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HYPOCHOLESTEROLEMIC EFFECT OF SOME PLANT BY-PRODUCTS IN BAKERY PRODUCTION: BIOCHEMICAL AND HISTOLOGICAL STUDY

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ABSTRACT

The current study was conducted to investigate the hypocholesterolemic effect of a mixture of three plant by-products (orange albedo, potato peel and carrot pomace), as a source of natural fibers on hypercholesterolemia and hyperglycemia in hypercholesterolemic rats. Furthermore, production of bread containing high natural fiber contents, formed from these by-products. The diet was supplemented with 10% and 20% of this mixture. The obtained results revealed that the hypercholesterolemic rats which fed on the diet supplemented with 20% mixture showed the highest significant improve in lipid profile, liver function, kidney function, blood picture and plasma glucose compared to positive and negative control. Also the results showed a high significant decrease in body weight gain compared to positive control. In addition, the sensory evaluation of the bread which formed from these by-products showed that the addition of the mixture to the bread significantly decreased the bread quality compared with control Balady Bread, but the bread that contained 10% mix showed a high

values in [taste, aroma, mouth feel, crumb texture, crumb color, (break & shred) and crust color] compared to the bread containing 20% mix.

Keywords: Hypercholesterolemia, dietary Orange Albedo powder (OrAP), potato peel powder (PoPP) carrot pomace powder (CaPP), and dietary fibers

INTRODUCTION:

In recent years, many reports have focused on how to decrease plasma lipid concentrations and the absorption of fat in the intestinal tract to reduce diet-related chronic disease. Fibers show some potent hypolipidemic effect [1]. The relationship between fruit and vegetable intake and health has been the focal point of much scientific investigation, in attempts to identify the specific plant components that convey health benefits. Fruits and vegetables, apart from being good sources of vitamins, minerals, and fiber, are also rich sources of potentially bioactive compounds known as phytochemicals [2-3].

Dietary fiber may reduce the risk of coronary heart disease (CHD) through a variety of mechanisms, such as improving blood lipid profiles [4-5], Depending on specific characteristics, a soluble fiber has been shown to reduce triglyceridaemia in human secondary CVD risk trials [6] and in obese Zucker rats [7].

Furthermore, dietary fibers can adsorb heavy metals [8-9] and act as a potential "functional food" that reduces the incidence of CVDs [10] by reducing the risk of type-2 diabetes, body weight, serum low-density lipoprotein–cholesterol levels and adsorbing bile acids [11].

Abd El-Ghany et al. [12] found that PoPP showed a significant decrease in cholesterol, triglycerides, LDL-C and VLDL-C, and significant increase in HDL-C comparing with positive control. Singh et al. [13] found that potato peel influence both glycemic index and antioxidant status in streptozotocin (STZ)-induced diabetic male Wistar rats. In that study, diabetic rats fed on PoPP supplemented diet showed significant decrease in blood glucose levels.

A study by Singh and Rajini [14] was conducted to investigate the protective effect of PoPP against acute liver injury in rats. The results demonstrated that pretreatment of rats with PoPP significantly prevented the increased activities of AST and ALT in serum prevented the elevation of hepatic MDA formation as well as protected the liver from GSH depletion.

Carrot fiber has become of interest to food processers due to the large quantities of carrot waste created in the cut and peel carrot and carrot juice industries. Carrot pomace is the wet carrot shavings produced from carrot processing which is subsequently dried to

form a powder, the water retention and swelling capacities of carrot pomace were relatively high compared to other agricultural byproducts such as apple, pear, and orange pomace [15]. Nawirska and Uklanska [16] found that carrot pomace have the highest percentage of soluble fiber when compared with apple, cabbage, strawberry, black currant, and chokeberry pomace.

Chau and Huang [17] demonstrated that the WIFF derived from Orang peel had very pronounced hypocholesterolemic and hypolipidemic effects as compared to cellulose. It could significantly decrease the levels of serum triglyceride, serum total cholesterol, liver total lipid, and liver cholesterol, the hypocholesterolemic action of WIFF is due to its ability to enhance cholesterol and bile acids excretion. WIFF could be a potential cholesterol-lowering ingredient in human diets, and offer industry an opportunity to develop new formulations of fiber-rich functional foods.

Pectin, the dietary fiber that is so effective in helping to reduce cholesterol, is present in large amounts in the white lining of citrus fruit (albedo). An easy way to increase pectin intake is to eat the white pith. Pectin is a major component of the kind of fiber that is known to lower cholesterol. Pectin is also helpful in stabilizing blood sugar. A single orange provides 3 grams of fiber, and dietary fiber has been associated with a wide range of health benefits [18].

MATERIALS AND METHODS

MATERIALS

Fresh potato peels **(***Solanum tuberosum, L.***)** were obtained from Chipsy for Food Industries Company - Egypt, fresh carrot pomace (*Daucus carota*) was obtained from local juice extraction shops, and orange was obtained from local market and the albedo was separated from the orange peels. The three samples were dried in an air-oven at 45 °C for 48 h. The dried samples were ground in a multi mill apparatus and passed through a 0.5-mm mesh sieve to obtain a fine powder. The powders were mixed well at equal weights.

Biological effects of the mixture powder:

Experimental animals and diets:

Thirty two male albino rats weighing (100-120 g) were obtained from the laboratory animal house, National Research Center, Egypt. The animals were housed individually in stainless steel cages in a controlled environment (25±2 ºC, 50-60% relative humidity and 12-hour light-dark cycle). The animals were fed ad-libitum with a basal diet and water for two weeks, and were then randomly assigned to 4 groups (8 rats each) as follows:

Group 1 (negative control): received basal diet consisting of starch 65% casein 10% corn oil 10% salt mixture 4% vitamins mixture 1%

and cellulose 10% Association of Official Agriculture Chemists, 2000.

Group 2 (positive control): received hypercholesterolemia-induced diet (high fat diet) which prepared as basal diet preparation, except that the 10% corn oil portion was replaced with 10% animal fat and it was supplemented with 1% cholesterol and 0.25% bile salts [19].

Group 3: received 80% high fat diet plus 20% mixture powder.

Group 4: received 90% high fat diet plus 10% mixture powder.

Experimental Design:

During the experimental period (6 weeks), water and diets were available ad- libitum. At the end of the experiment, all the animals were scarified by cervical decapitation. Blood samples were collected in two tubes. The first one (0.5 mL blood) was used for the determination of blood hemoglobin, red blood cells (RBCs), white blood cells (WBCs) and haematocrit (HCT), the 2nd heparinized tube was centrifuged at 2500 rpm at 37 ºC for 15 min to separate the plasma which was kept in the deep freezer for the subsequent investigation. Also, body weight, food consumption were recorded day after day.

Biochemical Analysis:

Lipid Profile:

Plasma total lipid (TL), triglycerides (TG), total cholesterol (TC), low density lipoprotein cholesterol (LDL-C) and high density lipoprotein cholesterol (HDL-C) "mg/dl" were determined according to Knight *et al*. [20], Fossati and Prencipe [21], Allain *et al*. [22], Levy [23] and Burstein [24], respectively.

Atherogenic Index (AI) was calculated according to Lee and Niemann [25].

$$\text{Atherogenic Index (AI)} = \frac{\text{Total Cholesterol} - \text{HDL-C}}{\text{HDL-C}}$$

Liver functions tests:

Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) activities were measured colorimetrically at 340 nm in plasma according to the method described by Reitman and Frankel [26]. Alkaline phosphatase (ALP) activity in plasma was determined colorimetrically at 405 nm according to the method of Rec [27].

Kidney functions tests:

For kidney functions urea in plasma was determined colorimetrically at 578 nm according to the methods described by Patton and Crouch [28] and the determination of plasma creatinine content was carried out colorimetrically at 510 nm according to the methods described by Faulkner and King [29].

Determination of glucose:

Plasma glucose level was determined colorimetrically at 510 nm according to Trinder [30].

Determination of total protein and albumin:

Plasma total protein and plasma albumin were determined colorimetrically according to the methods described by Henry [31] and Doumas and Peters [32] respectively.

Blood Picture:

Blood hemoglobin:

The concentration of blood hemoglobin was determined colorimetrically at 546 nm according to the method of International Committee for Standardization in Haematology [33].

Red blood cells (RBCs) and White blood cells (WBCs):

Red blood cells (RBCs) count and White blood cells (WBCs) count were measured according to the method of Natt and Herrick [34].

Hematocrit (HCT):

Hematocrit (HCT) is the percent volume of whole blood occupied by red blood cells and is determined by centrifuging blood in special (hematocrit) capillary tubes. It was determined according to the method of Campbell [35].

Histopathological examination:

Liver and kidney of the sacrificed rats were taken and immersed in 10 % formalin solution. The specimens were then trimmed, washed and dehydrated in ascending grades of alcohol. Dehydrated specimens were cleared in xylol, embedded in paraffin, sectioned at 4-6 microns thickness and stained with Heamtoxylin and Eosin for histopathological examination according to the method described by Carleton [36].

Preparation of bread samples:

Bread was prepared with 10 and 20 % of mixture powder by substituted with wheat flour comparing with the control balady bread. The baking formula according to the flour weight included salt (2%) and compressed yeast (3%), the used amount of water was varied.

Dough was made from wheat flour extract 79 %, flour blends were mixed in a mixer and baked into bread using the conventional straight dough process. After one hour of fermentation the dough mass was divided into loaves, rounded and allowed to rest for 20 minutes before sheeting and molding, then baked according to the conventional method.

Sensory evaluation of prepared bread:

The untrained panelists, consisting of 17 college students (10 females and 7 males) most of whom had no experience with sensory evaluation techniques, were given a general orientation session to be familiar with the test procedures, bread samples were evaluated within 6 hour after baking. Each batch of test samples had one slice of the control bread and tested bread samples of all levels while presented to each panelist in a dish labeled each sample with a three-digit random number code. The tested parameters were taste, aroma, mouth feel, crumb texture, crumb color, (break & shred), crust color and

symmetry shape. Categories were established similar to those of Kulp *et al.* [37] with a total scoring scale of 100 divided to each parameter as follows (taste (20), aroma (20), mouth feel (10), crumb texture (15), crumb color (10), (break & shred) (10), crust color (10) and symmetry shape (5)).

Statistical analysis:

Statistical analysis (standard error ''SE'') was carried out according to Fisher [38]. LSD (Least significant difference) test was used to compare the significant differences between means of treatment; Waller and Duncan [39]. The statistical package for social science SPSS (1999) program version was used for all analysis.

RESULTS

Table 1: The mean value of Body Weight Gain (g / d), Feed Efficiency Ratio and Food Intake (g / d) in the experimental rats

Parameters / Treatments	Body weight gain	FER	Food intake
G-1 Negative control	1.3 ±0.11 [bc]	0.07 ±0.00 [c]	18.0 ±0.60 [b]
G-2 Positive control	2.4 ±0.06 [a]	0.12 ±0.00 [a]	20.1 ±0.33 [a]
G-3 20% Mix	1.1 ±0.07 [c]	0.06 ±0.00 [c]	15.9 ±0.55 [c]
G-4 10% Mix	1.4 ±0.08 [b]	0.10 ±0.00 [b]	13.0 ±0.45 [d]

All values represented as mean ±S.E.; [a-d] Means with different letters are significantly different (p<0.05).

Table 2: Plasma total lipid, total cholesterol, triglycerides, LDL-C, HDL-C (mg/dl) and AI in the experimental rats

Parameters / Treatments	Total lipid	T. cholesterol	Triglycerides	LDL-C	HDL-C	AI
G-1 (Negative control)	631 ±38.1 [b]	199 ±6.2 [b]	124 ±7.6 [b]	140 ±3.46 [b]	48.4 ±3.01 [b]	3.11
G-2 (Positive control)	1252 ±36.5 [a]	388.9 ±11.8 [a]	395 ±17.2 [a]	324 ±11.5 [a]	37.8 ±3.52 [c]	9.28
G-3 (20%Mixiture)	496 ±24 [c]	157 ±3.4 [c]	73.1 ±2.2 [c]	86 ±3.26 [c]	59 ±2.22 [a]	1.66
G-4 (10%Mix)	591 ±30.2 [bc]	175.1 ±6.9 [c]	94 ±2.6 [c]	101.6 ±5.84 [c]	46.9 ±4.32 [bc]	2.73

[a-c]All values represented as mean ±S.E.; Means with different letters are significantly different (p<0.05).

Table 3: Complete blood picture of the experimental rats

Parameters / Treatments	Hemoglobin (g/dL)	RBCs Count (×million /mm³)	Hematocrit Value (%)	WBCs Count (×10³ /µL)
G-1 (Negative Control)	11.2 ±0.55 [b]	5.2 ±0.17 [a]	36.9 ±2.02 [b]	9.6 ±0.84 [a]

G-2 (Positive Control)	**9.3 ±0.55 [c]**	**3.6 ±0.18 [b]**	**29.1 ±1.03 [c]**	**3.3 ±0.24 [c]**
G-3 (20% Mix)	**13.8 ±0.53 [a]**	**5.5 ±0.21 [a]**	**42.7 ±1.62 [a]**	**7.7 ±0.6 [ab]**
G-4 (10% Mix)	**13 ±0.54 [a]**	**5 ±0.17 [a]**	**39.8 ±1.52 [ab]**	**6.7 ±1.02 [b]**

All values represented as mean ±S.E; [a-c] Means with different letters are significantly different (p<0.05).

Table 4: Plasma ALT, AST and ALP activities (IU/L) in the experimental rats

Parameters / Treatments	AST	ALT	ALP
G-1 (Negative control)	**28.4 ±2.32 [b]**	**25.6 ±2.25 [b]**	**91 ±4.81 [b]**
G-2 (Positive control)	**93.1 ±6.42 [a]**	**86 ±4.04 [a]**	**183 ±4.25 [a]**
G-3 (20% Mix)	**23.4 ±2.98 [b]**	**27.5 ±3.36 [b]**	**78.1 ±4.58 [c]**
G-4 (10% Mix)	**26.3 ±2.3 [b]**	**28.1 ±2.98 [b]**	**86.6 ±4.11 [bc]**

All values represented as mean ±S.E.
Means with different letters are significantly different (p<0.05).

Table 5: Plasma urea and creatinine (mg/dL) of the experimental rats

Parameters / Treatments	Urea	Creatinine
G-1 (Negative Control)	**26.6 ±3.1 [a]**	**0.9 ±0.08 [b]**
G-2 (Positive Control)	**12.9 ±1.73 [b]**	**1.2 ±0.07 [a]**
G-3 (20% Mix)	**27.4 ±2.8 [a]**	**0.8 ±0.07 [b]**
G-4 (10% Mix)	**26.8 ±2.78 [a]**	**1 ±0.07 [b]**

All values represented as mean ±S.E.; Means with different letters are significantly different (p<0.05)

Table 6: Plasma protein, albumin (g/dL) and plasma glucose (mg/dL) of the experimental rats

Parameters / Treatments	Total protein	Albumin	Glucose
G-1 (Negative control)	**7 ±0.26 [a]**	**4 ±0.28 [a]**	**87 ±6.67 [b]**
G-2 (Positive control)	**4.2 ±0.28 [b]**	**2.5 ±0.24 [b]**	**105 ±3.95 [a]**
G-3 (20% Mix)	**7.4 ±0.19 [a]**	**4.3 ±0.22 [a]**	**72.6 ±2.24 c**
G-4 (10% Mix)	**7.2 ±0.2 [a]**	**4.1 ±0.19 [a]**	**80.8 ±3.46 [bc]**

All values represented as mean ±S.E.; Means with different letters are significantly different (p<0.05).

Table 7: Relative organs weight (g) of the experimental rats

Organs / Treatments	Liver	Heart	Kidney	Spleen
G-1 (Negative control)	9.1 ±0.27 [d]	1.3 ±0.06 [a]	2.2 ±0.05 [c]	1.1 ±0.05 [a]
G-2 (Positive control)	20.0 ±0.85 [a]	1.4 ±0.05 [a]	2.7 ±0.06 [a]	1.3 ±0.06 [a]
G-3 (20% Mix)	11.1 ±0.63 [c]	1.3 ±0.03 [a]	2.4 ±0.06 [bc]	1.1 ±0.04 [a]
G-4 (10% Mix)	13.4 ±0.74 [b]	1.3 ±0.06 [a]	2.5 ±0.06 [b]	1.2 ±0.07 [a]

All values represented as mean ±S.E.; Means with different letters are significantly different (p<0.05)

Table 8: Effect of different levels of the mixture powder on Balady Bread sensory evaluation

Samples	Taste (20)	Aroma (20)	Mouth feel (10)	Crumb texture (15)	Crumb color (10)	Break & Shred (10)	Crust color (10)	Symmetry shape (5)	overall acceptance (100)
Balady Bread (Control)	15.3 ±0.75 [a]	15.9 ±0.51 [a]	8.5 ±0.23 [a]	11.6 ±0.40 [a]	8.5 ±0.21 [a]	8.6 ±0.38 [a]	9.4 ±0.15 [a]	4.4 ±0.17 [a]	82.1
20% Mix	8.8 ±0.16 [c]	9.6 ±0.59 [c]	2.9 ±0.22 [c]	5.4 ±0.15 [c]	5.4 ±0.15 [c]	2.5 ±0.12 [c]	5.2 ±0.18 [c]	1.5 ±0.17 [b]	41.2
10% Mix	10.4 ±0.15 [b]	11.3 ±0.22 [b]	5.6 ±0.36 [b]	9.8 ±0.16 [b]	6.5 ±0.15 [b]	5.4 ±0.17 [b]	7.1 ±0.20 [b]	4.1 ±0.12 [a]	60.1

All values represented as mean +S.E. Means with different letters are significantly different (p<0.05).

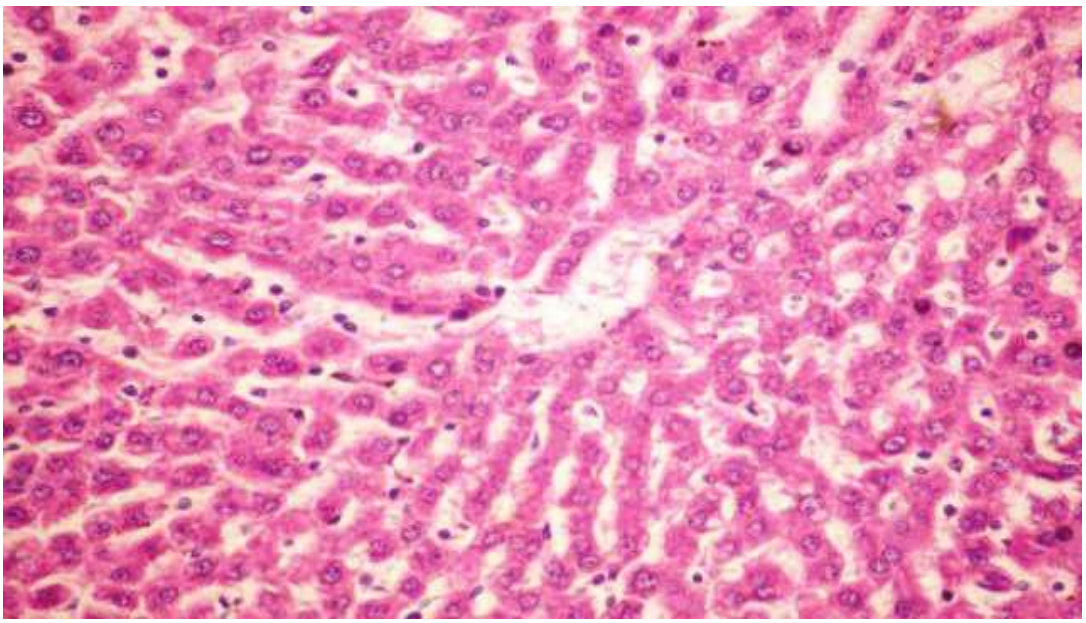

Fig. 1: Normal histological structure of hepatic lobule in rat's liver fed the basal diet "negative control group". (H and E ×400)

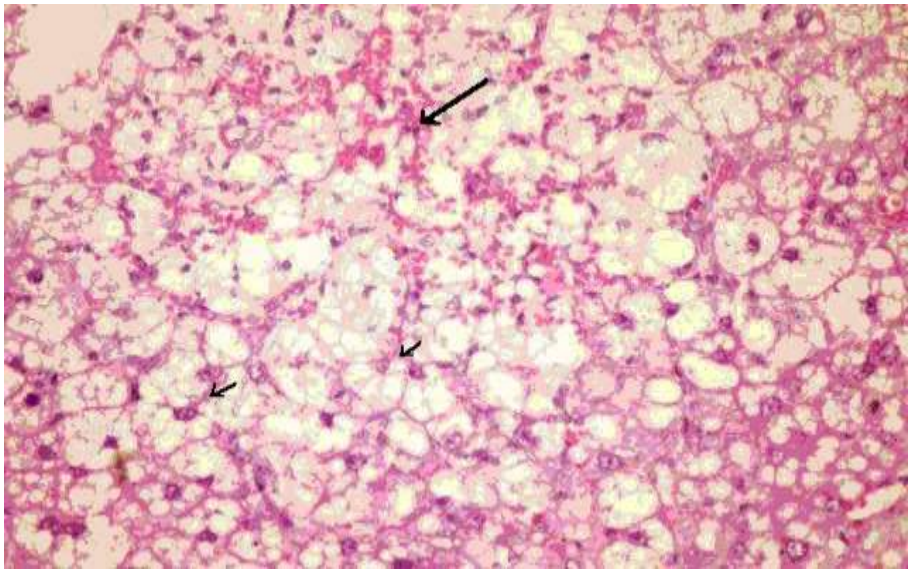

Fig. 2: Liver of rat from group 2 "positive control" showing enlarged hepatocytes, foamy cytoplasm (small arrow) as well as local hepatic hemorrhage (large arrow). (H and E ×400)

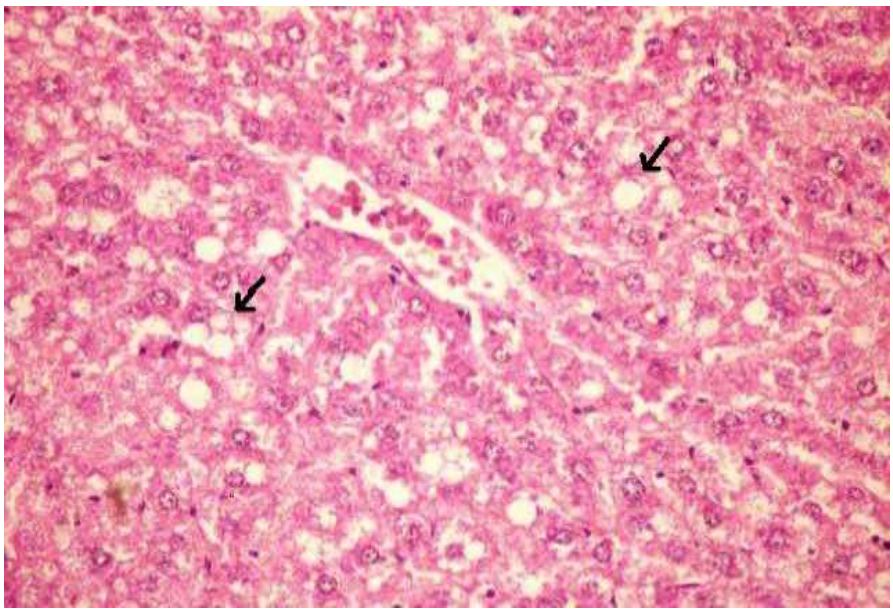

Fig. 3: Liver of rat from group 3 (20% Mix) showing fatty change of some hepatocytes (H and E ×400)

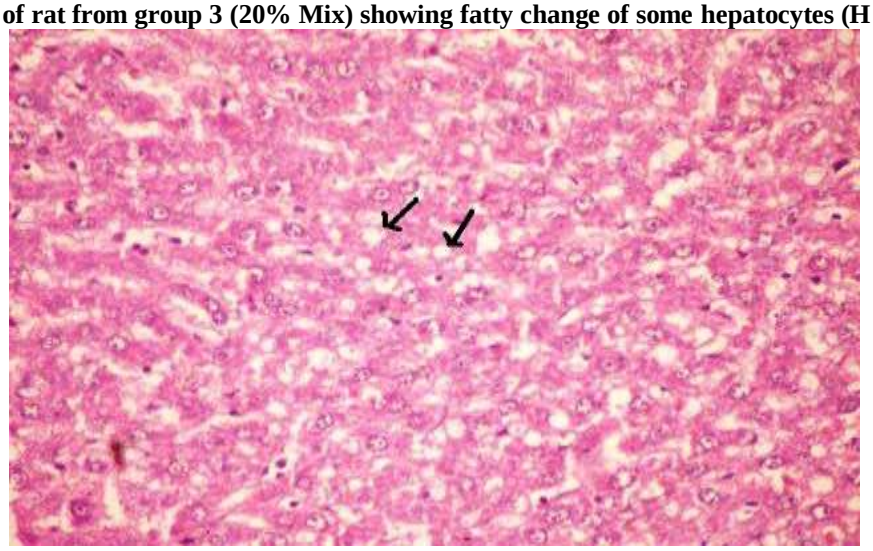

Fig. 4: Liver of rat from group 4 (10% Mix) showing fatty change of some hepatocytes (H and E ×400).

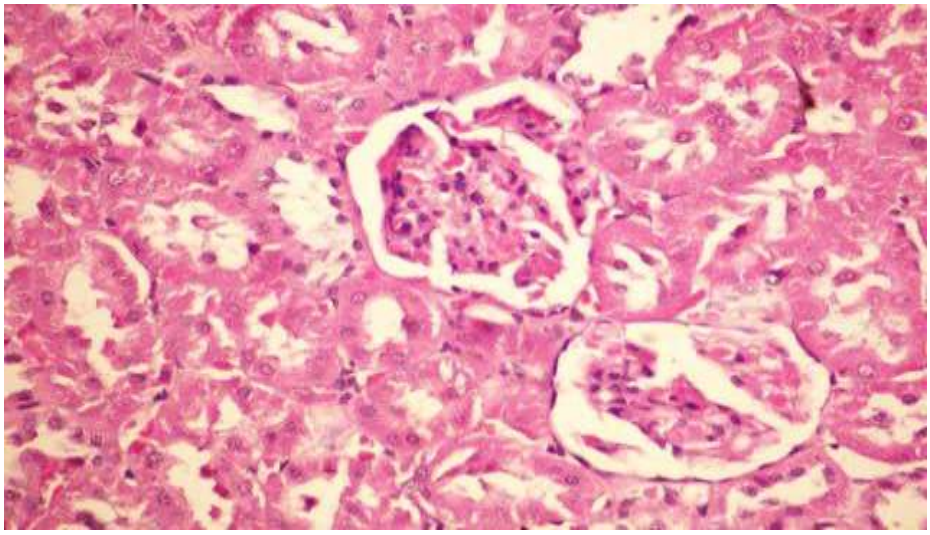

Fig. 5: Kidney of rat from group 1 (negative control) showing the normal histological structure of renal parenchyma (H and E ×400).

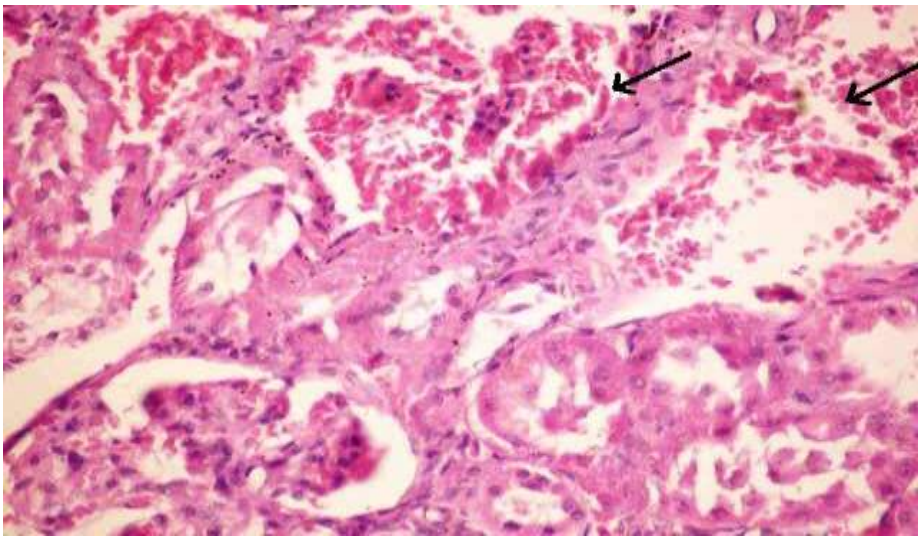

Fig. 6: Kidney of rat from group 2 (positive control) showing dilatation and congestion of renal blood vessels as well as local renal hemorrhage (H and E ×400).

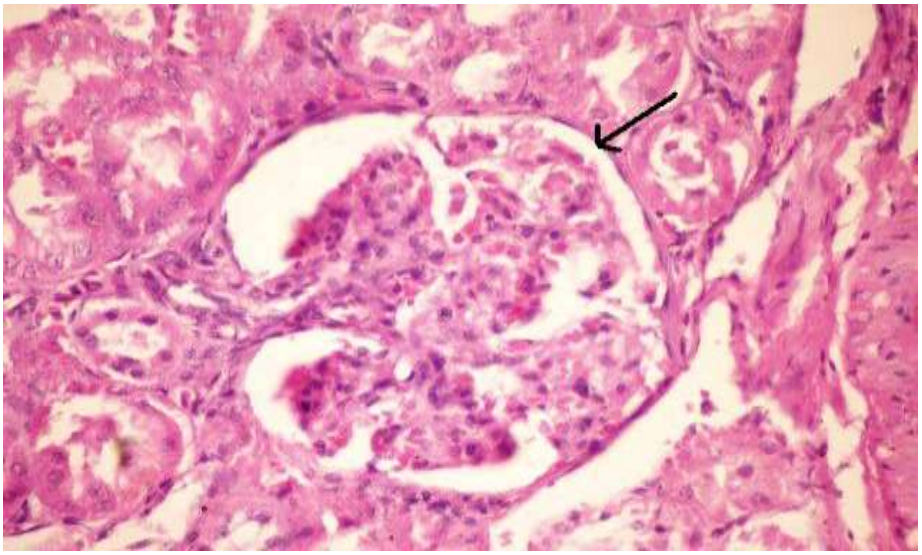

Fig. 7: Kidney of rat from group 2 (positive control) showing hypertrophy of glomerular tuft and thickening of the parietal layer of Bowman's capsule (H and E ×400).

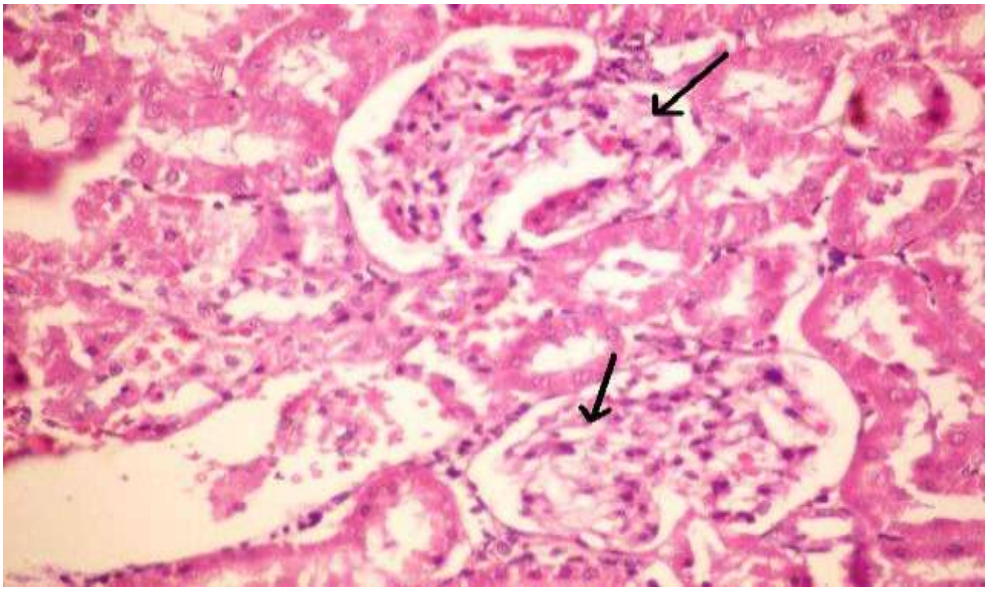

Fig. 8: Kidney of rat from group 3 (20% Mix) showing vacuolations of endothelial lining glomerular tufts (H and E ×400)

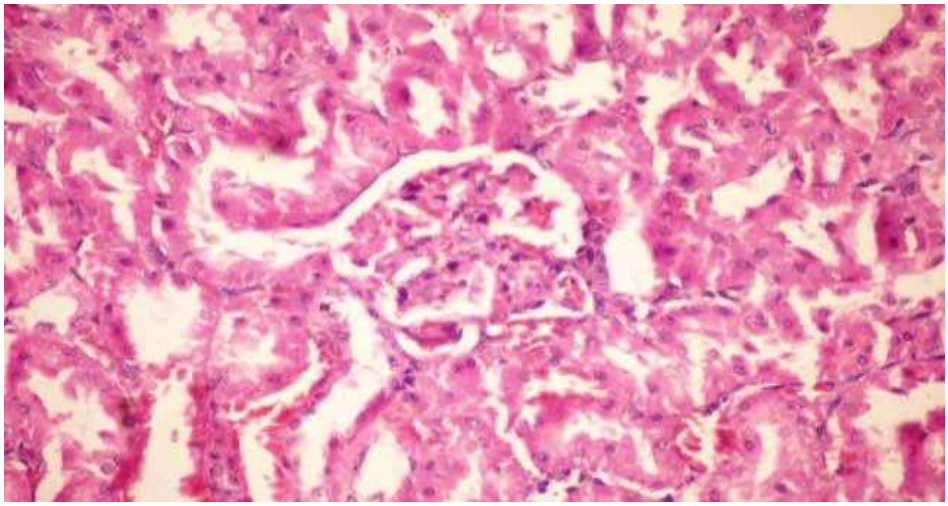

Fig. 9: Kidney of rat from group 4 (10% Mix) showing no histopathological changes. (H and E ×400)

DISCUSSION

The group of rats which fed on high ft diet (positive control) showed an increase in BWG which may be due to the presence of animal fat and cholesterol, these results agree with Milagro et al. [40] who reported that animals fed on the high fat diet showed higher body weight, fat deposition and total liver weight and increased energy intake compared with those on the standard-fat diet. Also results revealed that the reduction in BWG of the rats fed on diet containing 20% mixture indicate muscle tissue wasting this decrease may due to high content of fiber, these results were agreed with Singh et al. [13] who reported that the fiber content of diet reduce calories and losses weight, also these results are in a good agreement with Parveen et al. [41] who reported that the carrot residue fibers significantly reduce calories and body weight. However, Chau et al. [42] was on an opposite side, they found that consumption of water-

insoluble fiber rich fraction didn't affect the weights of hamsters. Also Nicolle et al., [43] were on contrary line with our study who found no significant changes on body weight gain and food intake in the groups feed on carrot diet with the control group.

Dietary factors such as continuous ingestion of high amounts of saturated fats and cholesterol are believed to be directly related to hypercholesterolemia and susceptibility to atherosclerosis. Lipid structure, composition, configuration, in addition to excessive fat and cholesterol consumption are also believed to affect the lipid profile in the plasma (Asashina et al., [44].

The intake of the designed formula in different concentrations induced significant improvement in the hypercholesterolemic state, as shown by significant increase in HDL-C concentration and significant decrease in all other lipid parameters compared to the positive control group.

Chau and Huang [17] found that the fibers derived from orange albedo could significantly decrease the levels of serum triglyceride, serum total cholesterol, liver total lipid, and liver cholesterol. And the hypocholesterolemic action of these fibers is due to its ability to enhance cholesterol and bile acids excretion, and so, it could be a potential cholesterol-lowering ingredient in human diets. On the other hand, Chau et al. [45] found that the inclusion of carrot pomace fiber in diet effectively decreased the concentrations of serum total cholesterol, serum triacylglycerides and liver cholesterol, and increased the concentrations of fecal total lipid, fecal cholesterol, and fecal bile acids, and showing pronounced hypolipidemic and hypocholesterolemic effects. Carrot pomace fibers having high cation-exchange capacity could entrap, destabilize and disintegrate the lipid emulsion, and thus reduce the diffusion and absorption of lipid as well as cholesterol [46].

The addition of mixture to the diet with 10, 20% concentrations lead to decrease in total lipid, total cholesterol, triglycerides and LDL-C but resulting increase of HDL-C. Our data was in the same line with Nicolle et al. [43] who found carrot consumption exerts a moderate lowering cholesterol effect (12% decreases). A significant 11% reduction of cholesterol has been observed in human subjects by Robertson et al. [47].

A study conducted by Hsu et al. [48], showed that insoluble fiber-rich fractions (IFRF) isolated from carrot pomace was found to have very pronounced hypocholesterolemic and hypolipidemic effects, these results suggested that the hypolipidemic and hypocholesterolemic actions of IFRF were attributed to its ability to enhance the excretion of cholesterol, lipid, and bile acids via feces.

The improvement in blood picture data was noticed especially in hemoglobin which recorded a significant increase in 20% mixture than control group also hematocrite was in significant increase in 10%, 20% mixture compared with positive control, this increase in blood picture may due to the high presence of iron. Gopalan et al. [49] found that carrot is a good source of minerals like Ca, P, Fe and Mg. On the other hand, the intake of mixture which contains orange albedo with different concentrations improved the blood picture of rats. These results could be contributed to the improvement in iron absorption from rat's gut after consumption of orange albedo due to its high content of ascorbic acid [50].

AST and ALT levels act as indicators of liver functions, hence, restoration of normal levels of these enzymes indicates normal functions of liver, the previous results indicated that the rats fed on hypercholesterolemic diet showed increased liver enzymes (AST, ALT and Alkaline phosphatase). Morphological alterations that occur in the liver affect many metabolic processes in the organism. Peroxide formation induced by hypercholesterolemia [51] result in the release of some enzymes by interacting with cellular structure and function. Thus, the serum activities of cellular enzymes such as transaminases, alkaline phosphatase, and lactate dehydrogenase do increase. With the increase in cellular membrane permeability, intracellular fluid transfers onto intercellular space, resulting in muscle and liver cell degeneration.

Rats fed on 10 and 20% mixture supplemented to hypercholesterolemic diet showed improved liver functions. This effect is mainly related to the presence of natural soluble and insoluble dietary fiber, and through increasing the plasma antioxidant activity that have been reported to protect the liver against oxidative stress in rats. Flavonoids also attenuate lipopolysaccharide-induced hepatotoxicity, possibly by preventing the cytotoxic effects of oxidants as nitrous oxide (NO) and oxygen-free radicals [52].

The present study also revealed that the ALP activity was increased when the liver functions abnormally [53], thus the study of liver ALP was done in the present study to find out the effect of the dietary fiber in the mixture on liver ALP. Results showed that mixture with 10 and 20% had a lowering effect on the activity of ALP in serum of hypercholesterolemic rats compared to the positive control, but this decrease was not significant with the negative control group. This effect is mainly related to the presence of natural soluble and insoluble dietary fiber [54].

he result of the present study also declared that mixture especially at 20% level has a significant improvement effect on the kidney

functions represented in urea and creatinine. This could be explained as consumption of food rich in dietary fibers stimulates the extrarenal route of nitrogen excretion. Younes et al., [55] found that indigestible carbohydrate/dietary fibers increased cecal weight and cecal blood flow, leading to accelerated diffusion of blood urea into the cecal lumen (by threefold), urea lysis to ammonia and protein synthesis by the microflora, and increased fecal excretion of nitrogen. Thus, reduce the role of kidney in the excretion of nitrogen and reduce blood urea concentration.

Rats fed on hypercholesterolemic diet alone (positive control) recorded increased blood glucose concentrations (hyperglycemia). These results agree with Akiyamaa et al. [56] who found that serum glucose and insulin concentrations before and after glucose loading in the rat group received high-fat hypercaloric diet (360 kcal/kg body wt./day) were significantly higher than those in the control diet (180 kcal/kg body wt./day).

The results showed that consuming of mixture at the two tested levels 10 and 20% as a source of fiber in food, markedly lowered the blood glucose levels in hypercholesterolemic rats, and these findings indicate that mixture which contains potato peel can act as hypoglycemic agents, which in true could be attributed to the presence of the phytochemicals, dietary fibers and polyphenols in potato peel as mentioned by Singh et al., [13] who found that feeding of potato peel at 5% and 10% levels to diabetic rats significantly decreased their blood glucose level. De Escalada Pla et al., [57] confirmed that the soluble fiber components which include pectic polysaccharides, may through their viscosity enhancing and gel forming properties delay gastric emptying and possibly reduce absorption rate in small intestine, a feature that is particularly impotent to diabetes.

Total protein and albumin were markedly increased with addition of 10%, 20% mixture especially with 20%, these results were on a opposite side to Eggum [58] who reported that dietary fiber had a negative influence on digestion and assimilation of proteins. Also results show a markedly decrease of glucose on hypercholesterolemic rats fed on 10 and 20% mixture. These results were confirmed by Chau et al. [59] who found that fiber has functional properties and in vitro hypoglycemic effects. Rodríguez et al. [60] declared that glucose of diabetic patients decreased by having diets rich in fiber.

The affected liver weight by hypercholesterolemia is similar to that reported by Jemai et al. [61] who found that the liver/body weight ratio increased in rats fed cholesterol rich diet compared with those fed control diet. Beynen et al., [62] reported that the appearance and weight of liver are

generally varied with the amount of cholesterol and lipid incorporated in the experimental diet. The fact that fiber source was the only variable in the ingredients of diet might explain the comparable liver weight among all groups. It could be noticed from our results that hypercholesterolemia increased organs weight especially liver and kidney compared with negative control, while their spleen and heart had no significant changes in their weight. However, 10% and 20% mixture supplemented to hypercholesterolemic treated groups had a significantly decreased in liver and kidney weights. Also these results are in agreement with Fryer [63] who reported that the increased liver weight may indicate the progression of damage in liver cell which happened due to oxidative stress in diabetic nephropathy. Although, some authors suggested that the consumption of water insoluble fiber-rich fraction as well as cellulose did not affect the weights of visceral organs of rats [42].

Examined sections of liver of rats from group (3 and 4) revealed no histopathological changes compared to positive control except slight fatty changes of hepatocytes. The use of the supplement at different concentration protect the liver cells from fatty degeneration as it lower the triglycerides level in the blood and improve all the other parameters of the lipid profile. This could be contributed to their antioxidant effects and high fiber contents of the supplements [52].

On the other hand, the experimental glomeruloscerosis is associated with hyperlipidemia and deposition of lipid in the glomeruli. Glomerulosclerosis is typically preceded by glomerular hypertrophy, and significantly correlated with proteinuria and leucocytic infiltration of the glomeruli. Mesangial foam cells derived from macrophages were associated with adhesions to Bowman's capsule. Hyperlipidemia exacerbates the development of glomerular hypertrophy and that this may be mediated by factors released during phagocytosis of lipoprotein deposits by macrophages [64]. These changes could be prevented by the use of the supplements which have high fibers content and antioxidant capacity.

The addition of mixture to the bread strongly affect the bread quality, Orr et al. [65] found that the sensory evaluation of the products made with potato peels revealed a musty aroma, while Toma et al., [66] found that the addition of peels to wheat flours at levels of 5, 10 and 15% resulted in breads of acceptable sensory quality.

The sensory evaluation of the bread indicate that the 10% concentration of mixture showed the highest value compared to 20% concentration which means that there is an inverse relationship between the concentration of sample an overall acceptance

of Balady Bread. Angioloni and Collar [67] stated that the dietary fibers with larger particle size and high viscoelastic profile resulted in highly sensory acceptable breads with higher amount of resistant starch, lower digestible starch bringing lower in vitro expected glycemic Index, and slightly lower protein digestibility.

CONCLUSION

The present study clearly refers to the possibility of using some plant by-product to develop new formulations of fiber-rich functional foods such as bakery products as a hypocholesterolemic agent and could be used in obese people for body weight loss.

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