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HISTOLOGICAL CHANGES INDUCED BY FEEDING ON FRIED POTATO AND PROTECTIVE ROLE OF CURCUMIN IN THE LIVER OF FEMALE RATS AND THEIR OFFSPRINGS

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ABSTRACT

This study deals with the evaluation of the histological effect of feeding on fried potato and the protective role of curcumin on the liver of pregnant rats and their offspring. The results showed that the using of fried potato caused different histopathological lesions in the liver of female rats and their offspring.

These lesions include vacuolated cytoplasm, dilated blood vessels, congested blood vessels, haemorrhage, pyknosis, necrosis and complete degenerated areas. The emerged data showed that fried potato induced histological alterations in the liver of rats similar to those caused due to acrylamide in food.

The present findings suggested that curcumin have no protective effect against the degenerative effect of fried potato..

Key words: Fried Potato – Curcumin – Liver – Rats - Offspring.

INTRODUCTION

In April of (2002) the discovery of the presence of a "probable carcinogen" in a range of fried and oven- cooked foods by Swedish Scientists caused worldwide concern carbohydrate rich foods were posed as a cancer threat in the headlines, and food manufactures consumers and health professionals quickly sought additional information guidance and reassurance (Ahn *et al.*, 2003).

Tareke *et al.* (2000) exhibited a strong increase of the level of acrylamide to induce Hb adduct in rats fed on fried diet, compared to control rat fed un-fried diet. Also Tareke *et al.* (2002) observed a reaction products (adducts) of acrylamide with N termini of hemoglobin. They added that moderate levels of acrylamide (5- 50 microgram/kg) were measured in heated protein- rich foods and higher contents (150-4000 microg/kg) in carbohydrate- rich foods, such as potato products and crisp bread. Acrylamide could not be detected in unhealed control or boiled foods (<5 microg/kg).

Mottram *et al.* (2002) and Stadler (2002).reported the presence of acrylamide in a range of fried and oven –cooked foods, these showed how acrylamide can be generated from food components during heat treatment as a result of Maillard reaction between amino acid and reducing sugars. They found that asparagine, a major amino acid in potatoes and cereals, is a crucial participant in the production of acrylamide by these pathway carcinogenic properties. Deng *et al.* (1993), Friedman *et al.* (1995), Lopachin *et al.* (2002) reported that acrylamide has been toxic effects on the nervous systems and neurotoxicity in humans.

On the other hand Mucci *et al.* (2003) reported that there was little evidence of an association between and specific backed or fried potato product and cancer risk in human. Histopathological alterations in liver, kidney, brain and erythrocyte due to acrylamide exposure were documented (Dixit *et al.*, 1982 and 1984; Segerback *et al.*, 1995 and Gamboa *et al.*, 2003).

In addition, the experimental groups fed on diet containing fried potato chips or treated with acrylamide from day 6 to day 20 of gestation produced similar histopathological alteration in liver, kidney and heart of delivered newly born highest intensity of lesions in those maternally fed on fried potato chips (El-Ghawet, 2003). Marlowe *et al.* (1986), orally administered male and pregnant female mice on day 13.5 and 17.5 day of gestation with 120 mg/kg (2, 3- 14 C) acrylamide and assayed the radioactivity of acrylamide labelled with radioactive carbon at 0.33, 1, 3, 9, 24, 72, and 216 hr. Absorption from the stomach was virtually complete by 3 hr; renal and hepatic elimination was essentially complete at 24hr. This study indicates that acrylamide is efficiently absorbed from the stomach and eliminated by the liver, kidney, and pancreas. Miller and Mc Queen (1986) exposed isolated hepatocytes to acrylamide and [3H] thymidine for 18 hr. They found that acrylamide concentrations exceeding 10 (-2) µ were cytotoxic to hepatocytes. Dearfield *et al.* (1988) reported that after absorption of acrylamide, it is rapidly metabolized

primarily by glutathione conjugation. Acrylamide can bind to DNA, which has implications for its genotoxic and carcinogenic potential. Field *et al.* (1990) found that oral administration of acrylamide on gestational day 6-17 to mice (0, 3, 15 or 45 mg/kg) and on gestational day 6-20 to rats (0, 2.5, 7.5 or 15 mg/kg) were found to induce reduced body weight gain in both species. Titenko *et al.* (1998) reported that there was significant increase in the proportion of morphologically abnormal embryos, following paternal interperitoneal exposure of 50 mg acrylamide /kg.

It is noteworthy that the use of medicinal plants or their crude extracts in the prevention and/ or treatment of several chronic diseases has been traditionally practiced in various different societies worldwide (Kawamori *et al.*, 1999). Curcumin is the major yellow colouring matter isolated from turmeric, the powdered rhizome of the medicinal plant curcuma Longa Linn. It has widely been used to colour and flavor foods, as a cosmetic and in some medicinal preparations (Limtrakul *et al.*, 2001).

Curcumin (diferulylmethane), a phenolic compound, has been shown to possess antioxidant, free radical scavenging, anti-inflammatory, anti-thrombotic, hypo-glycemic and hypocholestrolemic (Ruby *et al.*, 1995, Chuang *et al.*, 2000). Moreover, curcumin is not toxic human up to 800 mg/ kg/ day, Cheng *et al.*, 2001). So the present study aims to investigate the histological changes in the liver of rats (dams and their offspring's) fed on fried potato and the protective role of curcumin.

MATERIAL AND METHODS

Forty eight adult virgin female rats weighing (180-200 g) were obtained from the Experimental Breeding Farm at Helwan, Egypt. After 2 weeks period of acclimatization, female rats were randomly distributed into (8) groups each of six animals per cage on a 12-hr. light/12-hr. dark cycle.

Diet:

The composition of the standard diet, vitamin mixture and salt mixture (Mullar, 1964) used in this experament is: *Casein 12, Sucrose 5 , Corn oil 10, Minerals 4, Cellulose 4, Vitamins 1, Starch 64* .

Fried-potato chips supplementation:

Fried potato chips were prepared at laboratory , mixed with standard diet at concentration of 15% (low dose) and 30% (High dose) supplied for feeding female rats for 6 weeks before gestation and 6 weeks after gestation.

Curcumin supplementation:

Powder curcumin was obtained from the market and mixed with standard diet at concentration of 2% and supplied for feeding during experimental period (El-Gammal, 2003).

Acrylamide supplementation:

Acrylamide supplied from Aldrich chemical company was used in the present work Female rats were arranged into 8 groups each was composed of 6 female rats as follows:

1- Control female rats fed standard diet.

2- Control female rat fed standard diet containing 2% curcumin.

3- Acrylamide-treated female rats (each individuals reiceved daily oral doses of 25 µg/ kg for 6 weeks before gestation and 6 weeks after gestation) according to protocole used by El-Ghawet (2003).

4- Acrylamide-treated female rats + 2 % curcumin.

5- Fried potato chips supplementation: female rats were feeding standard diet mixing with fried potato chips at concentration of 85%, 15% for respectivily (for 6 weeks before gestation and 6 weeks after gestation).

6- Combined standard diet, Fried potato chips (85%, 15%) + 2% curcumin.

7- Fried potato chips supplementation: female rats were feeding standard diet mixing with fried potato chips at concentration of 70%, 30% for respectivily (for 6 weeks before gestation and 6 weeks after gestation)..

8- Combined standard diet, Fried potato chips (70%, 30%) + 2% curcumin.

After 6 weeks the control and all the experimental groups were placed in cages overnight with untreated males (in a ratio of 1 male to 3 females). Then, in every morning vaginal smears were prepared and examined under the microscope. The presence of sperms in the vaginal smear during estrus denoted the day zero of gestation.

The pregnant females of the control and experimental groups were scarificed after 12 weeks and weanling rats aging 3 weeks.

Livers of female and their weanling (three weeks old) were removed, fixed in 10% formal saline, dehydrated, cleared and embedded in paraffin wax. Sections were cut at 5-7µ thickness and stained with haematoxylin and eosin.

Histomorphemetric analysis:

The numerical abnormal cells were counted by image-pro version 4.5

The nucleocytoplasmic indices were calculated according to Lang *et al.* (1986):

NP=VN/ (VC-VN)

Where NP is nucleocytoplasmic index, VN is the volume of nucleus and VC is the volume of cell.

Volume =V= 4/3 πr^3; where r is the radius of the nucleus or cell.

RESULTS

Control liver: The normal structure of the liver of control rat is illustrated (Figs. 1 & 2). The liver is surrounded by a thin connective tissue capsule it is made up of two main constituents, the parenchyma and stroma. The parenchyma consists of the liver cells or hepatocytes, whereas the stroma is composed of connective tissue material extending from the tissue septa, which divide the tissue into ill-defined liver lobules. Each hepatic lobule contains a central vein from which radiate and branching cords or plates of hepatic cells, each of one or two cells thick. The spaces lying between these plates constitute the hepatic sinusoids surrounded by a discontinuous layer of flattened endothelial cells with flattened darkly stained nuclei and cytoplasm. The Kupffer cells are distinctly large and possess prominent nuclei, and a number, of them appear frequently bulging into the sinusoidal lumens. The hepatic cells or hepatocytes are polyhedral in shape, large in size and exhibit distinct cell boundaries. The hepatocytes enclosing a homogeneously fine granulated, acidophilic cytoplasm and embodying a centrally placed large spherical nucleus, a prominent nucleoli and distinct chromatin particles (Fig. 2).

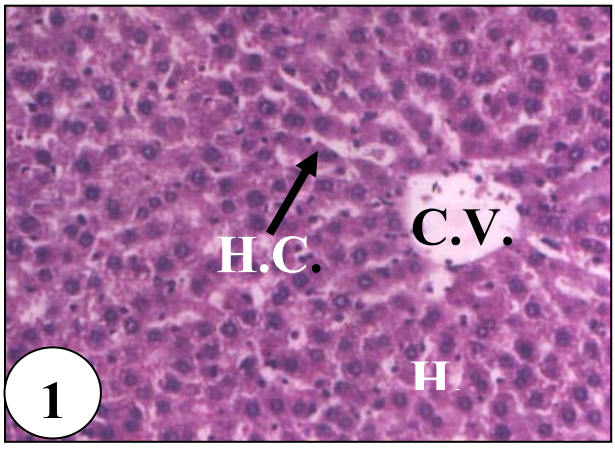


Figure (1): Liver section of control female rat showing central vein (C.V.), hepatic cords (H.C.) and hepatocytes (H.). (H/E X200).

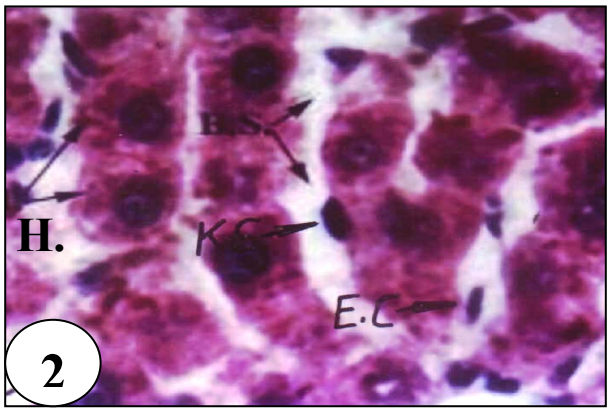


Figure (2): Liver section of control female rat showing hepatocytes (H.), blood sinusoids (B.S.), Kupffer cells (K.C.) and endothelial cells (E.C.). (H/E X1000).

Mothers fed on diet containing acrylamide:

The liver of female rats fed on diet containing acrylamide exhibited different histological changes. Where, pyknosis, cell degeneration, lymphocytic infiltration, vacuolation, congestion and haemorrage were perceptive in most parts of the hepatic tissue (Fig. 3).

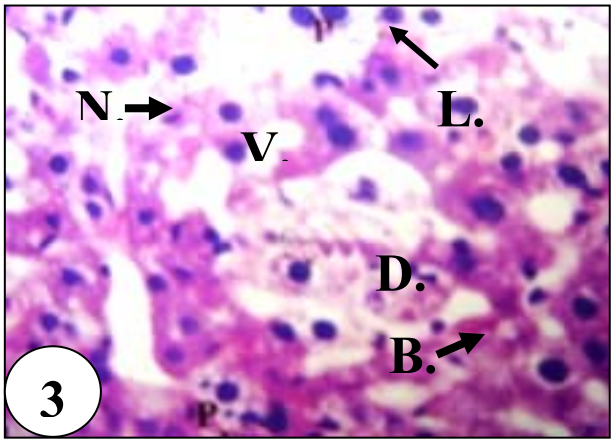


Figure (3): Liver section of female rat fed on diet containing acrylamide showing necrosis (N.), vacuolation (V.), proliferation of lymphocytes (L.), degeneration (D.) and haemorage (H.). (H/E X 400).

Mothers fed on diet containing acrylamide and curcumin:

The using of curcumin in the diet of female rats as a protective agent against the side effects of acrylamide exhibited ill defined protection in the liver. Inspected liver sections of rats of this group showed some of the degenerative changes recorded in the liver of rats fed on diet containing acrylamide. The cytoplasm of the hepatocytes showed less vacuoles and the nuclei were almost normal. Also probferation of the endothelial cells, hypertrophied Kupffer cells and dilated blood sinusoids were greatly encountered (Fig. 4).

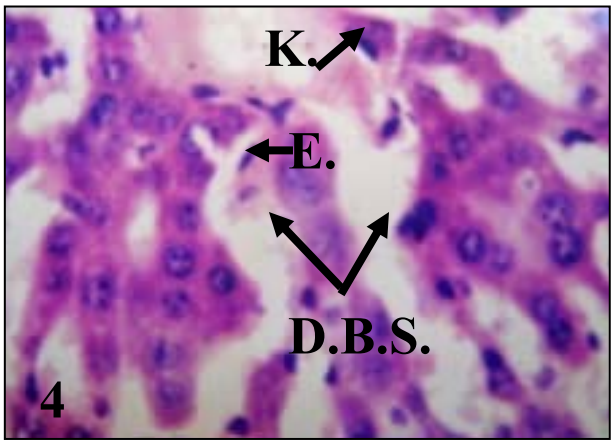


Figure (4): Liver section of female rat fed on diet containing acrylamide and curcumin showing hyperthrophied Kupffer cells (K.), increase of endothelial cells (E.) and dilated blood sinusoids (D.B.S.). (H/E X 400).

Mothers fed on diet containing 15% fried potato:

The obtained results indicated that feeding on diet containing 15% of fried potato for three months induced different histological changes in the liver of rats. These changes were mainly represented in prevailing vacuolated hepatocytes cytoplasm denoting fatty degeneration (Fig. 5). Also, congestion in the blood vessels and haemorrhage were delineated. Moreover, condensed nuclear materials or pyknotic nuclei were greatly encountered in most of the hepatocytes.

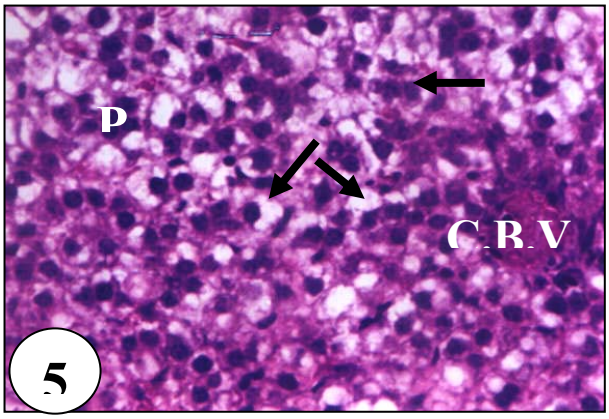


Figure (5): Liver section of female rat fed on diet containing 15% fried potato showing congested blood vessel (C.B.V.), vacuolation (V) and pyknotic nuclei (P.). (H/E X 200).

Mothers fed on mixed diet containing 15% fried potato and curcumin:

The results of the present work showed that the using of curcumin in the diet ameliorated the changes of the hepatic architecture of rats fed on fried potato, most of the hepatocytes and their nuclei were mostly similar to those of the control animals. But the resulted amelioration was mild, where some histological changes were ratified in the hepatic tissue. These histological changes were represented in dilatation of blood vessels and vacuolation of the hepatocytes cytoplasm in different area of the hepatic tissue (Fig. 6).

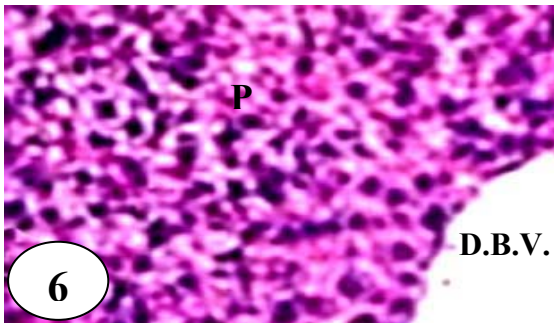


Figure (6): Liver section of female rat fed on diet containing 15% fried potato and curcumin showing dilated blood vessel (D.B.V.) and pyknotic nuclei (P.). (H/E X 400).

Mothers fed on diet containing 30% fried potato:

Investigated liver sections obtained from female rats fed on diet containing 30% fried potato for three months revealed severe histological lesions after three months of feeding on this diet. These lesions were represented by lymhpocytic infiltration, cytoplasmic vacuolation, pyknosis, focal areas of necrotic cells and congestion in blood vessels (Fig.7).

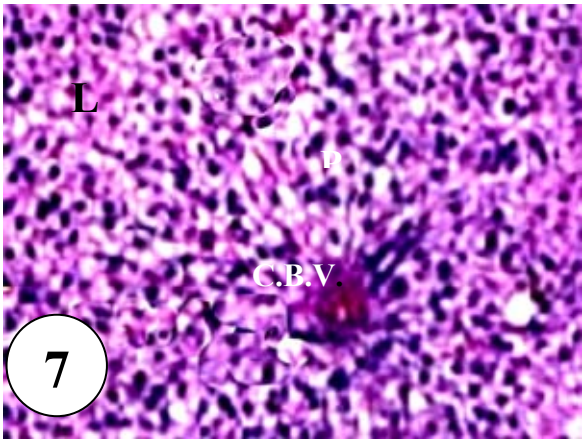


Figure (7): Liver section of female rat fed on diet containing 30% fried potato showing lymphocytic aggregation (L.) and necrotic area (N.A.). (H/E X 200).

Mothers fed on diet containing 30% fried potato and curcumin:

No amelioration was detected in the liver of female rats fed on diet containing fried potato and curcumin. The liver sections displayed mild dilatation in both blood vessels and sinusoids, congestion in blood vessels, vacuolation, pyknotic nuclei and some necrotic cells (Fig. 8).

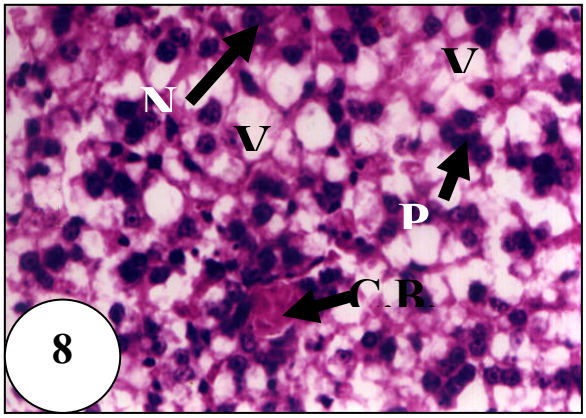


Figure (8): Liver section of female rat fed on diet containing 30% fried potato and curcumin showing congested blood vessels (C.B.V.), vacuolation (V.), pyknotic nuclei (P.) and necrotic cells (N.). (H/E X 400).

Control weanling

Liver sections of weanling are similar structure to those of mother rats.

Weanling of mother fed on diet containing acrylamide:

Weanling of mothers fed on diet containing acrylamide revealed severe histological changes in the liver. Investigated liver sections obtained from this group revealed prevailing vacuolated hepatocytes, congestion in blood vessels, necrosis in most parts of the hepatic tissue and cell degeneration (Fig. 9).

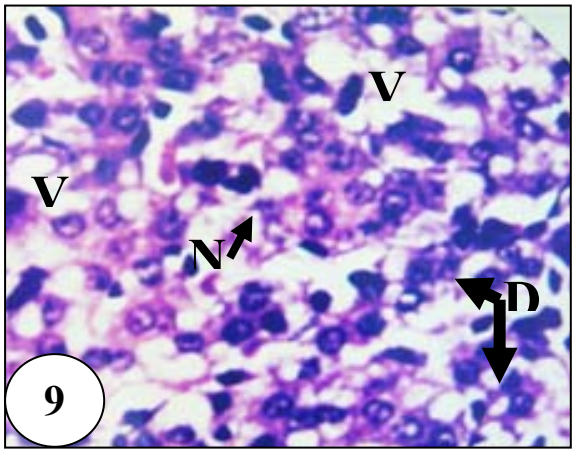


Figure (9): Liver section of weanling rat fed on diet containing acrylamide showing vacuolated hepatocytes (V.), necrosis (N.) and degenerated cells (D.). (H/E X 400).

Weanling of mother fed on diet containing acrylamide and curcumin:

Inspected liver sections of this group showed that no protective effect for curcumin could be detected against the effect of acrylamide. Figure 10 shows oedematous blood vessels, lymphocytic proliferation and pyknosis and necrosis in the hepatocytes.

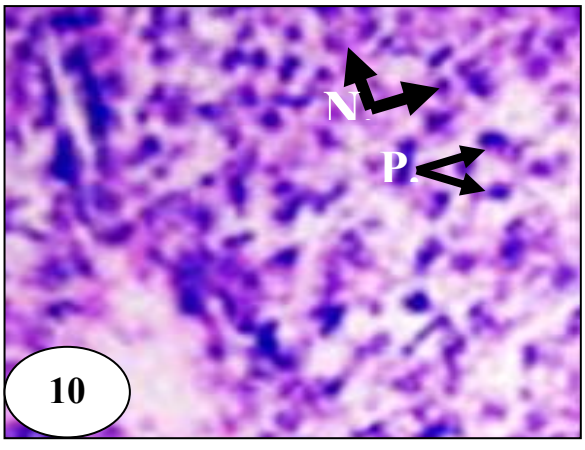


Figure (10): Liver section of weanling rat fed on diet containing acrylamide and curcumin showing pyknosis (P.) and necrosis (N.). (H/E X400).

Weanling of mother fed on diet containing 15% fried potato:

Liver sections obtained from this group displayed inflammatory response as indicated by the presence of inflammatory cells within the tissue, vacuolated hepatocytes cytoplasm, hypertrophied Kupffer cells and haemrrhage within the tissue (Fig.11).

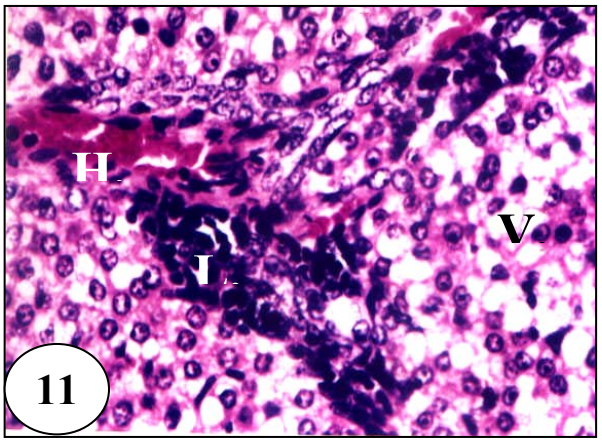


Figure (11): Liver section of weanling rat fed on diet containing 15% fried potato showing haemorrhage (H.), lymphocytic aggregation (L.) and vacuolation (V.). (H/E X400).

Weanling rats fed on mixed diet containing 15% fried potato and curcumin:

Inspected liver sections obtained from weanling offspring maternally fed with 15% fried potato in combination with curcumin after three months exhibited slight amelioration and showed peripheral regenerative feature. Where, congestion in blood vessels, vacuolation, lymphocytic infiltration and degenerative changes were still percepted in almost all parts of the tissue (Fig.12).

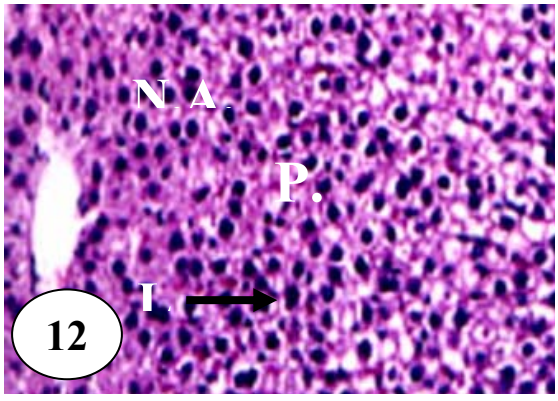


Figure (12): Liver section of weanling rat fed on diet containing 15% fried potato and curcumin showing congested blood vessel (C.B.V.) and necrotic area (N.A.). (H/E X200).

Weanling fed on diet containing 30% fried potato:

Investigated liver sections of weanling from maternal female rats fed on fried potato chips for three months showed dilated blood sinusoids, and blood vessels diffused Kupffer cells proliferation in between the hepatocytes. Also, vacuolation in the cytoplasm of the hepatocytes, pyknotic nuclei and nuclear fragmentation (karyolysis) were delineated (Fig. 13).

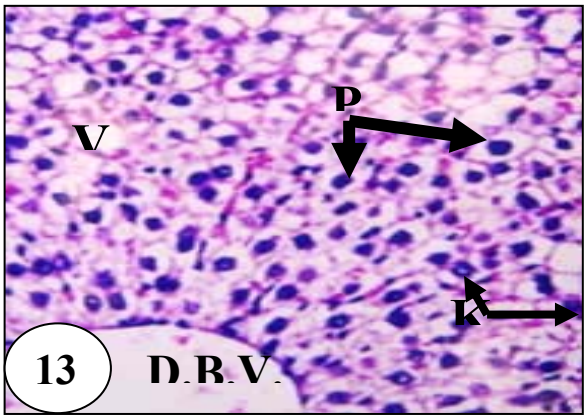


Figure (13): Liver section of weanling rat fed on diet containing 30% fried potato showing prevailing vacuolation (V.), karyolysis (K.), pyknosis (P.) and dilated blood vessel (D.B.V.). (H/E X400).

Weanling fed on diet containing 30% fried potato and curcumin:

The obtained data showed that the use of curcumin in the diet slightly minimize the side effects of fried potato. The most prominent amelioration was found in the restoration of the hepatocytes cytoplasm of weanling rats. But focal degenerated hepatocytes, karyolysis and proliferation of Kupffer and endothelial cells were encountered in the hepatic tissue (Fig. 14).

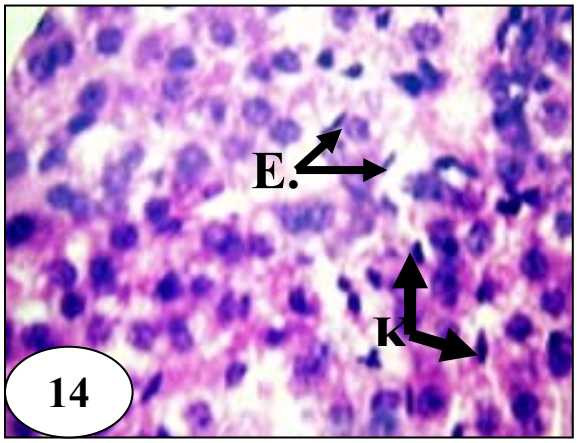


Figure (14): Liver section of weanling rat fed on diet containing 30% fried potato and curcumin showing increases of Kupffer (K.) and endothelial cells (E.). (H/E X 400).

Hestomorphemetric analysis:

The mean activity of hepatocytes can be measured by counting the normal, fragmented, necrotic and hypertrophied of hepatocytes in standard area of section in the control and treated liver of mother white rats and weanling (Tables 1&2). Table (1) shows the percentage of abnormality in control 25.47±7.39 and decrease to reach 19.89±8.238 in the curcumin group. Percentage of abnormality increased to 42.64±21.77, 39.63±17.45, 34.49±13.65, and 38.64±13.11 in groups treated with acrylamide, acrylamide + curcumin, low fried potato chips and high fried potato chips respectively. These percentages were recoveries when curcumin was used in last two treated groups to reach 28.84±9.33and 29.35±8.41 (Table 1 and Fig. 15).

Table (2) and Figure (16) show the percentage of abnormality in the control 21.60 ±7.39 and treated groups of weanling white rats. The percentages were increased to reached 41.88±15.47, 38.54±12.19, 27.1±13.53, and 36.41±14.11, in treated groups used acrylamide, acrylamide + curcumin, low fried potato chips and high fried potato chips, respectively. These percentages recovered in treated groups low fried potato chips + curcumin and high fried potato chips + curcumin to reach 26.53±9.31 and 24.42±11.65, respectively.

The volume of nucleus per volume of cytoplasm (nucleocytoplasmic index) in control mother white rats was 0.47±0.25 and increased to 0.51±0.22 and 0.63±0.26 in groups treated with curcumin and acrylamide. These indexes decreased to reach 0.2±0.07, 0.19±0.03, and 0.16±0.07 in groups treated with acrylamide, low fried potato chips and high fried potato chips respectively. There were some recoveries in groups treated with low and high fried potato chips + curcumin (Table 3 and Fig.17). Nucleocytoplasmic index in control and treated groups of weanling rats were 0.57±0.26, 0.14±0.09, 0.19±0.01, 0.22±0.02 and 0.22±0.01 in groups control, acrylamide, low and high fried potato chips and low and high fried potato chips +antioxidant respectively (Table 4 and Fig.18).

DISCUSSIOIN

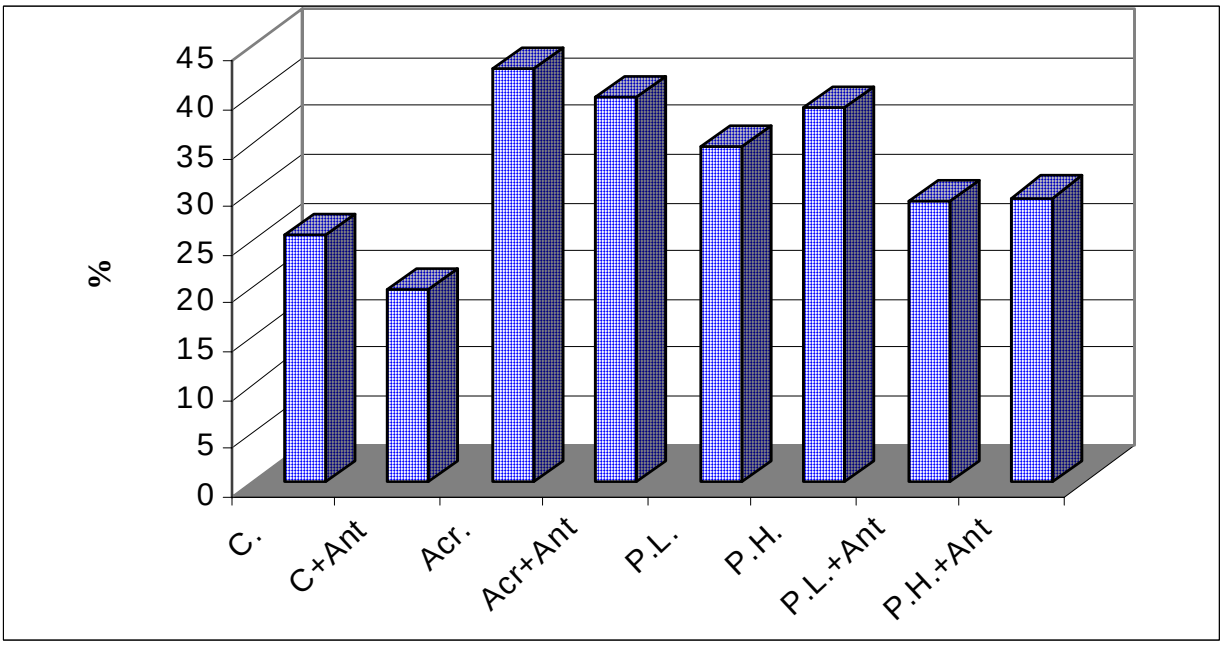
Acrylamide is a highly reactive and water-soluble polymer, which is commonly used in both industries and laboratories (Nordin *et al*., 2003). Recent studies reported the presence of acrylamide in heat treated food products and it has energized research efforts worldwide to define toxic mechanisms (Konings *et al*., 2003). The formation of acrylamide is particularly associated with high temperature cooking process for certain carbohydrate-rich foods, especially when asparagine (a naturally occurring amino acid found in carbohydrate foods) reacts with sugars (Mottram *et al*., 2002). Moreover, cooking at lower temperatures (e.g., by boiling) produces much lower level of acrylamide (Rydberg *et al*., 2003).

Table (1): Average quantitative analysis of hepatocytes in control and treated groups of mother rat.

Groups		Total No. of cells	Normal cells	Fragmented cells	Necrotic & hypertrophic	% of abnormality
C.	Average±S.D	59.38±12.26	42.99±9.22	8.31±3.53	4.52±2.76	21.61±7.39
C.±Anti	Average±S.D	62.38±15.17	49.12±10.12	9.21±4.16	5.13±3.12	22.98±7.48
	t test	0.162	0.371	0.094	0.731	0.761
W±Acry.	Average±S.D	95.23±14.85	53.34±11.55	31.77±8.93	8.11±3.49	41.87±15.47
	t test	0.002*	0.003*	0.000*	0.000*	0.000*
Acry.±Anti	Average±S.D	89.33±13.18	55.29±19.35	27.15±9.37	7.28±3.65	38.54±12.19
	t test	0.005*	0.000*	0.000*	0.000*	0.000*
W±P.L.	Average±S.D	82.35±22.65	50.16±17.43	17.93±11.4	4.39±2,55	27.11±13,53
	t test	0.000*	0.005*	0.000*	0.833	0.000*
W±P.H.	Average±S.D	104.59±41.55	66.48±28.41	31.9±14.72	6.18±2.61	36.41±14.11
	t test	0.000*	0.000*	0.000*	0.007*	0.000*
P.L.±Anti\	Average±S.D	98.5543.57	71.93±23.33	16.85±5.83	9.3±3.45	26.54±9.31
	t test		0.000*	0.000*	0.000*	0.052
P.H.±ant.	Average±S.D	111.36±40.72	74.7116.43	17.24±5.85	9.95±4.66	24.42±11.65
	t test	0.000*	0.000*	0.000*	0.000*	0.535

*=significant (P≤0.05)

Where: C= control, Anti= antioxidant (curcumin), Acr =Acrylamid, P= Potatos, L=low dose (15%)



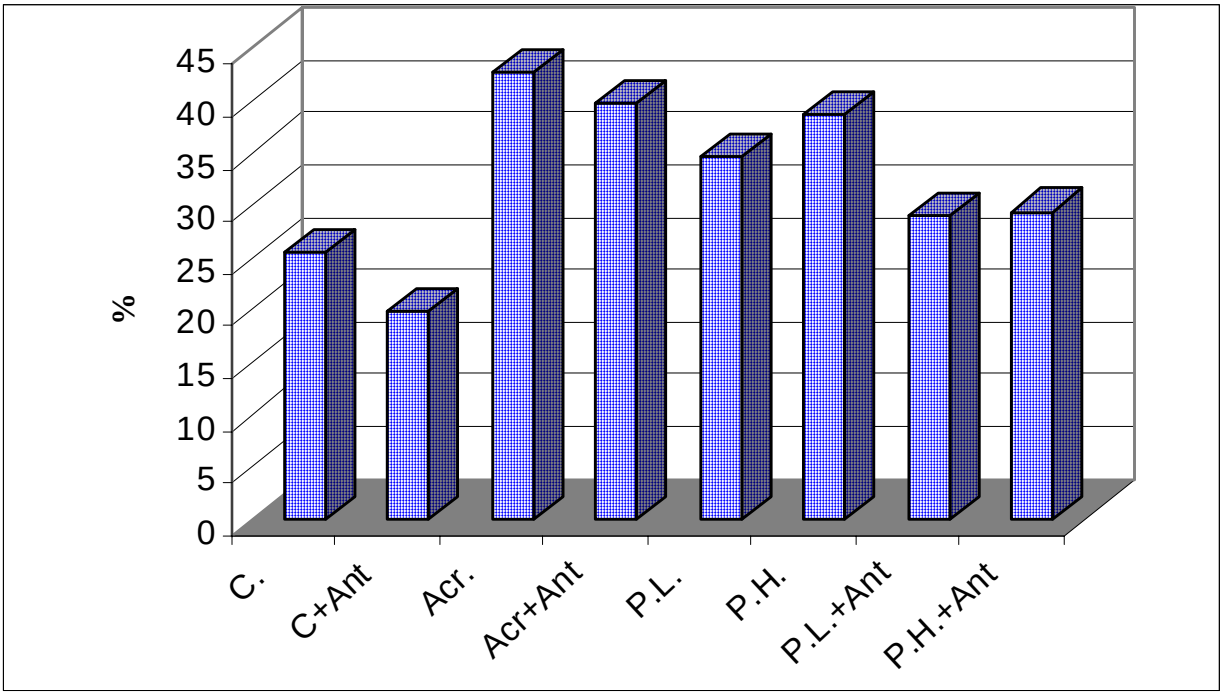
Histogram (16): Showing the percentage of abnormal hepatic cells in mother treated groups of white rats contrast to control.

Table (2): Average quantitative analysis of hepatocytes in control and treated groups of weanling rat

Groups		Total No. of cells	Normal cells	Fragmented cella	Necrotic & hypertrophic)	% of abnormality
C.	Average±S.D	56.38±9.54	42.99±11.87	9.14±3.79	5.22±2.51	25.47±7.33
C±Anti	Average±S.D	63.25±13.6	45.31±15.33	8.36±4.51	4.22±2.03	19.88±8.23
	t test	0.381	0.095	0.471	0.515	0.084
Acryl	Average±S.D	87.54±23.14	49.54±19.36	23.16±9.45	14.17±4.33	42.64±21.77
	t test	0.000*	0.000*	0.000*	0.000*	0.000*
Acry±Anti	Average±S.D	86.55±26.71	51.41±13.83	17.88±6.01	16.42±6.13	39.63±17.45
	t test	0.000*	0.001*	0.000*	0.000*	0.002*
P.L.	Average±S.D	73.25±11.37	49.85±15.72	14.32±7.91	10.94±4.21	34.49±13.65
	t test	0.000*	0.003*	0.000*	0.000*	0.001*
P.H.	Average±S.D	75.38±19.83	42.65±16.35	17.38±8.55	11.75±5.38	38.64±13.11
	t test	0.000*	0.000*	0.000*	0.000*	0.000*
P.L.±Anti	Average±S.D	83.43±26.44	49.11±13.07	14.63±6.27	9.43±4.73	28.84±9.33
	t test	0.000*	0.009*	0.000*	0.000*	0.152
P.H.±Anti	Average±S.D	85.17±31.05	63.23±17.35	15.11±4.66	9.89±4.32	29.35±8.41
	t test	0.001*	0.000*	0.000*	0.000*	0.047*

*=significant (P≤0.05)

Where: C= control, Anti= antioxidant (curcumin), Acry =Acrylamid, P= Potatos, L=low dose (15%), H= High (30%) and W=Weanling.



Histogram (16): Showing the percentage of abnormal hepatic cells in mother treated groups of white rats contrast to control.

Table (3): Nucleocytoplasmic index of hepatocytes in control and treated groups of white rat

Groups		Vol. of cell	Vol. of nucleus	Vol. of cytoplasm	Vol. of N/ Vol. of Cytoplasm
C.	Average	1709049.18	545886.09	1163163.09	0.47
	S.D.	586786.31	68030.70	942.03	0.25
C+Anti	Average	1646117.00	555574.02	1090542.98	0.51
	S.D.	29134.30	20130.19	80424.43	0.22
	%	-3.68	1.77	-6.24	8.55
	t-test	2.47	1.94	0.92	0.07
Acryl	Average	332173.14	128807.97	203365.17	0.63
	S.D.	25037.67	52178.56	12671.34	0.26
	%	-80.56	-76.40	-82.52	34.96
	t-test	0.00*	0.00*	0.00*	0.00*
Acry+Anti	Average	778892.43	128780.84	650111.60	0.20
	S.D.	30986.09	18731.99	1874.25	0.07
	%	-54.43	-76.41	-44.11	-57.79
	t-test	0.00*	0.00*	0.00*	0.00*
P.L.	Average	489002.19	79375.98	409626.21	0.19
	S.D.	42824.58	39045.92	91371.35	0.03
	%	-71.39	-85.46	-64.78	-58.71
	t-test	0.00*	0.00*	0.00*	0.00*
P.H.	Average	369096.27	51415.63	317680.64	0.16
	S.D.	120154.29	20271.47	57132.05	0.07
	%	-78.40	-90.58	-72.69	-65.51
	t-test	0.00*	0.00*	0.00*	0.00*
P.L.+Anti	Average	1817093.23	287062.50	1530030.73	0.19
	S.D.	1551461.39	228721.57	9823.47	0.05
	%	6.32	-47.41	31.54	-60.02
	t-test	0.73	0.00*	0.00*	0.00*
P.H.+Anti	Average	534811.20	92624.21	442186.99	0.21
	S.D.	148242.55	43118.28	20465.73	0.05
	%	-68.71	-83.03	-61.98	-55.37
	t-test	0.00*	0.00*	0.00*	0.00*

*=significant (P≤0.05)

Where: C= control, Anti= antioxidant (curcumin), Acry =Acrylamid, P= Potatos, L=low dose (15%) and H= High (30%).

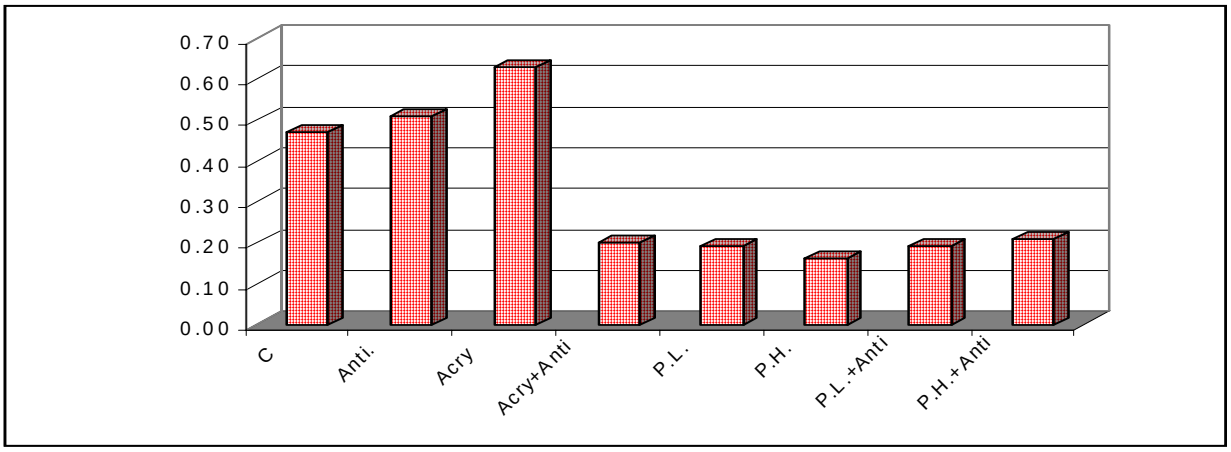


Fig.17: Nucleocytoplasmic index of hepatocytes in control and treated groups of mother white rats.

Table (4): Neucleocytoplasmic index of hepatocytes in control and treated groups of weallling white rat.

Groups		Vol. of cell	Vol. of nucleus	Vol. of cytoplasm	Vol. of N/ Vol. of Cytoplasm
C.	Average	1509049.18	545886.09	963163.09	0.57
	S.D.	586786.31	68030.70	942.04	0.26
C+Anti	Average	1618389.24	594208.98	1024180.26	0.58
	S.D.	139150.39	21071.20	74334.40	0.11
	%	7.25	8.85	6.34	2.37
	t-test	0.34	0.25	0.11	0.71
W+Acry.	Average	286080.59	34208.98	251871.61	0.14
	S.D.	117650.24	14471.78	804244.43	0.09
	%	-81.04	-93.73	-73.85	-76.04
	t-test	0.00*	0.00*	0.00*	0.00*
Acry.+Anti	Average	1781389.24	343208.98	1438180.26	0.24
	S.D.	317160.59	15461.93	31244.45	0.22
	%	18.05	-37.13	49.32	-57.89
	t-test	0.01*	0.00*	0.00*	0.00*
W+P.L.	Average	1817093.23	287062.50	1530030.73	0.19
	S.D.	1551461.39	228721.57	9453.07	0.01
	%	20.41	-47.41	58.85	-66.90
	t-test	0.89	0.00*	0.00*	0.00*
W+P.H.	Average	534811.20	62624.21	472186.99	0.13
	S.D.	148242.55	43118.28	62041.26	0.07
	%	-64.56	-88.53	-50.98	-76.60
	t-test	0.00*	0.00*	0.00*	0.00*
P.L.+Anti	Average	388311.09	70184.09	318127.00	0.22
	S.D.	38807.03	19105.92	30254.12	0.02
	%	-74.27	-87.14	-66.97	-61.07
	t-test	0.00*	0.00*	0.00*	0.00*
P.H.+ant.	Average	489792.06	89645.01	400147.05	0.22
	S.D.	222039.36	36321.25	32358.21	0.01
	%	-67.54	-83.58	-58.45	-60.47
	t-test	0.00*	0.00*	0.00*	0.00*

*=significant (P≤0.05)

Where: C= control, Anti= antioxidant (curcumin), Acry =Acrylamid, P= Potatos, L=low dose (15%), H= High (30%) and W= Weanling.

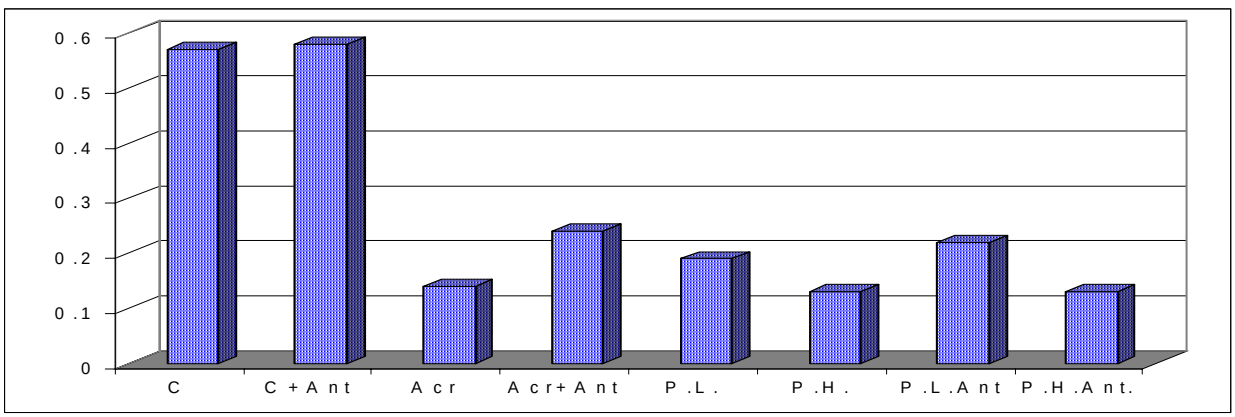


Fig.18: Nucleocytoplasmic index of hepatocytes in control and treated groups of weallling white rats.

In the present study, light microscopic examination of the liver of the female rats and their offsprings feeding to fried potato 3 months (6 weeks before gestation and 6 week after gestation), showed different sings of damage including cytoplasmic vacuolation, dilatation of sinusoidal and necrosis of the liver cells. Similar histopathological alterations in livers of offsprings maternally fed on fried diet. The histopatholgical effects in maternal tissues reflected the abnormal histological pattern in offsprings. These may be attributed to the transplacental passage of acrylamide and its metabolites to weanling tissues interfering with cell differentiation and exerted those drastic effects on their histological pattern (IKeda *et al.*, 1985).

In coincidence with the present findings, Sakr (2004) showed that acrylamide caused disturbance in the metabolic activity of fetal liver indicated by fat deposition in their liver cells. According to Lang *et al.* (1986) the nucleocytoplasmic index gives information about the cell status, cells in a good health when it has a high index. Abdel Aziz *et al.* (1994) reported that the carbamate insecticide caused carcinogenic effect in the body of exposed animals. Greatly reduced liver cells nucleocytoplasmic index was recorded which denoting disturbance in all hepatocytes activity and may resulted in necrobiotic changes in the liver.

Similar results were obtained by Miller and Mc Queen (1986) and Peterson (1987) in experimental animals hepatocytes cultivated *in vitro* as a result of acrylamide exposure. They observed marked necrosis of the liver cells and attributed such necrosis to acrylamide effect on the oxidase activity which showed marked decrease in the liver following acrylamide treatment.

From the present findings not only acrylamide which exerted intoxication but also swallowing of fried potato chips. In agreement, Diembeck *et al.* (1998) showed that acrylamide may also be readily absorbed from skin during handling of slices of fried diet while eating. Once acrylamide absorbed, it was conjugated by glutathion- S- transferase to N- acetyl- S- (3- amino-3 oxopropyl) cysteine or reacted with cytochrome P450 to produce glycidamide (Calleman *et al.*, 1990; Bergmark *et al.*, 1991 and Sumner *et al.*, 1999).

Furthermore glutathione- S- transferase is detoxifying enzyme in the liver and takes place many isoforms and plays a great role in maintaining cell function; conjugation of acrylamide or glycidamide with this enzyme may interfere with cell function and promote cell death (Bergmark *et al.*, 1991 and Barber *et al.*, 2001). They established that deleterious effects of acrylamide or its metabolite glycidamide seemed to be facilitate to form adduct with sulfhydryl groups on hemoglobin and other proteins. These may reduce the hemoglobin surface of carrying oxygen to tissue causing cell degeneration. The relationship between acrylamide and blood cells was reported by many investigators. Srivastave *et al.* (1983) found that acrylamide caused an increase in lipid peroxidation and a decrease in glutathione contents in rat liver. Rubin (1995) reported that the increase of lipid peroxidation might be the cause of the present hepatic lesions, because it leads to a loss of membrane integrity and in turn cause cell injury.

Segerbach *et al.* (1995) identified that injection of acrylamide form DNA adduct that may interfere with cell replication causing cell damage. Also, in coincidence with Gamboa *et al.* (2003), increased DNA adducts in liver may explain the systemic effect of the acrylamide and its metabolites and may illustrate the induced cell damage observed in the present study. Moreover, autoradiographic study carried out by (Marlowe *et al.*, 1986) showed that extensive distribution of acrylamide or its metabolite in the fetal tissues, showed increased rates of binding activity in RNA, DNA.

The present data indicated that fried potato may be caused a disturbance in the metabolic activity of females and weanling liver, as indicated by lesions in their liver cells. It seems that this is related to effect of fried potato, and it also might be due to decreased proteins available for triglyceride transport from the liver (Miller *et al.*, 1982). This is agreement with Nordin *et.al.* (2003) who reported a marked decrease in the general protein synthesis rate following acrylamide exposure.

Supporting this view, El-Ghawet (2003) reported that fed of fried potato chips mixed with standard diet at concentration of 50%, and administration of daily oral doses of 25µg acrylamide/kg. B.W. to pregnant mothers of albino mice from the 6th day of gestation caused histological abnormalities in liver, kidney, heart and femoral epiphyseal cartilage. These abnormalities were striking and unexpected and exhibited highest drastic effect comparing with acrylamid treatment. Recently, Sakr (2004) reported that light and electron microscopic examination of the fetal liver maternally exposed to acrylamide during the second and third week of pregnancy, showed cytoplasmic vacuolation, mitochondrial and rough endoplasmic reticulum damage.

The using of curcumin as a protective agent against different diseases has been reported (Jain and De Filipps, 1991 and Devasena *et al.*, 2002). The protective effect of curcumin against the adverse effects of fried potato feeding could not clearly detect in the present findings. Whereas slight ameliorative effect could be detected in the liver of female rats and their progeny fed on diet containing fried potato simultaneously with curcumin. On the other hand, Woo *et al.* (2003) demonstrated that curcumin can cause cell damage and induced cytotoxicity.

Devasena *et al.* (2002) examined the protective effect of a curcumin analog [bis-1,7-(2-hydroxyphenyl)-hepta-1,6-diene-3,5-dione] on hepatic lipid peroxidation and antioxidant status during 1,2-dimethylhydrazine-induced colon carcinogenesis in male Wistar rats. They observed that curcumin analog exerted chemopreventive effects against cancer development at extrahepatic sites by modulating hepatic biotransformation enzymes and antioxidant status. Tetrahydrocurcumin, an antioxidative

substance that is derived from curcumin by hydrogenation, has been shown to have a protective effect on oxidative stress in cholesterol-fed rabbits (Naito *et al*., 2002). Also, similar work proved that ethanol: water (1:1) extract of rhizomes of *Acorus calamus* prevent the neurotoxic effects induced by acrylamide in rats (Shukla *et al*., 2002).

Depending on the available results, more studies will be needed to evaluate the protective effect of curcumin and its derivatives against cytotoxicity induced by different chemicals.

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