): Role of residual estimation, and clinico-biochemical and pathological changes in the diagnosis of toxicity *Bubalus bubalis* caused by malathion. *Ind. J. Anim. Sci.*, 51(6): 616- 622.
- Harris, H.L. (1989): *Carletons histological technique.* 5th ed. Oxford Univ. Press New York, Toronto, pp. 33-48. Cited by Drury, R.A. and Wallington, A. (1980).
- Kiran, R.; Sharma, M. and Bansal, R. (1985): In vivo effect of carbaryl on some enzymes of rat liver, kidney and brain. *Pesticides* 19: 42-43.
- Krug, H.F. and Berndt, J. (1985): Inhibition by pesticides of prostaglandin formation in blood platelets. *Blut* 51: 19-23.
- Lucas, D.H. and Blank, R.E. (1977): Spectrophotometric standards in the clinical laboratory. *Animal lab.* 9: 77-83.
- Makhlouf, M.M. (1993): Teratogenic and embryotoxic effect of carbamates on rats. Thesis (M.V. Sc.), Fac. Vet. Med. Cairo University.
- Mount, M.E.; Dayton, A.D. and Oehme, F.W. (1981): Carbaryl residues in tissues and cholinesterase activities in brain and blood of rats receiving carbaryl. *Toxicol. Appl. Pharmacol.* 28: 282-296.
- Parafita, M.A. and Otera, F.M.P. (1984): The interaction of carbaryl with the metabolism of isolated hepatocytes. Effect on gluconeogenesis. *Gen. Pharmacol.* 15: 333-337.
- Patton, C.J. and Crauch, S.R. (1977): Enzymatic colorimetric method of urea determination. *Annul. Chem.*, 49: 464-469.

- Rahmy, N.A. (1994): Clinicopathological studies on the effect of carbamate insecticides in laboratory animals. Ph.D. Thesis, Fac. Vet. Med. Cairo University.
- Ray, S.K. and Podder, M.K. (1983): Carbaryl- induced elevation of corticosterone level and cholinergic mechanism. *Biosci. Rep.* 3: 973-977.
- Ray, S.K. and Podder, M.K. (1984): Effect of pentylenetetrazole on the carbaryl-induced changes of serotonin metabolism in rat brain hypothalamus. *Biosci. Rep.* 4: 427-432.
- Reitman, S. and Frankel, S. (1957): A colorimetric method for the determination of serum glutamic oxaloacetic acid, and glutamic pyruvic transaminase. *Am. J. Clin. Path.*
- Rizk-Alla, M.M. (1995): Biochemical studies on the toxic effect of some pesticides on experimental rats. M.Sc. Thesis, Fag. Agric., Ain Shams Univ.
- Saad, H. (1992): The role played by sex in modification of toxic action of some pesticides. Ph. D Thesis, Fac. Vet. Med., Cairo Univ.
- Siest, G.; Henny, J.; Schiele, F. and Young, D.S. (1985): Kinetic determination of creatinine. *Interpretation of Clinical Laboratory Tests.* p. 220-234.
- Snedecor, W.G. and Cochran, W.G. (1975): *Statistical methods.* 6th Ed., Iowa State Univ. Press, Ames, Iowa, USA.
- Sobbhy, H.M., (1987): Pharmacological and toxicological studies on the carbamate insecticide methomyl. M.Sc. Thesis, Fac. Vet. Med., Cairo University.
- Sonnenwirth, A.C. and Jarett, L. (1980): "Grawohl's Clinical Laboratory Methods and Diagnosis". 8th Ed. The C. V. Mosby Co. St. Louis. 135-138.
- Swilum, M.A. (1996a): Effect of long term administration of sublethal doses of carbaryl on the kidney of adult albino rat. *Al-Azhar Med. J.* 25(2): 123-130
- Swilum, M.A. (1996b): Effect of long term administration of sublethal doses of carbaryl on the testis of adult albino rat. *Al-Azhar Med. J.* 25(2): 131-140.
- Trinder, P. (1969): Determination of blood glucose using glucose oxidase with an alternative oxygen acceptor. *Ann. Clin. Biochem.* 6: 24-27.
- Wakakura, M.; Ishikawa, S. and Uga, S. (1978): Ultrastructural hepatic changes by carbamate pesticide (Sevin) in rats. *Environ. Res.* 16: 191-204.

تأثير تناول مبيد الكارباريل على الخصوبة ووظائف الكبد والكلى فى فئران التجارب

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استهدفت هذه الدراسة دراسة تأثير المبيد الحشرى الكارباريل على خصوبة ودرجة نشاط أنزيمات الكبد والكلى. حيث تم دراسة التأثيرات السمية لمبيد الكارباريل على الفئران وتم تحديد الجرعة المميتة لـ ٥٠% من عدد الفئران وأمكن معرفة الأعراض السمية الناتجة عن هذا المبيد كما تم دراسة تأثير المبيد على بعض مكونات مصل الدم وعلى نشاط بعض الأنزيمات والتغيرات التى تطرأ على الكبد والكليتين والأعضاء التناسلية وتم فحص خلايا الدم الحمراء عديدة الأصباغ وخلايا الدم الحمراء الكاملة النمو.

وأظهرت النتائج التى تم تحليلها إحصائيا ومقارنتها بالكنترول (العينات غير المعاملة) ما يلى:

- أن إعطاء مبيد الكارباريل عن طريق الفم لمدة ٩٠ يوم للفئران فى جرعات (٢,٤٥ ، ٣,٦٨ ، ٧,٣٥ مجم/١٠٠ جرام من وزن الحيوان على التوالى أدت إلى زيادة معنوية فى إنزيمات الكبد والبيليروبين واليوريا والكرياتينين.
- وجد زيادة معنوية فى مستوى حمض اليوريك ومستوى الدهون والكوليسترول والسكر فى الدم.
- وجد نقص معنوى فى مستوى الصوديوم والبوتاسيوم والبروتين ووجد نقص كبير فى نشاط أنزيم الكولين استيريز بالدم.
- حدث نقص فى عدد كرات الدم البيضاء والحمراء وتركيز الهيموجلوبين والصفائح الدموية.
- حدث نقص فى وزن الخصية وعدد الحيوانات المنوية ونقص ملحوظ فى نشاط الحيوانات المنوية ووجد زيادة فى نسبة الحيوانات المنوية الميتة والمشوهة مما يؤثر على الخصوبة فى ذكور الفئران.
- حدثت تغيرات هستوباثولوجية فى عدد من الأعضاء كان من أبرزها حدوث استمالة بخلايا الكبد والكلى والخصية بالإضافة إلى وجود بؤر نيكروزية فى الكبد.