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Prophylactic and Curative effects of Barley and its bran against Hyperlipaemia in Albino rats

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Abstract

The aim of this study was to investigate the prophglactic and curative effects of barley and it's bran against hyperlipaemia in albino rats. A total of 88 adult male albino rats "swiss strain" weighting about 80-100g were used in 2 main experiments. (**1**) The prophylactic effect of barley and bran against hyperlipaemia for 8 weeks and (2) The curative effect of barley and bran for 8 weeks after induction of hyperlipaemia (using cholesterol and cholic acid). The data revealed that there were increase in the levels of serum total lipids, total cholesterol, triglycerides, ALT, AST, ALP and LDH, while HDL-cholesterol level was decreased after the induction of hyperlipaemia. These results suggested that barley and bran may evoke different lipidaemic responses and that barley bran has more favorable effect on blood lipids than whole barley. Results were compared with those of Atorvastatin, a standard orally effective hypolipaemic agent.

Key words: Hyperlipiaemia, prophylactic, curative, barley, bran, lipid profile, liver function

Introduction

Hypercholesterolemia is a risk factor for early onset coronary heart disease. Increased consumption of dietary plant starch and non starch polysaccharides (NSP,"Fiber") and reduced consumption of total and saturated fat are known to lower plasma cholesterol [(1)].

Barley (**Hordeum vulgare L**.) contains relatively high concentration of the mixed-linkage (1-3) (1-4) β–D-glucans (β-glucan). Although β –glucan occurs in all cereals, its concentration is highest in oats and barley with values ranging from 2% - 16% [(2)]. Among the cereal grains, oats and barley have been reported to be the most effective in lowering serum total cholesterol and LDL-cholesterol in human and animals [(3-6)]. Cholesterol-lowering ability was first ascribed to oats but more recently to barley [(6)]. It has been hypothesized that, upon ingestion, β -glucan increases small intestinal viscosity due to its lower molecular weight and its tendency to form viscous gummy solution resulting in reduced bile acid and cholesterol or triglycerides absorption thus lowering plasma cholesterol [(7)] as well as altering digestive enzyme activity [(8)].

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Material and Methods

The barley was purchased from the Egyptian market . It was cleaned and powdered in a cyclotec mill to pass through a 60 mesh sieve. Atorvastatin choosed as a standard hypolipidaemic agent. It is obtained from Egyptian Int. Pharm Company; each tablet contains 10mg of the active material "vastatin".

Experimental animals; a total of 88 adult male albino rats (80-100g) were used in this study. Rats were provided from the NODCAR's Farm, Giza and allowed free access of water and fed on a standard synthetic diet for two weeks [(9)]. Hyperlipaemia was attained to rats using cholesterol/ cholic acid mixture (3:1) mixed with the synthetic diet in a dose calculated in the basis that each rat was received 0.5g of this mixture/kg b.w daily for 10 weeks. Two main experiments were conducted as follows:

1. The prophylactic effect against hyperlipaemia; to study the protective effect of the whole barley and bran against hyperlipaemia, a total of 40 rats were used and the experiment lasted for 8 weeks. Animals were divided randomly into equal five groups (8 rats each): **Group 1** fed on the standard synthetic diet and served as negative control (-ve).**Group 2** was daily attained to the hyperlipaemic diet (H.L.D) and served as positive control group (+ve). **Group 3** administered barley bran at a dose of 100mg/kg b.w (added to the H.L.D) daily. **Group 4** administered

barley bran at a dose of 200mg/kg b.w (added to the H.L.D) daily. **Group 5** administered whole barley at a dose of 200mg/kg b.w (added to the H.L.D).

2. The curative effect on hyperlipidaemic rats; in this experiment, a total of 48 rats were used. Eight rats were fed on the standard synthetic diet and served as negative control (-ve) **"Group1"**. The other rats were subjected to the induction of experimental hyperlipaemia for 10 weeks as described before. The hyperlipidaemic rats (40rats) were divided randomly into equal 5 groups (8 rats each): The first one; **Group 2** served as hyperlipaemic control (-ve control). **Group 3** received barley bran at a dose of 100mg/kg b.w. (added to the diet) daily. **Group 4** received 200mg/kg b.w.of barley bran (added to the diet) daily. **Group 5** received whole barley at a dose of 100mg/kg b.w. (added to the diet) daily. **Group6** received 0.9mg/kg b.w. Atorvastatin as a standard hypolipaemic agent (added to the diet) daily.

All doses administered to the animals were calculated according to the recommended therapeutic human dose and converted to the dose of the adult rats [(10)].

Blood sampling; in the first experiment, blood samples were collected before treatment and then after 4 and 8 weeks. In the second experiment, blood samples were collected before and after induction of hyperlipaemia and then 4 & 8 weeks after administration of the different treatments.

Analysis; AACC approved methods [(11)] were used to analyze the test samples for protein (Kjeldahl), fat (acid hydrolysis) and ash .β-Glucan content was measured [(12)]. Moisture was determined under vacuum (16 hr, 700^0 C, 25 mmHg).Total dietary fiber (TDF) and soluble fiber (SF) contents were determined [(13)]. The following parameters were assayed; serum total lipids [(14)], total cholesterol [(15)], HDL-cholesterol [(16)], triglycerides [(17)], transaminases enzymes (ALT& AST) [(18)], alkaline phosphatase (ALP) [(19)], lactate dehydrogenase (LDH) [(20)], total proteins [(21)], albumin [(22)], urea [(23)] and creatinine [(24)].The obtained results were statistically analyzed [(25)].

Results and Discussion

Clinical findings and post mortem changes

1. Chemical composition of Barley and Barley bran: The chemical composition of whole barley and barley bran was presented in Table (1). It is obvious that the whole barley contains fat higher than bran, while fat contents were 4.1 g/100g and 3.4 g/100g.Total carbohydrate amounts 51.8 g/100g and 52.1 g/100g, respectively. Barley bran contains higher amount of soluble fiber (12.5 g/100g) than whole barley (6.8 g/100g). B-Glucan concentrations were found in the same order, i.e. barley bran (13.2 g/100g) and whole barley (9.8 g/100g).

Table (1): Chemical composition of test material. **(Components g/100g)**

Material	Moisture	Protein (TN×6.25)	Fat	Ash	Total dietary fiber.	Soluble fiber	Carbo-hydrates	B-Glucan
Whole barley	10.2	11.7	4.1	2.1	20.1	6.8 (34)	51.8	9.8
Barley bran	9.4	11.2	3.4	2.0	21.9	12.5 (57)	52.1	13.2

2. Biochemical analysis

2.1. Prophylactic effect of Barley and Barley bran against hyperlipaemia: Table (2) revealed the effect of different treatments on serum lipid profile. It is clearly shown that the value of T. Lipids in the first group (-ve control) did not affected during the experimental period. In the (+ve control) group, which rats fed on the hyperlipaemic diet, serum T. Lipids were very highly significant increased by 180.2% after 8 wks of treatment compared with the corresponding control and by 98.9% , 51.0% and 88.6% for barley bran (100 , 200) and 200 mg/kg.b.w of whole barley, respectively. Serum triglycerides in the (-ve control) group did not affected during 8 weeks of treatment, while in the (+ve group), this level was increased by 144%. Barley bran in the two different doses and whole barley (200 mg/kg.b.w) causes a decrease by 5.60, 29.5 and 10.5, respectively. Serum total cholesterol level was increased by 225% in the (+ve control) group, while it was increased only by 4.11% in the negative control group. Barley bran in dose of (100,200) mg/kg. b.w. and whole barley (200 mg/kg.b.w) increases the T. Cholesterol by 36.2% , 20.8% and 37.7%, respectively. Serum HDL-cholesterol level was decreased by 19.4%, 11.4% and 17.6% in barley bran (100, 200 mg/kg.b.w) and whole barley groups, respectively. While in the (+ve control) group this value was decreased by 42.6% after 8 weeks. The risk ratio in the (+ve control) group was increased by

466%, while barley bran in two different doses and whole barley (200 mg/kg.b.w) decrease this value only by 91.8%, 35.8% and 66.7%, respectively compared to the corresponding control.

A marked elevation in serum ALT, AST, ALP and LDH by 68.2%, 89.1%, 150% and 33.5%, respectively was observed in the +ve control group after the induction of hyperlipaemia (Table3) . The daily dose of (100,200) mg/kg.b.w. of barley bran and 200mg/kg.b.w of whole barley decreases the levels of S-ALT , AST , LDH and ALP compared to +ve control group. Table (4) revealed the effect of bran and whole barley on serum total proteins, albumin, globulin and Alb / Glob. The daily dose of 200 mg/kg.b.w. of bran and whole barley caused a moderate increase in bl. urea level after 8 weeks of treatment. Both doses of barley bran and the dose of whole barley induced a slight increase in S.creatinine level after 8 weeks of treatment.

2.2. Curative effect of Barley and Barley bran on hyperlipaemic rats: In this experiment, rats were fed on the hyperlipaemic diet for 10 weeks, and then treated with the different treatments for 8 weeks. Atorvastatin was used as reference standard hypolipaemic agent. It was revealed appreciated effects on the different lipid parameters of hyperlipaemic rats after treatment for 8 weeks (Table 6). Also, this agent reduced ALT, AST, ALP and LDH levels. Bran and whole barley were also decreased these levels (Table 7). Serum total protein concentration was significantly decreased by 14.8%, 18.5%, 24.2% and 25.0% in the groups of barley bran, whole barley and Atorvastatin (Table 8), respectively. The values of serum albumin were also decreased by 31.0%, 28.6%, 30.5% and 31.7% in the groups of barley bran, whole barley and Atorvastatin. The high dose only of barley bran caused a slight increase in bl.urea level after 8 weeks of treatment. There is no any effect of different treatments on serum creatinine level during 8 weeks of treatment.

1. Induction of hyperlipaemia; Induction of hyperlipaemia was performed using cholesterol: cholic acid mixture at a ratio 3: 1 [(26)]. In addition, saturated fats (10%) and sucrose (50%) were added to the diet. Cholic acid was used to overcome the difficulty of cholesterol absorption.

As can be seen from the data shown in Table (5), very highly significant elevations were indicated in the level of serum total lipids, total cholesterol, risk ratio, triglycerides, ALT, AST, ALP, LDH and Alb/Glob ratio. While slight elevations were indicated in the level of serum creatinine and albumin after 10 weeks from the induction of hyperlipaemia. Also, highly significant reductions were indicated in the level of serum HDL-C. The level of serum total proteins and blood urea did not affect. The slightly elevation in serum creatinine after induction of hyperlipaemia is statistically not significant and could lies in the normal range. Elevations indicated in serum total lipids seem to be logic and runs parallel with the excess of saturated fat and sugar available in the diet. Elevations in serum total lipids were also indicated after the induction of experimental hyperlipaemia [(27)] .The increase indicated in the level of cholesterol runs parallel with the similar elevations indicated by the previous authors. Reductions indicated in HDL-C may be an important because it is stimulate the removal of cholesterol from the peripherol cells back to the liver for excretion. The increase in the level of triglycerides could be referred to the presence of excess saturated fats in the dietary intake. This excess of the need of the body leads to their conversion into triglycerides in the liver. These triglycerides are packaged into VLDL and released into the circulations for delivery to various tissues for storage or production of energy through oxidation [(28)].

Elevations indicated in serum ALT, AST, ALP and LDH after the induction of hyperlipaemia may be due to the destruction of some liver parenchymal cells or by an enhancement of the activity of the enzyme itself to face the damaging effect of free radicals accompanied with hyperlipaemia [(29)].

In most human studies as well as experimental animals there is a positive correlation between cardiovascular disease and blood cholesterol level. Free radicals play an important role in this concept. Lipid oxidation and generation of free radicals are considered to be natural phenomenon in biological system. The formation of reactive free radicals is mediated by a number of agents and mechanisms such as xenobiotic metabolism. The free radicals formed are highly reactive with molecular oxygen forming peroxy radicals and hydroperoxides and thus initiating a chain reaction. Pro-oxidant states cause cellular lesions in all major organs by damaging cellular components and cell function. The free radical has been implicated in the etiology of several genetic as well as acquired metabolic disorders. One of these diseases is hyperlipidaemia which favors the formation of free radicals, leading to arterial damage and platlet aggregation.Cholesterol oxidation products have received a lot of attention because of their involvement in the development of coronary artery disease [(30)].

Oxygen free radicals and lipid peroxidation

are major factors in the etiology of atherogenesis and its associated clinical disorders, which include coronary artery disease, stroke, ischemic dementia and various other atherosclerotic disorders [31]. Atherosclerosis is a vascular disease with a complex etiology [32]. The oxidative modification hypothesis of atherosclerosis proposes that oxidation of LDL leading to the accumulation of lipid peroxides and other oxidized radicals, is a major cause of atherosclerosis. It is now known that the level of serum LDL is positively correlated with the incidence of hyperlipidaemea and then atherosclerosis. One widely accepted theory for explaining this phenomenon is that oxidation of LDL. However, epidemiological studies have shown that the concentration of serum HDL was inversely correlated to the risk of atherosclerosis. Experimental evidences have suggested that HDL can protect LDL against oxidation. However, the HDL – cholesterol may be increased by N – acetyl cysteine suggesting the possibility that a decrease in HDL – cholesterol may be related to changes of the thiol level and / or the thiol / disulfide redox status (REDST) in the plasma. They concluded that there is a strong possibility that the changes in plasma thiol level / plasma and intracellular thiol disulphide redox status of peripheral blood mononuclear cells may play a causative role in the pathophysiology of the arteriosclerotic process and the development of coronary heart disease. This conclusion is in line with the fact that abnormally high "total homocysteine levels" which are also typically associated with an oxidative shift in REDST have been identified as an independent risk factor for CHD. The oxidative shift in REDST may therefore be a consensus risk factor common to several or all independent other risk factors [33].

2. Evaluation of barley and its bran; barley bran more lowered serum total cholesterol and serum triglycerides in the rats than whole barley (Table 6). Elevated serum HDL- Cholesterol levels, unlike total cholesterol levels, is reported [34] to provide protection against heart disease. In barn fed animals, however, an interesting pattern again emerged. The differing results may be due to diet, to the amount of soluble fiber and β-glucan in the diet. For example, rats fed the diet containing the highest level of soluble fiber and β-glucan (diet formulated with barley bran) showed the lowest serum cholesterol levels throughout the eight- weeks test period (Table 6). The physiological effects of dietary fiber have proven to be more complex than once thought. Currently, the major nutritive effect of fiber receivins the most focus is its hypolipidaemic effect, which is more pronounced by soluble fiber such as β-glucan than non soluble fiber. The most widely held hypothesis of the mechanism by which fiber influences lipid metabolism is that it interrupts enterohepatic circulation by binding the circulating bile acids and preventing their subsequent reabsorption [35, 36] .Thus, an increased proportion of cholesterol produced by the liver is converted to bile acids, thereby making less cholesterol available for packaging into lipoproteins.

Barley reportedly contains other factors that affect blood plasma cholesterol [37, 38 and 39]. It was reported that [40] barley contains compounds that inhibit 3-hydroxy-3-methylglutaryl coenzyme A (HMG-COA) reductase in chickens, the rate-limiting enzyme in the cholesterol biosynthetic pathway. One of these compounds was isolated from high-protein barley flour (HPBF) and identified as α-tocotienol. The present authors hypothesized that the presence of dietary HPBF would partially alleviate the hypercholesterolaemia resulting from dietary cholesterol because reports indicate that various fibers moderate the increase in plasma and liver cholesterol diets [41, 42]. Furthermore, Gallaher et al. found a significant serum lipid- lowering effect of a β-glucan rich barley fraction in hypercholesterolaemic men. These findings also indicate that the β-glucan in barley influences sterol metabolism [43].

Barley also contains lipids and phytosterols, which have both been postulated to reduce serum cholesterol [44]. Recently, barley oil was found to have a lipid- lowering effect similar to that of barley bran flour when added to a low-fat diet [44]. The authors suggested that the lipids of certain barley fractions, such as brewer's spent grain, may have lipid-lowering properties. β-glucans in barley increased intestinal viscosity and decreased plasma cholesterol of male broiler chicks fed barley and cholesterol [45]. In the chicken model [37] reported a cholesterol-lowering effect in barley due to a decrease in a rate- limiting enzyme in cholesterol synthesis; subsequently, the authors identified α-tocotrienol as an inhibitor of this enzyme .However, if human subjects respond like the rats used in this study, it would suggest that the ability of soluble fiber (SF) in barley meal to lower cholesterol is negated by some mechanism (activation of β-glucanses, for example).

Elevated serum triglycerides (TG) levels are viewed by some as an independent risk factor in heart disease [34]. Barley diets appeared to be strong predictors

caused a reduction in of the cholesterol – lowering in serum and the serum triglycerides content in liver of rats .The viscous property of soluble β-glucan may result in reduced absorption, or reabsorption of lipids [46].

β- glucan decreased LDL- Cholesterol and increased HDL-Cholesterol. High density lipoprotein may hasten the removal of cholesterol from peripheral tissue to the liver for catabolism and excretion. Also, high levels of HDL may complete with LDL receptor sites on arterial smooth muscle cells and thus partially inhibit uptake and degradation of LDL. The increase of HDL concentration could protect LDL against oxidation in – vivo because the lipids in HDL are preferentially oxidized before those in LDL [47].

A considerable attention has been devoted to the role of the different natural antioxidants as inhibitors of LDL oxidation and their possible therapeutic effects to prevent hyperlipaemia and atherosclerosis. It was reported that total and LDL-Cholesterol were reduced [48], both decreases being significantly correlated with soluble β-glucan content. It has been hypothesized that soluble β-glucan tends to increase intestinal viscosity due to its low molecular weight and tendency to form viscous solutions, resulting in reduced bile acid and cholesterol production ,and increased faucal fat bile acid excretion thus reducing plasma cholesterol [48,49].

These results suggested that barley and its bran may evoke different lipidaemic responses. Thus, it seems that the bran fraction of barley that is rich in soluble fiber and β- glucan would likely exert a more favorable effect on blood lipids than any fractions from whole barley.

Table (2): Prophylactic effect of whole barley and barley bran on serum lipids (mg/dl ± S.D) and % variation from the corresponding control during the induction of hyperlipaemia for 8 weeks in male rats.

GROUP NUMBER	TREATMENT	ITEM	Time intervals (Wk's)				
			0	4	% var.	8	% var.
1	Negative control	T.L	293 ± 55.4	299± 35.3†	2.05 ↑	301 ± 60.9†	2.73 ↑
		Trig.	50.2±5.47	51.3±1.70 †	2.19 ↑	49.9±7.00 †	0.60↓
		T.Ch.	53.5 ±7.78	56.7±6.10 †	5.98 ↑	55.7±4.23†	4.11 ↑
		HDL.Ch.	36.6±2.42	36.5±4.30 †	0.27↓	35.3±1.62†	3.55 ↓
		T./HDL	1.46±0.12	1.55±0.27 †	6.16 ↑	1.58±0.09†	8.22 ↑
2	Positive control	T.L	268 ± 53.6	723 ± 41.4***	169.8 ↑	751 ± 53.0***	180.2 ↑
		Trig.	54.4±7.81	65.0±1.39*	19.5 ↑	132.6±6.15***	144 ↑
		T.Ch.	55.4 ± 6.71	128.1±5.24 ***	131.2 ↑	179.9±14.4***	225 ↑
		HDL.Ch.	34.0±2.93	26.2±1.81***	22.9 ↓	19.5±2.58***	42.6 ↓
		T./HDL	1.63±0.08	4.89±0.44 ***	200.0 ↑	9.23±1.20***	466 ↑
3	Barley Bran (100 mg/kg b.w)	T.L	273 ± 64.2	419 ± 79.2***	53.5 ↑	543 ± 76.7***	98.9 ↑
		Trig.	50.0±6.28	39.1±5.95 *	21.8 ↓	47.2±5.49†	5.60↓
		T.Ch.	55.6±5.18	113.0±4.08***	103.2↑	75.7±3.99***	36.2 ↑
		HDL.Ch.	37.7±2.56	27.5±2.18***	27.1 ↓	30.4±2.64**	19.4 ↓
		T./HDL	1.47±0.06	4.10±0.49***	178.9 ↑	2.49±0.23***	91.8 ↑
4	Barley Bran (200 mg/kg b.w)	T.L	251 ± 27.6	612 ± 63.6***	143.8 ↑	379 ± 78.4***	51.0 ↑
		Trig.	57.0±9.41	42.1±9.70 *	26.1 ↓	40.2±9.40*	29.5 ↓
		T.Ch.	53.0±6.06	109.9±2.95***	107.4↑	64.0±5.95**	20.8 ↑
		HDL.Ch.	35.9±2.71	25.5±2.73***	28.9 ↓	31.8±2.93*	11.4 ↓
		T./HDL	1.48±0.07	4.31±0.58***	191.2 ↑	2.01±0.29**	35.8 ↑
5	Whole Barley (200 mg/kg b.w)	T.L	281 ± 30.5	500 ± 99.7**	77.9 ↑	530 ± 62.1***	88.6 ↑
		Trig.	48.8±6.44	40.6±8.72†	16.8 ↓	43.7±6.70 †	10.5 ↓
		T.Ch.	55.4±7.67	111.5±3.84***	101.3↑	76.3±6.38***	37.7 ↑
		HDL.Ch.	36.3±1.92	27.2±1.65***	25.1 ↓	29.9±3.44**	17.6 ↓
		T./HDL	1.53±0.13	4.10±0.35***	167.9 ↑	2.55 ± 0.47**	66.7 ↑

* P < 0.05. ** P < 0.01. *** P < 0.001. T.L.: total lipids. T.C.: total cholesterol.

TG.: triglycerides. HDL-C: HDL-cholesterol. T. /HDL-C: total cholesterol / HDL-cholesterol. EGb: Ginkgo biloba extract.

Table (3): Prophylactic effect of whole barley and barley bran on serum enzymes (U/L ± S.D) and % variation from the corresponding control during the induction of hyperlipaemia for 8 weeks in male rats.

GROUP NUMBER	TREATMENT	ITEM	Time intervals (Wk's)				
			0	4	% var.	8	% var.
1	Negative control	ALT	32.3±5.74	32.4±3.57 †	0.31 ↑	32.1±5.50 †	0.62 ↓
		AST	65.3±2.77	64.9±6.06 †	0.61 ↓	66.2±13.3†	1.38 ↑
		ALP	30.3±3.22	30.9±1.98 †	1.98 ↑	32.8±3.89 †	8.25↑
		LDH	694±89.9	683±66.2 †	1.59 ↓	698±86.9 †	0.58 ↑
2	Positive control	ALT	33.7±1.53	61.7±3.48 ***	83.1 ↑	56.7±8.33**	68.2 ↑
		AST	63.3±9.80	109.8±11.4 ***	73.5 ↑	119.7±17.9***	89.1 ↑
		ALP	29.7±2.21	26.5±3.79 †	10.8 ↓	74.4±6.39 ***	150 ↑
		LDH	741±80.1	694±64.4 †	6.34 ↓	989±47.8***	33.5 ↑
3	Barley Bran (100 mg/kg b.w)	ALT	31.4±4.20	36.3±1.69 *	15.6 ↑	36.2±5.22*	15.3
		AST	60.2±5.04	98.9±10.8 **	64.3 ↑	75.7±10.0**	25.7 ↑
		ALP	30.5±4.77	25.2±3.41*	17.4 ↓	34.3±6.20 †	12.5 ↑
		LDH	716±95.8	729±57.8 †	1.82 ↑	303±19.9***	57.7 ↓
4	Barley Bran (200 mg/kg b.w)	ALT	30.6±4.92	34.1±4.67 †	11.4 ↑	33.8±6.99 †	10.5 ↑
		AST	67.6±4.16	115.8±8.94 ***	71.3 ↑	68.1±12.8 †	0.74 ↑
		ALP	31.5±5.55	41.6±5.98**	32.1 ↑	23.5±4.41 **	25.4 ↓
		LDH	771±64.9	560±74.9**	27.4 ↓	472±27.1***	38.3 ↓
5	Whole Barley (200 mg/kg b.w)	ALT	34.8±3.07	36.5±1.37 †	4.88 ↑	41.9±6.94 **	20.4 ↑
		AST	60.4±6.92	90.7±2.29***	50.2 ↑	92.8±5.53 ***	53.6 ↑
		ALP	30.1±3.52	41.5±4.58**	37.9 ↑	43.3±9.82 **	43.8 ↑
		LDH	739±91.4	628±90.2 †	15.0 ↓	384±58.0***	48.0 ↓

* P < 0.05. ** P < 0.01. *** P < 0.001. ALT: Alanine aminotransferase. AST: Aspartat aminotransferase. ALP: Alkaline phosphatase. LDH:Lactate dehydrogenase. EGb: Ginkgo biloba extract.

Table (4): Prophylactic effect of whole barley and barley bran on serum proteins, urea and creatinine (mg/dl ± S.D) and % variation from the corresponding control during the induction of hyperlipaemia for 8 weeks in male rats.

GROUP NUMBER	TREATMENT	ITEM	Time intervals (Wk's)				
			0	4	% var.	8	% var.
1	Negative control	T.P.	6.03±0.62	6.44±0.19 †	6.80 ↑	6.42±0.23 †	6.47 ↑
		Alb.	3.54±0.20	3.77±0.24 †	6.50 ↑	3.72±0.15 †	5.08 ↑
		Glob.	2.49±0.40	2.67±0.15 †	7.22 ↑	2.70±0.14 †	8.43 ↑
		Alb/Glob	1.42±0.26	1.41±0.15 †	0.70 ↓	1.38±0.13 †	2.82 ↓
		Urea	25.2± 5.09	24.6 ± 1.50†	2.38 ↓	24.9 ± 3.14†	1.19 ↓
		Creat.	0.39 ± 0.09	0.40 ± 0.02†	2.56 ↑	0.41 ± 0.02†	5.13 ↑
2	Positive control	T.P.	6.28±0.61	7.07±0.38*	12.6 ↑	6.40±0.29 †	1.91 ↑
		Alb.	3.92±0.28	4.33±0.38*	10.5 ↑	4.37±0.45 †	11.5 ↑
		Glob.	2.36±0.42	2.74±0.34 †	16.1 ↑	2.03±0.49 †	13.9 ↓
		Alb/Glob	1.66±0.30	1.58±0.30 †	4.82 ↓	2.15±0.43*	29.5 ↑
		Urea	23.9 ± 1.59	31.1 ± 2.79***	30.1 ↑	27.3 ± 2.46*	14.2 ↑
		Creat.	0.55 ± 0.09	0.83 ± 0.02**	50.9 ↑	0.59 ± 0.06†	7.27 ↑
3	Barley Bran	T.P.	6.08±0.14	6.76±0.39**	11.2 ↑	6.40±0.27*	5.26 ↑

	(100 mg/kg b.w)	Alb.	3.82±0.23	3.84±0.24 †	0.52 ↑	3.90±0.34 †	2.09 ↑
		Glob.	2.26±0.18	2.92±0.24 ***	29.2 ↑	2.50±0.35 †	10.6 ↑
		Alb/Glob	1.69±0.21	1.31±0.12**	22.5 ↓	1.56±0.17 †	7.69 ↓
		Urea	25.6 ± 4.24	28.6 ± 3.99†	11.7 ↑	27.0 ± 3.72†	5.47 ↑
		Creat.	0.49 ± 0.03	0.61 ± 0.03**	24.5 ↑	0.59 ± 0.04*	20.4 ↑
4	Barley Bran (200 mg/kg b.w)	T.P.	6.54±0.20	6.84±0.39 †	4.59 ↑	6.67±0.43†	1.99 ↑
		Alb.	4.15±0.22	3.85±0.26*	7.23↓	4.22±0.16 †	1.69 ↑
		Glob.	2.39±0.24	2.99±0.35 **	25.1 ↑	2.45±0.32 †	2.51 ↑
		Alb/Glob	1.74±0.25	1.29±0.20**	25.9 ↓	1.72±0.18 †	1.15 ↓
		Urea	26.6 ± 3.19	45.3 ± 8.00**	70.3 ↑	41.7 ± 8.60**	56.8 ↑
		Creat.	0.50 ± 0.05	0.83 ± 0.02***	66.0 ↑	0.61 ± 0.07*	22.0 ↑
5	Whole Barley (200 mg/kg b.w)	T.P.	6.17±0.26	6.97±0.65*	12.9 ↑	6.43±0.47 †	4.21 ↑
		Alb.	3.81±0.28	4.00±0.49 †	4.99 ↑	3.80±0.34 †	0.26 ↓
		Glob.	2.36±0.28	2.97±0.49 *	25.8 ↑	2.63±0.35 †	11.4 ↑
		Alb/Glob	1.61±0.36	1.35±0.30†	16.2 ↓	1.44±0.27 †	10.6 ↓
		Urea	26.3 ± 1.65	34.9 ± 2.65***	32.7 ↑	34.2 ± 4.30**	30.0 ↑
		Creat.	0.51 ± 0.03	0.83 ± 0.02***	62.7 ↑	0.60 ± 0.02*	17.6 ↑

†P > 0.1. * P < 0.05. ** P < 0.01. *** P < 0.001. T.P.: Total protein. Alb.: Albumin. Glob: Globulin. Alb/ Glob: Albumin/ Globulin. Creat: Creatinine. EGb: Ginkgo biloba extract

Table (5): Arithmetic mean values ± S.D and % changes from the corresponding control of different biochemical parameters before and after induction of hyperlipaemia in male albino rats

Parameters	Normal level	Hyperlipaemia Level	% change	Parameters	Normal level	Hyperlipaemia Level	% change
S.T. lipids (mg /dl)	266 ± 39.7	739 ± 46.4***	178 ↑	S.T.Proteins (g/dl)	6.24 ± 0.32	6.17 ± 0.271†	1.12 ↓
S.Triglyc.(mg /dl)	51.5± 7.26	130 ± 8.51***	152 ↑	S.Alb.(g/dl)	3.78± 0.21	4.37 ± 0.39*	15.6 ↑
S.T.chol.(mg /dl)	55.1± 6.16	181 ± 12.9***	228 ↑	S.Glob.(g/dl)	2.46 ± 0.15	1.80 ± 0.22**	26.8 ↓
S.HDL-chol.(mg /dl)	35.7 ± 2.45	19.3 ± 0.42***	45.9 ↓	S.Alb. / Glob	1.54± 0.07	2.43 ± 0.26***	57.8 ↑
R.R (T/HDL-chol.)	1.54 ± 0.11	9.38 ± 0.41***	509.1 ↑	Bl. Urea (mg/dl)	25.1± 3.21	26.6 ± 4.35†	5.98 ↑
S.ALT.(U/ml)	32.2 ± 3.70	58.1 ± 5.25***	80.4 ↑	S. Creat. (mg/dl)	0.46 ± 0.08	0.52 ± 0.04*	13.0 ↑
S.AST.(U/ml)	62.7 ± 6.29	117.6 ± 12.8***	87.6 ↑				
S.ALP. (IU/L)	30.6 ± 3.85	66.1 ± 4.61***	116 ↑				
S. LDH.(U/l)	747 ± 75.6	986 ± 58.6***	32.0 ↑				

†P > 0.1. * P < 0.05. ** P < 0.01. *** P < 0.001. T.L.: total lipids. T.C.: total cholesterol. TG: triglycerides. HDL-C: HDL-cholesterol. T. /HDL-C: total cholesterol / HDL-cholesterol. ALT: Alanine aminotransferase. AST: Aspartat aminotransferase. ALP: Alkaline phosphatase. LDH:Lactate dehydrogenase. EGb: Ginkgo biloba extract.

Table (6): The Curative effect of whole barley and barley bran on serum lipids (mg/dl ± S.D) and % variation from the corresponding control during the induction of hyperlipaemia for 8 weeks in male rats.

GROUP NUMBER	TREATMENT	ITEM	Before induction of hyperlipaemia	After induction of hyperlipaemia	% var.	Time intervals (Wk's) 4	% var.	8	% var.
1	Negative control	T.L	293±55.4	301±45.5†	2.73↑	299±28.5†	0.66↓	298±46.2†	1.00↓
		Trig.	50.2±5.47	49.9±7.00†	0.60↓	48.9±4.38†	2.00↓	53.0±6.40†	6.21↑
		T.Ch.	53.5±7.78	55.7±4.23†	4.11↑	53.7±8.54†	3.59↓	56.9±6.13†	2.15↑
		HDL.Ch.	36.6±2.42	35.3±1.62†	3.55↓	36.6±4.60†	3.68↑	35.7±4.15†	1.13↑
		T./HDL	1.46±0.12	1.58±0.09†	8.22↑	1.47±0.12†	6.96↓	1.59±0.30†	0.63↑
2	Positive control	T.L	268±53.6	751±53.0***	180↑	854±44.4**	13.7↑	861±34.8**	14.6↑
		Trig.	54.4±7.81	132.6±6.15***	144↑	143.6±8.16*	8.30↑	138.7±12.1†	4.60↑
		T.Ch.	55.4±6.71	179.9±14.4***	225↑	188.5±9.50†	4.78↑	168.4±4.28†	6.39↓
		HDL.Ch.	34.0±2.93	19.5±2.58***	42.6↓	19.1±1.22†	2.05↓	20.1±0.41†	3.08↑
		T./HDL	1.63±0.08	9.23±1.20***	466↑	9.87±1.05†	6.93↑	8.38±0.19†	9.21↓
3	Barley Bran (100 mg/kgb.w)	T.L	268±33.1	740±54.4***	176↑	516±52.6***	30.3↓	446±91.2***	39.7↓
		Trig.	49.7±4.99	131.3±7.23***	164↑	123.0±18.0†	6.32↓	99.7±10.8**	24.1↓
		T.Ch.	55.8±5.74	190.0±12.2***	241↑	148.6±17.8**	21.8↓	100.4±9.48***	47.2↓
		HDL.Ch.	35.8±4.07	19.2±3.29***	46.4↓	28.0±1.18***	45.8↑	31.5±2.21***	64.1↑
		T./HDL	1.56±0.06	9.90±0.97***	535↑	5.31±0.75***	46.4↓	3.19±0.20***	67.8↓
4	Barley Bran (200 mg/kgb.w)	T.L	288±46.1	755±47.3***	162↑	507±49.2***	32.8↓	370±67.4***	51.0↓
		Trig.	54.6±9.64	133.0±9.08***	144↑	117.5±21.3†	11.7↓	82.1±9.02***	38.3↓
		T.Ch.	53.0±3.13	178.9±18.7***	238↑	145.6±19.9**	18.6↓	84.3±8.26***	52.9↓
		HDL.Ch.	35.9±2.09	19.6±3.69***	45.4↓	28.7±2.28**	46.4↑	33.1±2.39***	68.9↑
		T./HDL	1.48±0.04	9.13±1.06***	517↑	5.07±1.03***	44.5↓	2.55±0.35***	72.1↓
5	Whole Barley (200 mg/kgb.w)	T.L	261±53.8	749±77.7***	187↑	562±77.5**	25.0↓	401±42.2***	46.5↓
		Trig.	55.1±6.34	128.5±6.09***	133↑	123.9±13.3†	3.58↓	88.9±5.68***	30.8↓
		T.Ch.	53.2±6.47	175.4±10.7***	230↑	147.4±16.1**	16.0↓	93.4±6.43***	46.8↓
		HDL.Ch.	35.2±3.90	19.4±3.31***	50.6↓	27.6±1.80***	42.3↑	31.7±2.32***	63.4↑
		T./HDL	1.51±0.10	9.04±1.20***	499↑	5.34±0.49***	40.9↓	2.95±0.15***	67.4↓
6	Atorvastatin (0.9mg/kgb.w)	T.L	251±35.9	746±87.7***	197↑	618±64.3*	17.2↓	443±85.2**	40.6↓
		Trig.	54.0±8.62	132.8±9.42***	146↑	118.3±7.50*	10.9↓	74.7±10.1***	43.7↓
		T.Ch.	55.2±5.77	179.6±12.0***	225↑	132.0±11.9***	26.5↓	73.9±9.70***	52.9↓
		HDL.Ch.	34.4±3.16	19.4±2.06***	43.6↓	31.2±2.05***	60.8↑	34.4±2.74***	77.3↑
		T./HDL	1.60±0.05	9.30±1.19***	481↑	4.23±0.60***	54.5↓	2.15±0.24***	76.9↓

†P > 0.1. * P < 0.05. ** P < 0.01. *** P < 0.001. T.L.: total lipids. T.C.: total cholesterol.
TG: triglycerides. HDL-C: HDL-cholesterol. T. /HDL-C: total cholesterol / HDL-cholesterol. EGb: Ginkgo biloba extract.

Table (7): The Curative effect of whole barley and barley bran on serum enzymes (U/L ± S.D) and % variation from the corresponding control during the induction of hyperlipaemia for 8 weeks in male rats.

GROUP NUMBER	TREATMENT	ITEM	Before induction of hyperlipaemia	After induction of hyperlipaemia	% var.	Time intervals (Wk's) 4	% var.	8	% var.
1	Negative control	ALT	32.3±5.74	32.1±5.50†	0.62↓	31.6±2.09†	1.56↓	31.4±4.80†	2.18↓
		AST	65.3±2.77	66.2±13.3†	1.38↑	60.3±3.87†	8.91↓	60.3±5.88†	8.91↓
		ALP	30.3 ± 3.22	32.8 ± 3.30†	8.25 ↑	34.0 ± 1.76†	3.36 ↑	34.1 ±4.44†	3.96↑
		LDH	694 ± 89.9	698 ± 86.9†	0.58 ↑	674 ± 16.6†	3.44 ↓	670 ± 57.5†	4.01↓
2	Positive control	ALT	33.7±1.53	56.7±8.33**	68.2↑	56.9±5.88†	0.35↑	56.6±5.48†	0.18↓
		AST	63.3±9.80	119.7±17.9***	89.1↑	104.3±12.5†	12.9↓	112.7±7.73†	5.85↓
		ALP	29.7 ± 2.21	74.4 ± 6.39***	150 ↑	73.7 ± 1.88†	0.94 ↓	73.2 ±2.13†	1.61↓
		LDH	741 ± 80.1	989 ± 47.8***	33.5 ↑	986 ± 49.1†	0.30 ↓	953 ±31.0†	3.64↓
3	Barley Bran (100 mg/kg b.w)	ALT	36.0±4.90	58.7±7.50***	63.1↑	48.9±3.21**	16.7↓	43.9±3.55**	25.2↓
		AST	61.2±2.12	120.0±12.5***	96.1↑	97.0±11.4**	19.2↓	63.9±5.25***	46.7↓
		ALP	32.8 ± 4.36	65.6 ± 5.21***	100 ↑	62.7 ± 5.80†	4.42 ↓	52.0 ±5.33***	20.7↓
		LDH	715 ± 77.4	996 ± 59.1***	39.3 ↑	761 ± 61.0***	23.6 ↓	690 ±61.9***	30.7↓
4	Barley Bran (200 mg/kg b.w)	ALT	31.6±2.01	58.5±6.15***	85.1↑	45.5±6.36*	22.2↓	37.0±4.41***	36.7↓
		AST	61.7±3.84	120.1±10.4***	94.7↑	86.8±10.5***	27.7↓	63.6±6.34***	47.0↓
		ALP	23.9 ± 3.09	63.7 ± 4.10***	120 ↑	54.6 ± 7.50*	14.3 ↓	43.6 ±4.81***	31.6↓
		LDH	721 ± 78.2	979 ± 53.5***	35.8 ↑	731 ± 70.2***	25.3 ↓	692 ±79.0***	29.3↓
5	Whole Barley (200 mg/kg b.w)	ALT	31.1±4.28	63.7±1.89***	105↑	48.3±3.38***	24.2↓	42.6±3.84***	33.1↓
		AST	60.1±6.51	118.0±4.08***	96.3↑	98.1±6.18***	16.9↓	62.7±4.92***	46.9↓
		ALP	33.0 ± 3.57	65.9 ± 6.24***	100 ↑	60.1± 1.20*	8.80 ↓	46.5 ±2.51***	29.4↓
		LDH	740 ± 85.8	995 ± 64.3***	34.5 ↑	710 ± 96.9***	28.6 ↓	679 ±50.2***	31.8↓
6	Atorvastatin (0.9mg/kg b.w)	ALT	31.3±3.79	57.0±5.33***	82.1↑	41.9±4.87***	26.5↓	38.6±4.53***	32.3↓
		AST	60.9±4.44	115.1±5.04***	29.0↑	90.2±5.93***	21.6↓	56.9±2.53***	50.6↓
		ALP	31.0 ±5.40	63.9 ± 8.87***	106 ↑	51.2±5.35**	19.9 ↓	35.3 ±5.40***	44.8↓
		LDH	703 ± 88.2	992 ± 94.9**	41.1 ↑	731 ± 81.0**	26.3 ↓	674 ± 61.8**	32.1↓

†P > 0.1. * P < 0.05. **P < 0.01. *** P < 0.001. ALT: Alanine aminotransferase. AST: Aspartat aminotransferase. ALP: Alkaline phosphatase. LDH:Lactate dehydrogenase. EGb: Ginkgo biloba extract.

Table (8): The Curative effect of whole barley and barley bran on serum proteins, urea and creatinine (mg/dl ± S.D) and % variation from the corresponding control during the induction of hyperlipaemia for 8 weeks in male rats.

GROUP NUMBER	TREATMENT	ITEM	Before induction of hyperlipaemia	After	% var.	Time intervals (Wk's) 4	% var.	8	% var.
1	Negative control	T.P.	6.03 ± 0.62	6.42 ± 0.23†	6.47 ↑	6.48± 0.51†	0.93↑	6.73± 0.36†	4.83↑
		Alb.	3.54± 0.20	3.72 ± 0.15†	5.08↑	3.91± 0.42†	5.11↑	3.60± 0.18†	3.32
		Glob.	2.49± 0.40	2.70± 0.14†	8.43↑	2.57± 0.42†	4.81↓	3.13± 0.16†	15.9↑
		Alb/Glob	1.42± 0.26	1.38± 0.13†	2.82↓	1.52± 0.21†	10.1↑	1.15± 0.12**	16.7↓
		Urea	25.2±5.09	24.9±3.14†	1.19↓	24.6±2.88†	1.20↓	24.3±3.28†	2.41↓
		Creat.	0.39±0.09	0.41±0.02†	5.13↑	0.44±0.02†	7.31↑	0.43±0.10†	4.88↑
2	Positive control	T.P.	6.28± 0.61	6.40 ± 0.29†	1.91↑	6.73 ± 0.53†	5.16↑	4.90 ±0.62**	7.81↑
		Alb.	3.92± 0.28	4.37 ± 0.45†	11.5↑	4.05 ± 0.37†	7.32↓	3.03 ±0.33**	30.7↓
		Glob.	2.36± 0.42	2.03 ± 0.49†	13.9↓	2.68 ± 0.13*	32.0↑	1.87 ±0.25†	7.88↓
		Alb/Glob	1.66± 0.30	2.15 ± 0.43*	29.5↑	1.51 ± 0.17*	29.8↓	1.62 ±0.19†	24.6↓
		Urea	23.9±1.59	27.3±2.46*	14.2↑	32.0±3.46*	17.2↑	27.0±2.57†	1.10↓
		Creat.	0.53±0.09	0.59±0.06†	7.27↑	0.42±0.06**	28.8↓	0.49±0.10†	16.9↓
3	Barley Bran (100 mg/kg b.w)	T.P.	6.40 ± 0.51	6.30 ± 0.26†	1.56↓	6.53 ± 0.52†	3.65↑	5.37 ±0.89*	14.8↓
		Alb.	3.91 ± 0.21	4.36 ± 0.33*	11.5↑	3.86 ± 0.27*	11.5↓	3.01 ±0.39***	31.0↓
		Glob.	2.49 ± 0.39	1.94 ± 0.34*	22.1↓	2.67 ± 0.44**	37.6↑	2.36 ±0.39*	21.6↑
		Alb/Glob	1.57 ± 0.18	2.25 ± 0.25**	43.3↑	1.45 ± 0.12***	35.6↓	1.28 ±0.32***	43.1↓
		Urea	23.5±3.53	26.2±4.49†	11.5↑	25.9±5.20†	1.15↓	26.7±2.11†	1.91↑
		Creat.	0.52±0.14	0.47±0.04†	9.61↓	0.45±0.03†	4.26↓	0.49±0.06†	4.26↑
4	Barley Bran (200 mg/kg b.w)	T.P.	6.17± 0.62	6.20 ± 0.30†	0.49↑	6.44 ± 0.33†	3.87↑	5.05 ±0.78**	18.5↓
		Alb.	3.83± 0.42	4.34 ± 0.42†	13.3↑	3.96± 0.30*	8.76↓	3.10 ±0.17***	28.6↓
		Glob.	2.34± 0.11	1.86 ± 0.23**	20.5↓	2.48± 0.19***	33.3↑	1.95 ±0.27†	1.91↑
		Alb/Glob	1.63± 0.23	2.33 ± 0.44**	42.9↑	1.60± 0.29**	31.3↓	1.59 ±0.37**	31.8↓
		Urea	23.9±1.21	26.0±3.94†	8.79↑	21.8±2.53*	16.2↓	29.3±1.93*	12.7↑
		Creat.	0.48±0.11	0.53±0.08†	10.4↑	0.44±0.03*	17.0↓	0.46±0.06†	13.2↓
5	Whole Barley (200 mg/kg b.w)	T.P.	6.01 ± 0.29	6.20 ± 0.43†	3.16 ↑	6.40 ± 0.75†	3.23↑	4.70±0.74**	24.2↓
		Alb.	3.73 ± 0.30	4.33 ± 0.38**	16.1 ↑	3.84 ± 0.32*	11.3↓	3.01±0.31***	30.5↓
		Glob.	2.28 ± 0.36	1.87 ± 0.27*	18.0↓	2.56 ± 0.30**	36.9↑	1.69±0.40†	9.63↓
		Alb/Glob	1.63± 0.33	2.32 ± 0.41**	42.3↑	1.50 ± 0.24**	35.3↓	1.78±0.16**	23.3↓
		Urea	23.6±2.68	25.6±5.11†	8.47↑	24.3±4.39†	5.08↓	23.3±1.96†	8.98↓
		Creat.	0.51±0.05	0.50±0.08†	1.96↓	0.45±0.02†	10.0↓	0.47±0.06†	6.00↓
6	Atorvastatin (0.9mg/kg b.w)	T.P.	6.36 ± 0.35	6.24 ± 0.13†	1.39 ↓	6.13± 1.00†	1.76↓	4.68 ± 0.42***	25.0↓

		Alb.	3.90 ± 0.13	4.35 ± 0.45*	11.5 ↑	3.86± 0.57†	11.3↓	2.97 ± 0.08***	31.7↓
		Glob.	2.46 ± 0.25	1.89 ± 0.18**	23.2↓	2.27± 0.54†	20.1↑	1.71± 0.18†	9.52↓
		Alb/Glob	1.58 ± 0.23	2.30 ± 0.38**	45.6↑	1.70± 0.14**	26.1↓	1.74± 0.11**	24.3↓
		Urea	24.6±3.20	29.5±2.35*	19.9↑	33.0±4.69†	11.9↑	28.8±3.15†	2.37↓
		Creat.	0.54±0.11	0.50±0.06†	7.41↓	0.46±0.02†	8.00↓	0.47±0.03†	6.00↓

†P > 0.1. * P < 0.05. ** P < 0.01. *** P < 0.001. T.P.: Total protein. Alb.: Albumin. Glob: Globulin. Alb/ Glob: Albumin/ Globulin. Creat: Creatinine. EGb: Ginkgo biloba extract.

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