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ANTIOXIDANT ACTIVITIES OF SOME VEGETABLES IN EXPERIMENTAL MICES

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ABSTRACT

This study was carried out to evaluate the antioxidant (anticarcinogenic) effect of some vegetables (celery, green pepper and broccoli) to prevent the risk of precrcinogenic diets (10% oil used several times for fring) in the experimental animals. The results showed that the animals treated with vegtables exerted a positive effect on daily body weight, food conversion ratio, triglyceride, low density lipoprotein cholesterol (LDLc,) cholesterol, and total lipid. The inhibitory effect of vegetables on liver and renal function which was demonstrated by decreased concentrations of aspartate amino transferase (AST), (ALT), uric acid and creatinine. It could be also shown from results that vegetables diet caused significant effect in some blood parameters and blood picture. Also, laboratory analysis had beneficial effect on relative weight of liver, spleen, kidenys and brain. The experimental rats showed different pathological changes varied from mild to sever inflammation and adenoma in different organs of spleen, brain, liver, and kidneys. From this study, it is concluded that, consumption of vegetables can lower the risk of very fried oil.

INTRODUCTION

Dietary antioxidants protect against free radicals such as reaction oxygen species in the human body (Nilsson *et al.*, 2004). Free radicals are known to be a major contributor to degenerative diseases of aging (Atoui *et al.*, 2005), cancer and atherosclerosis (Ames *et al.*, 2002). During cooking the fat or oil was keeping hot (about 180 °c) for long periods of time and was exposed to both moisture and oxygen. It was noticed a gradual increases in peroxide value, benzidine value, acid value, absorbance at 400 and 450 nm and the amount of polymeric and oxidized glycerids during frying heating up to 35 hr. Complex chemical and physical changes occurred under these condition caving fat deterioration with might reach a point where the flavor, odor, color, nutritional value and safety of the food might be affected (Becher *et al.*, 1998). Eating a lot of fat in the diet causes problems, whether it's toxic compounds that come out through the heating process such 4-hydroxy-trans-2-nonenal (HNE), alcohol, aldehyeds and hydrocarbons, or other compounds of the fat such as high cholesterol levels (Menendez *et al.*, 2002). Mammalian cells possess elaborate defense mechanisms for radical detoxification. Antioxidants are agents, which scavenge the free radicals and prevent the damage caused by them. Some of these compounds are of exogenous nature and are obtained from food (Halliwell and Gutteridage, 1998). Several antioxidant mechanisms counterbalance the potential deleterious effect of reactive oxygen species (ROS). Among the enzymatic ROS scavengers are superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPX) (Brigelius–Flohe, 1999). Also, there are hydrophilic radical scavengers such as ascorbic acid (Vitamin C), urate and glutathione as well as lipophilic radical scavengers such as tocopherol (Vitamin E), carotenoids and flavonoids. Therefore, vegetables could conceivably protect against the molecular effects of lipid peroxidation, free radicals and ROS and also delay the progress of many chronic diseases (Lai *et al.*, 2001 and Gulacin *et al.*, 2003).

The present study was designed to determine some chemical composition in some vegetables (green pepper, celery and broccoli) as antioxidant sources. We focused our attention on its effects on the serum lipid pattern parameters, liver function, kidneys function, test of some blood indicators and enzymes, as well as the histopathological examination of liver, kidneys, spleen and brain.

MATERIALS AND METHODS

Materials :-

Fresh vegetable: Celery (*Apium graveolens*), broccoli (*Brassico oleracea*) and green pepper (*Capsicum annum*) were obtained from the local markets and were dried at 60 °C, and crushed to powder.

Oil used several time for frying for not less about 50 hours .

Experimental animals; this work was carried out 45 adult male of white albino mices of Sprague dawely strain. The mean weight of male rats ranged from 16 to 19 g. Provided from National Research Center, Cairo, Egypt, were housed as groups in wire cages under the normal laboratory conditions and fed on basal diet according to NRC 1995 . Food and water was provided ad-libtum. The standard diet was composed as following : - 200g casein, 497g Corn starch, 100g Sucrose, 20g Vitamin mixture, 100g Mineral mixture, 50g Corn oil, 3g DL- methionine, 30g Cellulose.

Methods :-

Chemical Analysis :-

Moisture, ash, crude protein, crude fat, and crude fiber were determined according to the methods of the (A.O.A.C.1990). While, total carbohydrates were estimated by subtracting the difference from initial weight of the samples as follows :

% Carbohydrates = 100 – (% moisture + % protein + % fat + % ash + % fiber).

Iron, Calcium and zinc determination were conducted following the methods of (Pearson, 1970). Phosphorus was determined according to colorimetric method as described by (Page, 1982).

Selenium content was determined by using the methods described by (Carnrick *et al.*, 1983 and Pupsa *et al.*, 1994). Carotenoids was determined by using the method described by (Wettestein, 1957). Ascorbic acid was determined by using 2.6 dichlorophin-olindophinol titration methods as described by (Ranganna, 1979).

Determination of total phenols compounds (mg/100 g of dry matter) was determined by using the method described by (Daniel and George 1972 and A.O.A.C. 1990).

Experimental rats design; The rats were randomly classified as follows :-

First group, Rats fed on basal diet (negative control) .

Second group, Rats fed on basal diet with 10% of oil used several times for frying (positive control).

Group 3, Rats fed on diet contained 10% of oil used several times for frying supplemented with 10% of dried green pepper .

Group 4, Rats fed on diet contained 10% of oil used several times for frying supplemented with 10% of dried celery.

Group 5, Rats fed on diet contained 10% of oil used several times for frying supplemented with 10% of dried broccoli .

The duration of the study was four weeks. The rats were subjected daily to physical examination for observation of healthy condition such as external appearance, color of hair, body condition and activity of rats. Food intake was recorded daily and body weight of rats was measured once weekly. The total body weight gain and food intake during the experimental period (4 weeks) were also calculated. Food convention ratio was calculated at the end of experiment as follows: -

Food conversion ratio = body weight gain (g) / food intake (g).

At the end of the experiment period, the rats were anaesthetized by diethyl ether and sacrificed . Blood samples of each rat were withdrawn in two test tubes. The whole blood in the heparenized tube was used for estimation of some biochemical analysis and also to obtain blood pictures . The other tubes of blood were left for coagulation then centrifuged at 3000 rpm for 15 minutes to obtain serum for further analysis.

Spleen, liver, kidneys and brain for every rat were collected and weighted then immersed in 10 % neutral buffered formalin as fixative and then sent to Pathological Department of Veterinary Medicine, Cairo University for histopathological examination.

Serum cholesterol was measured according to Richmond, (1973). Serum triglyceride (TG) was determined using the method of Buccolo and David, (1973). Serum high density lipoprotein cholesterol (HDL-c) was determined according to Grodon and Amer, (1977). Serum total lipid was measured according to Knight *et al.*, (1972) . Low density lipoprotein cholesterol (LDL-c) and phospholipids were calculated according to Lee and Nieman, (1996). Total lipids was estimated according to Zollner and Kirsch, (1962). Serum lanine amino transferase (ALT) and aspartate amino transferase (AST) enzymes activity were determined according to the method of Reitman and Frankel, (1957). Serum uric acid was estimated according to Trinder,(1969) Serum creatinine was estimated according to Henry, (1974). Serum glucose level was estimated according to Trinder, (1969). Hemoglobin was estimated according to Drabkin, (1949). Packed cell volume (PCV) hematocrit was estimated according to Mc-Inory, (1954). Bood glutathione peroxidase was estimated according to Beuther *et al.*, (1987) Srum glutathione-S-transferase activity was estimated according to Yaggi, (1987). Speroxide dismutase (SOD) was estimated according to Beuchamp and Fridovich, (1971). Lipid peroxides was estimated according to Yaggi, (1987).

Histopathological examination:- The fixed samples of spleen, kidney and brain in 10 %neutral buffered formalin were cleared in xylol and embeded in paraffin 4-5 µm thick section were prepared and stained with Hematoxylin and Eosin (H&E) for subsequent histopathological examination Bancroft *et al.*, (1996).

All the obtained data were statistically analyzed by SPSS computer soft ware (Fouad, 1978).

RESULTS AND DISCUSSION

The chemical composition of dried green pepper, celery and broccoli are given in Table (1) . The results of the analyses were established to give nutrient values per 100g. Crude protein, fat, fibers, ash and carbohydrates were found 13.5%, 1.82%, 16%, 16.64 % and 43.39% in green pepper respectively, while, 19.29%, 1.46%, 11.3%, 8.48% and 40.67% in celery respectively, and 23%, 1.38%, 14.6%, 6.83% and 45.59% in broccoli respectively.

Mineral contents, vitamins and total phenolic compounds are presented in the same table as mg/100g. According to results, calcium (Ca), phosphorus (P), iron (Fe), zinc (Zn), selenium (Se), vitamin (C) beta-carotene and total phenolic compounds were 10 mg, 200 mg, 10 mg, 0.019 mg, 0.026 mg, 263.8mg, 0.26 mg and 62.3 mg in green pepper respectively, and 15.5 mg, 270 mg, 70 mg, 0.029 mg, 0.022 mg, 77.35 mg, 0.60 mg and 13.9 mg in celery respectively, while, 20 mg, 310 mg, 10 mg, 0.036 mg, 0.027 mg 237.9 mg, 0.08 mg and 176.2 mg in broccoli respectively.

Table (1): Chemical composition and total phenolic compounds of different dried raw material.

Samples / Components	Vegetables (D.W.B.)		
	Green pepper	**Celery**	**Broccoli**
Protein %	13.50	19.29	23.00
Fat %	1.82	1.46	1.38
Ash %	16.64	8.48	6.83
Carbohydrate, by difference %	43.39	40.67	45.59
Fiber %	16.00	11.30	14.60
Calcium, Ca (mg/100g)	10.00	15.50	20.00
Phosphorus, P (mg/100g)	200.00	270.00	310.00
Iron, Fe (mg/100g)	10.00	70.00	10.00
Zinc, Znn (mg/100g)	0.019	0.029	0.036
Selenum, (mg/100g)	0.026	0.022	0.027
Vit. C (mg/100g)	263.80	77.35	237.00
Beta-carotene (mg/100g)	0.26	0.60	0.08
Total phenolic compounds (mg/100g)	62.30	13.90	176.20

Data in table (2) showed the values of body weight gain, food intake and food conversion ratio of the control and experimental rats.

It is noticeable from this table (2) that rats fed on pre-carcinogenic diet (10% oil used several times for frying) and 10% green pepper or 10% celery or 10% broccoli recorded the highest body weight gain 17± 2.74 g, 17± 2.10 g and 17± 2.48 g respectively . The improvement in body weight gain of rats fed on antioxidant diets related to beneficial effect of antioxidants in retarding the harmful effect of frying oil on reducing body weight gain. These one results are in agreement with those reported by Liu and Lee, (1998).

The data of the same table show the body weight gain per day. These is no significant difference (P < 0.05) between negative control and positive control, (0.57± 0.06 gm/d/rat) and (0.59± 0.06 gm/d/rat). While, these are significant difference (P < 0.05) between these two groups and experimental groups. The same table show that these is no significant difference of daily food intake between all groups of experimental animals. The data show significant decrease difference of food conversion ratio between the positive control (25.4 ± 0.44 ratio) and all treatment groups. These result are agree with those reported by (Gad, 2000).

Table (2): Mean values ± SD of body weight gain, food intake and food conversion ratio of rats fed on precarcinogenic (frying oil used several times) and antioxidant (anticarcinogenic) diets.

Groups	Initial Wight (g)	Final Wight (g)	Body W. gain (g)	Daily body Wight (g)	Daily food Intake (g)	Food conversion (ratio)
Group (1): Negative control Basal diet	19.0 ± 1.35 ^{a}	35.0 ± 3.10 ^{a}	16.0 ± 1.80 ^{b}	0.57 ± 0.06 ^{c}	15.0 ± 2.28^a	26.35 ± 0.19 ^{a}
Group (2): Positive control Basal diet + 10% frying.	19.5 ± 2.21 ^{a}	35.0 ± 1.10 ^{a}	16.5 ± 1.41 ^{b}	0.59 ± 0.06 ^{b}	15.0 ± 3.94^a	25.4 ± 0.44 ^{a}
Group (3): Green Pepper Basal diet + 10% frying.	16.0 ± 1.31 ^{c}	33.0 ± 2.47 ^{a}	17.0 ± 2.74 ^{a}	0.61 ± 0.06^a	15.0 ± 1.30^a	24.00 ± 0.44 ^{b}
Group (3): Celery Basal diet + 10% frying.	16.0 ± 1.20 ^{c}	35.0 ± 1.50 ^{a}	17.0 ± 2.10 ^{a}	0.61 ± 0.06 ^{a}	15.0 ± 1.36^a	24.1 ± 0.50 ^{b}
Group (3): Broccoli. Basal diet + 10% frying.	18.0 ± 0.32 ^{b}	35.0 ± 1.05 ^{a}	17.0 ± 2.48 ^{a}	0.61 ± 0.06^a	15.0 ± 1.35^a	24.6 ± 0.06 ^{b}

- **Significant at (P < 0.05 P < 0.01) .**

Mean values in each column having different superscript (a,b,c,d,….) are significantly different at P < 0.05 .

Results in table (3) showed the plasma total cholesterol content, triglycerides, HDL-c, LDL-c and total lipids of rats fed on all investigated diets. The results indicated increases in plasma total cholesterol content of rats fed on either frying oil with broccoli (36.0 ± 0.71 mg/dl) or those fed on fried oil alone (35.5 ± 1.07 mg/dl). The lowest value of total cholesterol is (31.5 ± 1.01 mg/dl) for group fed on green pepper. The same table show that the highest value of triglycerides is (22.5 ± 2.42 mg/dl) for the positive control group. While for tested groups, the group fed on green pepper show the lowest value of triglycerides (17.5 ± 1.52 mg/dl) , followed by group fed on celery (20.5 ± 2.02 mg/dl) . The data in the same table revealed that the highest value of HDL-c is (18.5 ± 0.25 mg/dl) for the group fed on broccoli. There are significant difference (P < 0.05) between the positive control and the experimental groups. The hypo cholesterol mice effects of plant occur by the increase of HDL-c level which led to increase in the follow of cholesterol to liver converted to other compounds (Ibrahim, 1997).

Table (3): Mean values ± SD of some serum lipid pattern parameters (mg/dl) of rats fed precarcinogenic(frying oil used several times) and antioxidant (anticarcinogenic) diets.

Variable / Groups	Total Cholesterol	Triglycerides (T.G)	HDL-c	LDL-c	Total Lipids
Group (1): Negative control Basal diet	33.0 ± 0.85 [b]	20.0 ± 0.63[b]	15.0 ± 0.25 [c]	41.5 ± 0.30 [c]	430 ± 7.02 [c]
Group (2): Positive control Basal diet + 10% frying.	35.5 ± 1.07 [b]	22.5 ± 2.42 [a]	17.0 ± 0.13 [b]	56.8 ± 8.35 [a]	580 ± 9.06[a]
Group (3): Green Pepper Basal diet + 10% frying.	31.5 ± 1.01 [c]	17.5 ± 1.52 [c]	18.3 ± 0.66 [a]	48.3 ± 2.79 [b]	490 ± 4.61[b]
Group (3): Celery Basal diet + 10% frying.	34.8 ± 1.10 [b]	20.5 ± 2.02 [b]	18.0 ± 0.04[a]	53.6 ± 7.53 [a]	533 ± 9.75 [a]
Group (3): Broccoli. Basal diet + 10% frying.	36.0 ± 0.71 [a]	22.0 ± 2.13 [a]	18.5 ± 0.25 [a]	55.7 ± 1.21 [a]	570 ± 2.28 [a]

- **HDL-c : High density lipoprotein cholesterol .**
- **LDL-c : Low density lipoprotein cholesterol .**
- **Significant at ($P < 0.05$, $P < 0.01$) .**
- **Mean values in each column having different superscript (a,b,c,d,….) are significantly different at $P < 0.05$.**

Also, results in table (3) revealed that the highest value of LDL-c is (56.8 ± 8.35 mg/dl) for the positive control. While, the group fed on green pepper show the lowest value of LDL-c (48.3 ± 2.79 mg/dl), followed by group fed on celery (53.6 ± 7.53 mg/dl) and group fed on broccoli (55.7 ± 1.2 mg/dl). The same table (3) revealed that the total lipids in rats serum show significant increase difference ($P < 0.01$) between the positive control (58.0 ± 9.06 mg/dl) and the negative control (43.0 ± 7.02 mg/dl). The value is decrease significantly upon group fed on broccoli, group fed on celery and group fed on green pepper, (57.0 ± 2.28 mg/dl), (53.3 ± 9.75 mg/dl) and (49.0 ± 4.61 mg/dl) respectively. These result are in line with Ibrahim, (1997) who found that, feeding of hyper lipidemic rats on diets containing some of natural products as protein leads to reduce the level of total lipids.

Table (4) demonstrate the effect of pre-carcinogenic (10% oil used several times for frying) and antioxidant (anti-carcinogenic) diets on some blood antioxidant enzymes. With respect to super-oxide dismutase (SOD) the positive control recorded the lowest value (23.85 ± 1.95 u/ghb), which show significant increase difference ($P < 0.01$) with the other groups. Increased significantly up heave treatment to (46.23 ± 1.12 u/ghb) for group fed on green pepper, (30.67 ± 1.51 u/ghb) for group fed celery and (29.15 ± 1.54 u/ghb) for group fed broccoli. Also, the data which tabulated in table (4) revealed that the highest value of glutathione peroxides (GPX) is (49.4 ± 0.92 u/hgb) for the negative control while the highest value of the other experimental groups is the group fed on green pepper (36.4 ± 0.58 u/ghb). There is significant difference ($P < 0.05$) between the positive control and other experimental groups. Glutalhions-S-tranferase level for the control and experimental groups of animals are presented in the same table (4).

Table (4): Mean values ± SD of oxidant/ antioxidant enzymes status (SOD – GPX – GSH – LP) of rats fed on(frying oil used several times) and antioxidant diets.

Variable Groups	SOD μ /ghb	GPX μ /ghb	GSH Mmol/ghb	LP Mmol/ml
Group (1): Negative control Basal diet	60.79 ± 1.01 [a]	49.4 ± 0.92 [a]	5.62 ± 0.38 [a]	2.2 ± 0.94 [b]
Group (2): Positive control Basal diet + 10% frying.	23.85 ± 1.95 [d]	18.6 ± 0.05 [c]	3.21 ± 1.19 [b]	4.8 ± 0.51 [a]
Group (3): Green Pepper Basal diet + 10% frying.	46.23 ± 1.12 [b]	36.4 ± 0.58 [b]	6.8 ± 0.96 [a]	2.94 ± 0.31 [b]
Group (3): Celery Basal diet + 10% frying.	30.67 ± 1.51 [c]	18.6 ± 0.40 [c]	3.8 ± 0.65 [b]	4.9 ± 1.53 [a]
Group (3): Broccoli. Basal diet + 10% frying.	29.15 ± 1.54 [d]	17.1 ± 0.94 [c]	3.42 ± 0.78 [b]	4.5 ± 0.34 [a]

SOD : Superoxide dismutase .
Significant at (P < 0.05 , P < 0.01) .
GPX : Glutathione peroxidase .
a,b,c,d,e : Means within the same column with different superscripts are significant .
GSH : Glutathione . -
LSD : (Least significant difference) .
LP : Lipid peroxides (MDA) .

The highest value of (GSH) is (6.8 ± 0.96 Mmol/ghb) for group fed on green pepper. The positive control increased significantly up heave treatment to (3.8 ± 0.65 Mmol/ghb) and (3.42 ± 0.78 Mmol/ghb) for group fed on celery and group fed on broccoli respectively. The increase of (SOD) (GPX) and (GSH) for group fed on green pepper may be due to its high amount of vitamin C. this are agree with (Johnston *et al.*, 1999). Results in the same table (4) showed that the highest value of lipid peroxide (LP) is (4.9 ± 1.53 Mmol/ml) for group fed on celery. Then it decreases upon to (4.5 ± 0.34 Mmol/ml) and (2.94 ± 0.31Mmol/ml) for group fed on broccoli and group fed on green pepper respectively. There are significant increase of lipid peroxide difference (P < 0.01) between the positive control (4.8 ± 0.51 Mmol/ml) and the negative control (2.2 ± 0.94 Mmol/ml). These results consider as indicator for increase free radical and are in line with the study by Garcia, (2003).

Results in Table (5) showed some values of liver and renal function parameters in the control and treatment rats. The results indicated increases of (ALT), (AST) in rats fed on fried oil alone and there is significant decrease difference (P < 0.01) between positive control and all groups of experimental animals. These results can be confirmed by the results obtained by (Battino *et al.*, 2002) and (Ibrahim *et al.*, 2005).

Healthy kidneys take uric acid and creatinine out of the blood and put them in the urine to leave the body. When kidneys were not working well, uric acid and creatinine builds up in the blood (Obata *et al.*, 2000). Results in Table (5) showed increase in uric acid of rats fed on fried oil alone compared with negative group and the other groups. The lowest value of uric acid is (7.5 ± 0.06 mg/dl) for group fed on green pepper, then it increased to (8.3 ± 0.03 mg/dl) and (8.8 ± 0.03 mg/dl) for group fed on celery and group fed on broccoli. Regarding, creatinine there is significant difference (P < 0.01) between positive control and negative control. While, there are no significant difference (P < 0.01) of creatinine between positive control and all groups of experimental animals .The biochemical results obtained are in agreement with Hropot *et al.*, (2001).

Table (5): Mean values ± SD of kidney/liver functions tests (ALT - AST - Uric Acid - Creatinine) & hematological parameters (Sugar - Hb - PCV) of rats fed on (frying oil used several times) and antioxidant diets.

Variable / Groups	ALT μ /L	AST μ /L	Uric Acid mg/dl	Creatinine mg/dl	Glucose mg/dl	Hemoglobin mg/100 ml	PVC mg/100 ml
Group (1): Negative control Basal diet	204 ± 7.50 [b]	153± 7.22[c]	8.3 ± 0.05 [a]	1.2 ± 0.01 [b]	120 ± 0.98[b]	5.6 ± 0.02[b]	18.0 ± 0.04[b]
Group (2): Positive control Basal diet + 10% frying.	370 ± 6.77[a]	280 ± 5.80 [a]	9.0 ± 0.04 [a]	2.0 ± 0.02[a]	165 ± 0.72[a]	7.8 ± 0.20 [a]	22.9 ± 0.16[a]
Group (3): Green Pepper Basal diet + 10% frying.	122.5± 9.45[c]	235 ± 4.30 [b]	7.5 ± 0.06 [b]	2.0± 0.03 [a]	83 ± 0.70[c]	4.3 ± 0.01[c]	13.2 ± 0.04[c]
Group (3): Celery Basal diet + 10% frying.	300 ± 2.20 [a]	114 ± 1.96[c]	8.3 ± 0.03 [a]	2.0 ± 0.06 [a]	79 ± 0.50[c]	7.8 ± 0.20[a]	20.6 ± 0.17 [a]
Group (3): Broccoli. Basal diet + 10% frying.	210 ± 4.91 [b]	103 ± 5.74[c]	8.8 ± 0.03[a]	2.0± 0.06[a]	129 ± 0.20 [b]	3.57 ± 0.01[c]	11.0 ± 0.01[c]

(ALT) : Alanine aminotransferase . (AST) : Aspertate aminotransferase .
PCV : packeol cell volume.
a,b,c,d,e : Means within the same column with different superscripts are significant .

It could be observed from the same table (5) that the highest values of serum glucose is (165 ± 0.721 mg/dl) for positive control then, it decreased significantly (P < 0.01) for group fed on broccoli, group fed on green pepper and group fed on celery (129 ± 0.20 mg/dl, 83 ± 0.70 mg/dl and 79 ± 0.50 mg/dl) respectively. These results are comistence with the observations of Todd *et al.*, (2002). Also, data in table (5) illustrate the effect of pre-carcinogenic and antioxidant diets on hemoglobin and hematocrit (PCV) of the experimental groups. It is noticed that, there is significant increase difference (P < 0.01) between negative control and positive control. The highest value of hemoglobin and PCV of experimental groups are (7.8 ± 0.20 mg/100 ml) and (20.6 ± 0.17 mg/100 ml), for group fed on celery. While, the lowest value is (3.57 ± 0.01 mg/100 ml and 11 ± 0.01 mg/100 ml) for group fed on broccoli.

Table (6) show the weight and the relative weight of some organs of rats fed on basal diet and experimental groups of rats fed on pre-carcinogenic and antioxidants diets. The relative weight of liver has show the highest ratio (1.43 ± 0.4 ratio) for positive control, and it significantly decreases upon to (1.33±0.28 ratio) for group fed on green pepper, (1.31 ± 0.22 ratio) for group fed on celery and (1.23 ± 0.24 ratio) for group fed on broccoli respectively.

The increase in ratio of organs may related to the toxic effect of frying oil on both physiological and biological cell functions which may inturne cause cellular cell damage. The results are in agreement with these obtained by Khalil, (1999) and Ahmed, (1994). The same table show that the highest value of the relative weight of kidneys are (0.43 ± 0.07 ratio) for group fed on broccoli. Then it decreases significantly upon to (0.39 ± 0.11ratio) and (0.38 ± 0.12 ratio) for group fed on green pepper and group fed on celery respectively. The spleen relative weight of positive control show significant increase difference (P < 0.01) compared with the other groups. The lowest ratio of relative spleen weight are (0.18 ± 0.09 ratio) and (0.18 ± 0.13 ratio) for both group fed on green pepper and group fed on celery respectively. Relative brain weight show increase significant difference (P < 0.01) between the positive control and other experimental groups. The highest value of relative weight of brain is (1.09 ± 0.15 ratio) for group fed on green pepper.

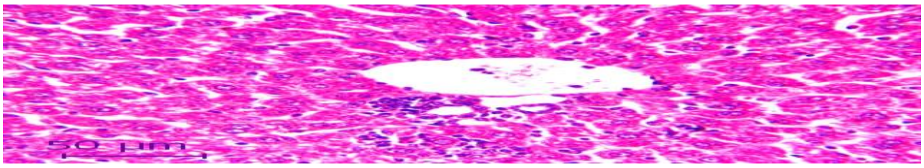
Table (6): Weight of liver, kidneys, spleen and brain of rats fed on precarcinogenice (frying oil used several times)and antioxidant (anticarcinogenic) diets.

Variable / Groups	Liver		Kidney		Spleen		Brain	
	Weight (g)	Relative (ratio)	Weight (g)	Relative (ratio)	Weight (g)	Relative (ratio)	Weight (g)	Relative (ratio)
Group (1) Negative control . Basal diet	0.48± 0.49[a]	1.37± 0.38[b]	0.15± 0.04[a]	0.43± 0.20[a]	0.07± 0.39[b]	0.20± 0.15[b]	0.36± 0.37[a]	1.03± 0.25[a]
Group (2) Positive control . Basal diet + 10% frying oil .	0.50± 0.001[a]	1.43± 0.40[a]	0.13± 0.43[b]	0.37± 0.19[b]	0.08± 0.09[a]	0.23± 0.12[a]	0.33± 0.14[c]	0.94± 0.22[c]
Group (3) . Basal diet + 10% frying oil + 10% Green pepper .	0.44± 0.39[b]	1.33± 0.28[c]	0.13± 0.11[ab]	0.39± 0.11[ab]	0.06± 0.004[c]	0.18± 0.09[c]	0.36± 0.31[a]	1.09± 0.15[a]
Group (4) . Basal diet + 10% frying oil + 10% Celery	0.46± 0.52[bc]	1.31± 0.22[b]	0.13± 0.36[b]	0.38± 0.12[b]	0.06± 0.03[cd]	0.18± 0.13[cd]	0.33± 0.32[b]	0.95 0.14[b]
Group (5) . Basal diet + 10% frying oil + 10% Broccoli .	0.43± 0.14[c]	1.23± 0.24[c]	0.15± 0.32[a]	0.43± 0.07[a]	0.07± 0.08[b]	0.20± 0.17[b]	0.35± 0.28[b]	1.00± 0.10[b]

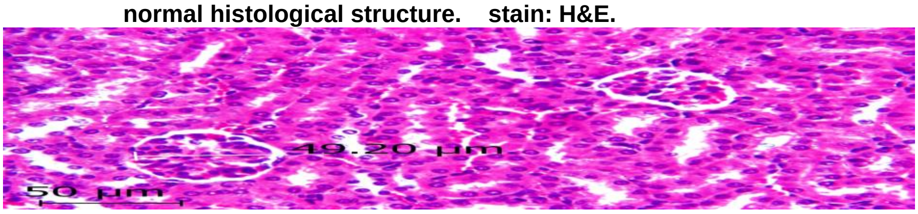
- **Significant at (P < 0.05 P < 0.01). Mean values in each column having different superscript (a,b,c,d,….) are significantly different at P < 0.05 .**

The control untreated rats showed the normal histological structure of hepatic lobule which consists of central vein and centrically arranged hepatocytes of liver (pict.1), normal histological structure of kidneys from renal cortex and renal medulla (pict.2), normal histological structure of spleen (pict.3), no histopathological changes in brain (pict.4).

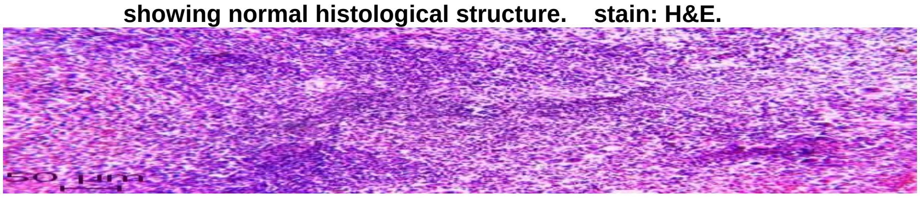
The positive control (fed on pre-carcinogenic oil used several time for fring) effect on liver, kidneys, spleen and brain. The biochemical results obtained are in agree with histopathological examination indicated that frying oil produced a toxic pre-carcinogenic effect on both physiological and the histology of organs (pict. 5,6,7, and 8). However, in groups fed on antioxidant diets shown enhancement in both physiological and histological parameters. Rats fed on green pepper are show a higher state of improvement in hematological parameters followed by broccoli and celery (pict. 9-20). These results concentrated much interest on benefit of vegetables as antioxidants (anti-carcinogenic) agents due to their photochemical content which play a critical role in the prevention of oxidative damage and associated pathologies.



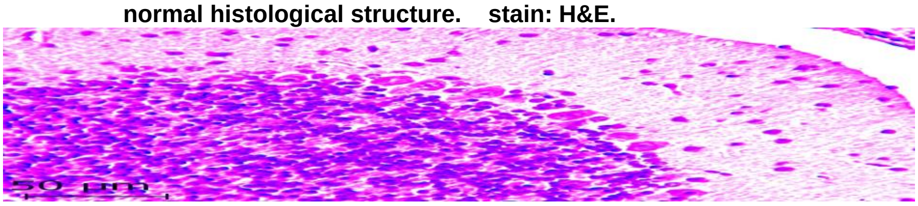
(Pict. 1):Group 1 (negative control), A photomicrograph of mice liver showing normal histological structure. stain: H&E.



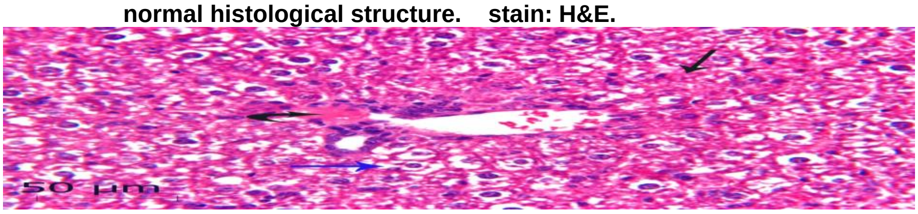
(Pict. 2):Group 1 (negative control), A photomicrograph of mice kidneys showing normal histological structure. stain: H&E.



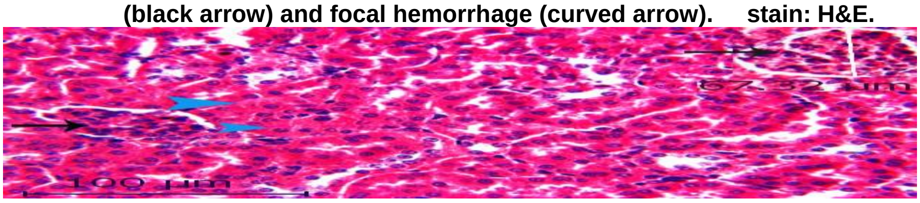
(Pict. 3):Group 1 (negative control), A photomicrograph of mice spleen showing normal histological structure. stain: H&E.



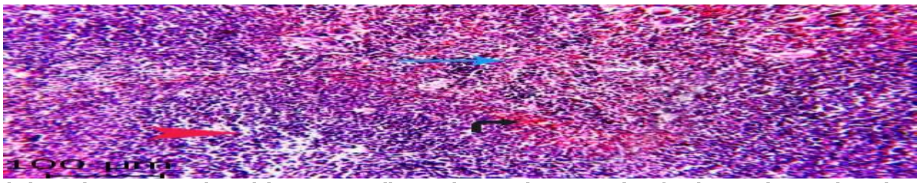
(Pict. 4):Group 1 (negative control), A photomicrograph of mice brain showing normal histological structure. stain: H&E.



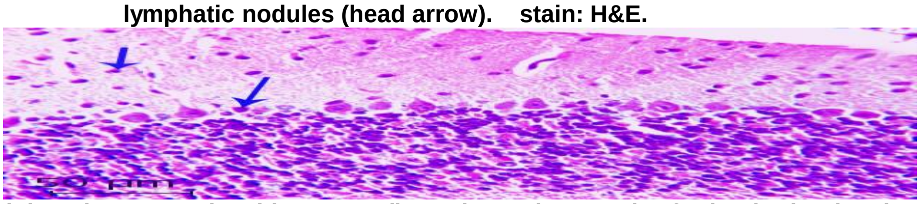
(Pict. 5):Group 2 (positive control), A photomicrograph of mice liver showing hydropic degeneration of the hepatocyts (blue arrow), damaged cell (black arrow) and focal hemorrhage (curved arrow). stain: H&E.



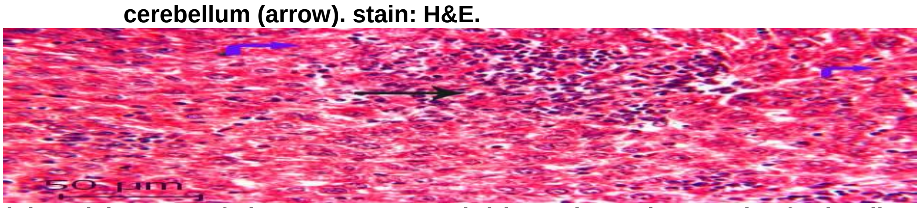
(Pict. 6):Group 2 (positive control), A photomicrograph of mice kidneys showing enlarged glomerulus (straight arrow), cloudy swelling of some tubules (head arrow) and stare shape lumen (curved arrow). stain: H&E.



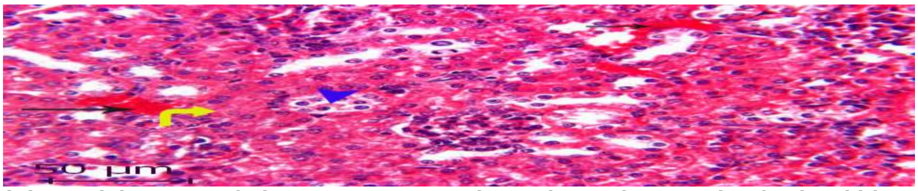
(Pict. 7):Group 2 (positive control), A photomicrograph of mice spleen showing lymphocytopenia (straight arrow), hemorrhage (curved arrow) and decrease in the number of the lymphocyte in the center of the lymphatic nodules (head arrow). stain: H&E.



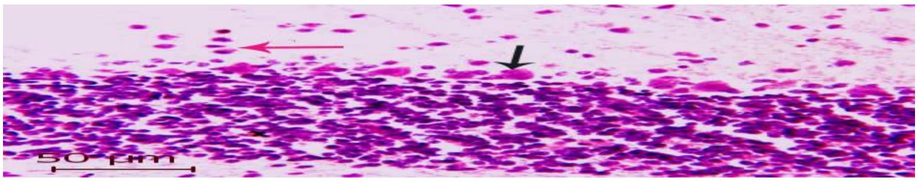
(Pict. 8):Group 2 (positive control), A photomicrograph of mice brain showing necrobiosis of some purkinje cells and molecular cells of the cerebellum (arrow). stain: H&E.



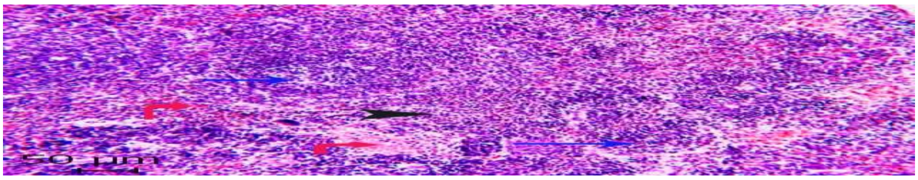
(Pict. 9):(Group 3 fed on green pepper) (4), A photomicrograph of mice liver showing hydropic degeneration of some hepatocyts (head arrow), coagulative necrosis of other cells (curved arrow) and focal aggregation of mono nuclear inflammatory cells (straight arrow). stain: H&E.



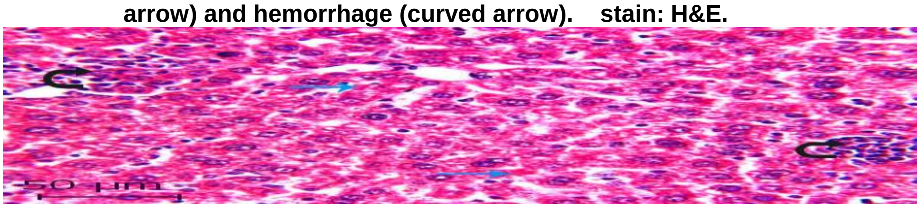
(Pict. 10):(Group 3 fed on green pepper), A photomicrograph of mice kidney showing hyaline casts (head arrow), congestion of the blood vessels (straight arrow) and cloudy swelling of some tubules (curved arrow). stain: H&E.



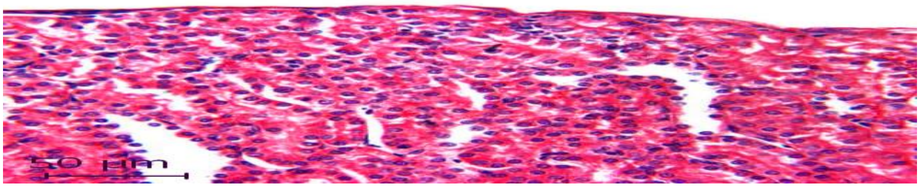
(Pict. 11):(Group 3 fed on green pepper), A photomicrograph of mice brain showing necrobiosis of some purkinje cells (black arrow) and molecular cells (red arrow) of the cerebellum. stain: H&E.



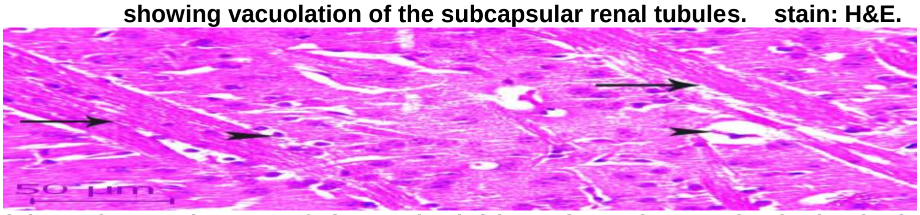
(Pict. 12):(Group 3 fed on green pepper), A photomicrograph of mice spleen showing decrease in the size of the lymphatic nodules (straight arrow) with sever necrosis of the lymphocyte in the white pulp (head arrow) and hemorrhage (curved arrow). stain: H&E.



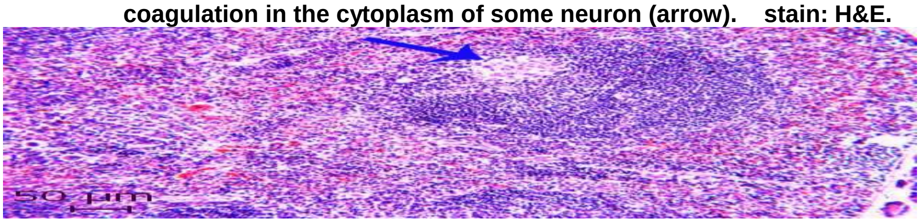
(Pict. 13):(Group 4 fed on celery) (6), A photomicrograph of mice liver showing showing hydropic degeneration of the hepatocyts (head arrow), focal hemorrhage (straight arrow) and focal aggregation of mono nuclear inflammatory cells (curved arrow). stain: H&E.



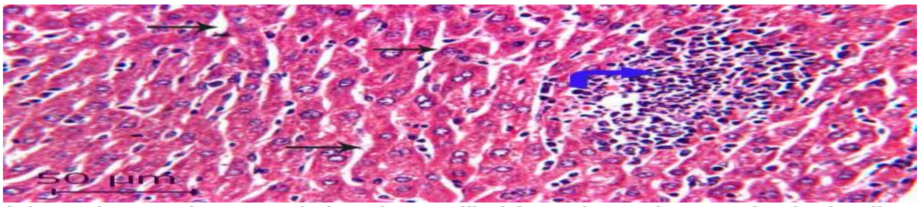
(Pict. 14):(Group 4 fed on celery) (6), A photomicrograph of mice kidney showing vacuolation of the subcapsular renal tubules. stain: H&E.



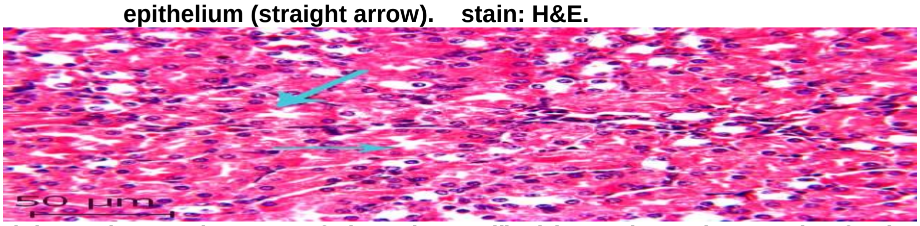
(Pict. 15):Group(Group 4 fed on celery) (6), A photomicrograph of mice brain showing prenuronal edema in the cerebrum (head arrow) and coagulation in the cytoplasm of some neuron (arrow). stain: H&E.



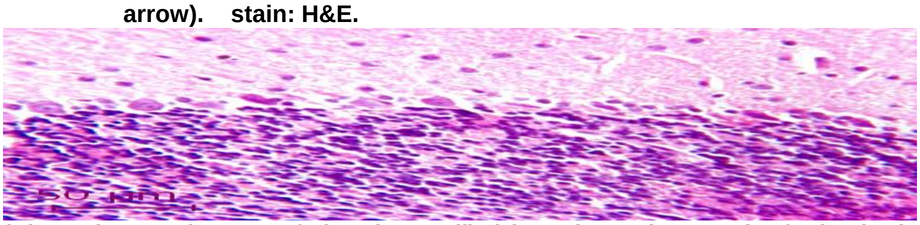
(Pict. 16):Group(Group 4 fed on celery) (6), A photomicrograph of mice spleen showing granule lymphocytopenia (blue arrow). stain: H&E.



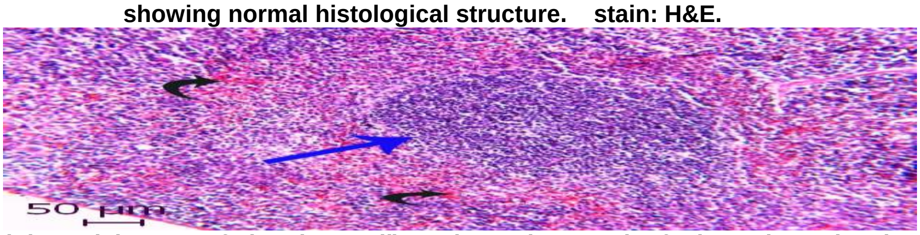
(Pict. 17):Group(Group 5 fed on broccoli), (9), A photomicrograph of mice liver showing focal aggregation of mono nuclear inflammatory cells (curved arrow) widening of the sinus and hyperplasia of its lining epithelium (straight arrow). stain: H&E.



(Pict. 18):Group(Group 5 fed on broccoli), (9), A photomicrograph of mice kidney showing stare shape lumen of the renal tubules (straight arrow). stain: H&E.



(Pict. 19):Group(Group 5 fed on broccoli), (9), A photomicrograph of mice brain showing normal histological structure. stain: H&E.



(Pict. 20):(Group 5 fed on broccoli), A photomicrograph of mice spleen showing necrosis of the lymphocyte in the white pulp (straight arrow) and mild hemorrhage (curved arrow). stain: H&E.

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نشاط المواد المضادة للأكسدة لبعض الخضروات على فئران التجارب

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أجريت هذه التجربة على فئران التجارب التى تغذت على أغذية مسرطنة (١٠% زيت مغلى لعدة مرات) لتقييم نشاط بعض الخضروات (١٠% فلفل أخضر أو ١٠% بروكلى أ و ١٠% كرفس) كمضاد للأكسدة (مضاد للسرطان).

وقد أكدت النتائج أن تناول الفئران لهذه الخضروات كان له أكبر الأثر فى تحسين كل من الزيادة فى وزن الجسم ، ونسبة كفاءة تحول الغذاء ، و الدهون الثلاثية، و والليبوبروتينات منخفضة الكثافة (LDL-c) و الدهون الكلية.

كما أثبتت النتائج أثر الخضروات المثبط للمركبات المسببة للسرطان وذلك لنقص تركيز كل من (AST)،(ALT) و حمض اليوريك في سيرم الدم و أيضا للأثر المعنوى فى كل من مقا ييس و صورة الدم . كما أظهرت النتائج الأثر الجيد على وظائف الكبد والكلي و أيضا على وزن الكبد والطحال و في نسبة وزن الكلي والمخ.

بالمقارنة بالمجموعة الضابطة سجلت نتائج التغيرات النسيجية للأنسجة الأعضاء المختبرة أن هناك تغيرات مرضية تبدأ من الحادة والمتوسطة والخفيفة مع وجود تغيرات هستوباثولوجية في معظم الأعضاء المختبرة (الطحال والكبد والكلي والمخ) . وتظهر بوضوح في الفئران المتناولة للزيت المغلى عدة مرات بدون معالجة .

وتوصى هذه الدراسة بضرورة زيادة استهلاك الخضروات المختبرة و التى كان لها أثر فعال في خفض التأثير الضار للمركبات المسببة للسرطان.

قام بتحكيم البحث

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