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Beneficial Effects of Dietary Intake of Soybean and Barley on Hyperlipidemic Albino Rats

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Abstract This research explores the impact of incorporating soybean and barley into the diet of hyperlipidemic albino rats. The study assessed how these dietary interventions affected various parameters, including serum lipids, atherogenic indexes, oxidative stress markers, and antioxidant enzyme activity. We divided fifty male Sprague-Dawley rats into a negative control group and hyperlipidemic groups. The hyperlipidemic groups were further categorized into positive control groups and treated groups, which received dietary interventions involving soybean, barley, or a combination of both. After 60 days, blood samples were collected when the rats were sacrificed. Nutritional analysis revealed no significant differences in food intake and feed efficiency ratio between the negative control group and the treated groups. However, these values were markedly lower ($p \leq 0.05$) compared to the positive control group. Among the hyperlipidemic diet groups, the group supplemented with a 20% blend of soybean and barley showed the most notable improvements in serum lipid levels and atherogenic indexes. Both the barley and blend groups also showed significant reductions in serum oxidant markers and notable increases in the activity of serum antioxidant enzymes. These findings suggest that a combination of soybean, barley, and their blend may effectively mitigate complications associated with hyperlipidemia.

Keywords Hyperlipidemia, Soybean, Barley, Serum Lipids, Antioxidant Enzymes

1. Introduction

Hyperlipidemia, characterized by elevated levels of plasma lipids including cholesterol, triglycerides, and lipoproteins, is a medical condition with significant implications for cardiovascular health, particularly with the heightened risk of cardiovascular diseases and atherosclerosis, primarily driven by hypercholesterolemia [1, 2]. The intricate connection between hyperlipidemia and these cardiovascular conditions underscores the critical need for effective strategies to mitigate blood cholesterol levels, focusing especially on lowering low-density lipoprotein while concurrently increasing high-density lipoprotein levels.

Recognizing the clinical relevance of hyperlipidemia, recent dietary trends have shifted towards approaches aimed at reducing cholesterol levels and mitigating the adverse effects associated with synthetic agents [3]. Barley, an ancient grain utilized for animal feed, malting, and human consumption, emerges as a pivotal contributor to cardiovascular health by not only lowering cholesterol levels but also enhancing glucose tolerance [4, 5]. Of particular significance is barley flour, distinguished by its rich content of soluble dietary fibers such as arabinoxylans, ß-glucans, and pectin, with ß-glucans exhibiting prominence compared to other cereals. The consumption of barley flour brings about a range of beneficial effects, including diminished bile acid absorption, increased cholesterol breakdown, reduced lipoprotein cholesterol secretion, improved bile acid secretion, and an overall reduction in the body's cholesterol pool [6, 7].

Extensive documentation supports the therapeutic impact of barley on cholesterol levels and glucose tolerance, with soluble dietary fibers being identified as key contributors to these effects [8]. Mechanisms involving decreased bile acid absorption, enhanced cholesterol catabolism, and reduced lipoprotein cholesterol secretion contribute collectively to the cholesterol-lowering attributes of barley [9].

Soybean-based foods, acknowledged for their positive influence on human health, have witnessed increased consumption owing to their recognized nutritional benefits. A notable co-product, soybean hulls, constituting about 8% of the entire seed, stand out for their richness in complex carbohydrates, particularly known for their capacity to elevate high-density lipoprotein (HDL) cholesterol, often referred to as "good" cholesterol [10, 11].

The extensive study of soybean hulls, comprising approximately 8% of the soybean seed, has consistently highlighted their impact on high-density lipoprotein cholesterol. The complex carbohydrates within soybean hulls position them as an effective source of dietary fiber, consistently demonstrating an ability to elevate HDL cholesterol levels, a positive outcome intricately linked to enhanced cardiovascular health [12]. This dual significance underscores the value of soybean-based foods, not only as a nutritionally advantageous dietary choice but also as a source of co-products, like soybean hulls, with potential health-promoting effects.

This study endeavors to shed light on potential dietary interventions for managing hyperlipidemia and improving lipid profiles by systematically evaluating and comparing the effectiveness of diets supplemented with either barley or soybean in hyperlipidemic rats.

2. Material and Methods

2.1. Materials

Corn starch, corn oil, dietary lipids, and sucrose were purchased from the local market in Riyadh, Saudi Arabia. Other chemicals, including casein, vitamin mixture, salt mixture, choline chloride, and cholesterol, were purchased from Sigma Aldrich Co., USA, and were of analytical grade.

2.2. Methods

2.2.1. Plant Preparation

Before any processing, soybeans and barley were carefully inspected to eliminate any impurities or contaminants. Both soybeans and barley were subjected to a controlled drying process in an oven set at a temperature of 55 ℃ for a duration of 24 hours. This step was crucial to reduce moisture content and enhance the stability of the plant materials. Following the drying process, soybeans and barley were finely ground using a high-quality electric mixer. The grinding procedure aimed to obtain a homogeneous and fine powder, facilitating better incorporation into the experimental diets. The ground soybean and barley powders were subjected to particle size standardization to ensure uniformity. This step involved sieving the powders to achieve a consistent particle size suitable for inclusion in the experimental diets. The final powdered soybean and barley preparations were stored in airtight containers in a cool and dark environment to prevent degradation and maintain the nutritional content.

2.2.2. Animals Used in Experiments

A total of fifty male Sprague-Dawley rats weighing 80±5g were obtained from King Saud University's Animal Lab in Riyadh, Saudi Arabia. These rats were housed in a climate-controlled setting with meticulous humidity, temperature control, and a 12-hour light-dark cycle. As per the criteria provided by the Institute of Laboratory Animal Resources, Commission on Life Sciences, and National Research Council, they had free access to tap water and food [13]. Prior to the investigation, the rats were given a regular diet and spent a week becoming used to the research setting.

2.2.3. Diet Composition

The basal diet employed in this study was meticulously formulated based on specific quantities (expressed in grams per kilogram of diet) of selected ingredients, following the recommendations of the National Research Council [13]. The aim was to induce hyperlipidemia in the experimental rats and establish a hyperlipidemic rat model. The composition of the basal diet comprised corn starch (497 g), casein (200 g), corn oil (50 g), a vitamin mixture (20 g), a salt mixture (100 g), cellulose (30 g), choline chloride (3 g), and sucrose (100 g). To induce hyperlipidemia, the remaining rats were fed a high-fat diet containing cholesterol (1%) and dietary lipids (20%) for four weeks.

2.2.4. Experimental Design

Rats were randomly divided into five groups (n=10):

- Negative control group: continuous feeding with the normal diet.
- Positive control group: feeding with hyperlipidemic diets.
- Soybean-treated group: feeding with hyperlipidemic diets supplemented with 20% soybean powder.
- Barley-treated group: feeding with hyperlipidemic diets supplemented with 20% barley powder.
- Soybean and barley treated group: feeding with hyperlipidemic diets supplemented with a 20% blend of soybean and barley powder.

2.2.5. Blood Sample and Organ Collection

After 60 days of the experiment, euthanasia was performed on the rats, and blood as well as liver samples were collected for subsequent analysis.

2.2.6. Nutritional Parameters

The calculations were conducted for multiple parameters, including assessing body weight gain, food intake, and the feed efficiency ratio.

2.2.7. Biochemical Parameters

As described by Friedewald et al. and Bhardwaj *et al.* [14,15], a number of biochemical parameters were evaluated, including the lipid profile (total cholesterol, LDLc, VLDLc, and LDLc/HDLc), as well as the atherogenic indexes (TC/HDLc and LDLc/HDLc). Biochemical evaluations of liver function markers, including lactate dehydrogenase (LDH), alkaline phosphatase (ALP), and alanine and aspartate transferase (ALT & AST), were conducted using procedures described by Kind and King, Kumar et al., and Reitman and Frankel, respectively [16,17, 18]. The techniques developed by Mariami and Yoshikawa, Saydam et al., and Bergmeyer and Grabe [19, 20, 21] were used to measure the activity levels of serum glutathione peroxidase (GPX), superoxide dismutase (SOD), and catalase enzymes. The methods reported by Draper et al. and Green et al. [22, 23] were also used to evaluate the levels of serum nitric oxide (NO) and malondialdehyde (MDA). Estimates of liver cholesterol, total lipids, and glycogen levels were made in accordance with studies by Yoshioka et al. and Chabrol and Castellano [24, 25].

2.2.8. Histopathological Examination

The histopathological examination of the liver followed the protocols of Bancroft *et al.* [26].

2.3. Statistical Analysis

Quantitative measurements were statistically analyzed using one-way analysis of variance (ANOVA) and expressed as mean ± standard deviation (SD). The Duncan test was used to compare the means of the groups, and P values < 0.05 were used to indicate statistical significance.

3. Results

Table 1 provides a comprehensive overview of the impact of incorporating 20% barley, soybean powder, or a combination of both into hypercholesterolemic diets fed to rats, with a specific focus on nutritional outcomes. Following a sixty-day duration, the hypercholesterolemic positive control group exhibited a significant increase in both body weight gain and body weight gain percentage in comparison to the negative control group. In contrast, the groups treated with soybean, barley, and the soybean and barley blend demonstrated a consistent and gradual reduction in body weight gain, accompanied by a decline in body weight gain percentage throughout the experiment, presenting a notable contrast to the positive control group.

While food intake showed no significant differences between the negative control group and the groups treated with soybean, barley, and the blend, there was a noteworthy decrease in both food intake and feed efficiency ratio in comparison to the positive control group. These findings suggest that the dietary interventions involving soybean, barley, or their combination have a substantial impact on the nutritional parameters of the rats, influencing body weight gain, body weight gain percentage, food intake, and feed efficiency ratio.

The distinct letters (a, b, c, d) representing mean values in each column emphasize the statistically significant ($p \leq 0.05$) differences between the groups, underscoring the effectiveness of the dietary interventions in modulating the observed nutritional outcomes.

Table 2 delineates the repercussions of administering hypercholesterolemic rat groups with soybean or barley powder, or a combination of both, in comparison to the negative and positive control groups. The evaluation focused on serum lipid parameters at the conclusion of the experimental period. Rats treated with soybean, barley, or the soybean and barley blend exhibited a significant increase in total cholesterol (TC), triglycerides, LDL cholesterol, and VLDL cholesterol, accompanied by a notable decrease in HDL cholesterol when compared to the negative control group. Conversely, TC, triglycerides, LDLc, and VLDLc values demonstrated a significant reduction, while HDLc levels increased in comparison to the positive control group.

Table 1. Impact of feeding rats with hypercholesterolemic diets supplemented with 20% soybean or barley or a blend of both on nutritional values.

Groups	Body weight gain (g)	Body weight gain (%)	Food intake(g)	Feed efficiency ratio
Negative control	61.66±2.11^{c}	44.04±3.17b^{c}	18.16±1.02^{b}	0.0565±0.002^{b}
Positive control	90.77±5.14^{a}	63.92±5.11^{a}	21.27±2.11^{a}	0.0711±0.004^{a}
Soybean powder	65.88±3.12^{b}	46.06±3.17^{b}	18.88±1.11^{b}	0.0581±0.001^{b}
Barley powder	66.77±4.11^{b}	46.36±3.19^{b}	19.07±1.03^{b}	0.0564±0.003^{b}
Soybean and barley blend	64.78±3.17b^{c}	44.67±2.11b^{c}	18.97±1.20^{b}	0.0569±0.002^{b}

The atherogenic indexes (TC/HDLc and LDLc/HDLc) showed an increase in all hypercholesterolemic rat groups compared to the negative control group. However, the rat groups that received soybean, barley, or the combination demonstrated an improvement in atherogenic indexes, indicated by lower values compared to the positive control group. Particularly noteworthy is the rat group fed with the hypercholesterolemic diet supplemented with a 20% blend of soybean and barley, which exhibited the most significant improvement in serum lipid values and atherogenic indexes compared to the positive control group.

The distinct letters (a, b, c, d) representing mean values in each column underscore the statistically significant differences between the groups, emphasizing the effectiveness of the dietary interventions in modulating lipid profiles.

In the present study, the positive control group exhibited a highly significant increase in liver cholesterol and total lipid levels, coupled with a significant decrease in liver glycogen, when compared to the negative control group. The rat groups fed hypercholesterolemic diets supplemented with 20% soybean powder or barley powder displayed elevated levels of liver cholesterol and total lipids, accompanied by a significant decrease in liver glycogen compared to the negative control group. However, these rat groups, despite showing increased levels in liver cholesterol and total lipids, demonstrated a significant decrease in liver glycogen when compared to the positive control group.

Contrastingly, the rat group fed with a blend of soybean and barley did not exhibit such outcomes. The values of liver cholesterol, total lipids, and glycogen in this group were somewhat similar to those of the negative control group, as depicted in Table (3). This indicates that the combination of soybean and barley in the diet mitigated the adverse effects observed in the positive control group, maintaining liver cholesterol, total lipids, and glycogen levels at a comparable range to the negative control group.

The distinct letters (a, b, c, d) representing mean values in each column emphasize the statistically significant differences between the groups, underscoring the potential protective effects of the soybean and barley blend against the alterations induced by the hypercholesterolemic diet.

Table 2. Impact of feeding rats with hypercholesterolemic diets supplemented with 20% soybean or barley or a blend of both on lipid profiles.

Groups	Cholesterol (mg/dl)	Triglycerides (mg/dl)	HDLC (mg/dl)	LDLC (mg/dl)	VLDLC (mg/dl)	cholesterol / HDLC	LDLC/ HDLC
Negative control	125.71±10.24^{d}	105.21±9.22^{d}	40.77±4.11^{a}	63.9±6.11^{d}	21.04±2.11^{c}	3.08±0.33^{b}	1.56±0.23^{d}
Positive control	259.71±28.11^{a}	193.21±17.78^{a}	22.41±2.60^{c}	198.66±20.21^{a}	38.64±3.70^{a}	11.58±2.16^{a}	8.86±1.61^{a}
Soybean powder	175.66±18.66^{b}	131.71±11.22^{b}	35.11±3.71^{b}	114.21±10.21^{b}	26.34±3.11^{b}	5.01±1.12^{b}	3.25±0.55^{b}
Barley powder	170.44±17.76^{b}	130.96±12.14^{b}	34.71±3.60^{b}	109.54±9.6^{bc}	26.19±2.99^{b}	4.91±1.03^{b}	3.15±0.45^{b}
Soybean and barley blend	155.41±15.22^{c}	124.22±11.77^{c}	37.41±3.51^{ab}	93.16±9.07^{c}	24.84±2.88^{b}	4.15±1.01^{b}	2.49±0.30^{c}

Table 3. Impact of feeding rats with hypercholesterolemic diets supplemented with 20% soybean or barley or a blend of both on liver cholesterol, total lipids, and glycogen

Groups	Liver cholesterol (mg/g tissue)	Liver total lipids (mg/g tissue)	Liver glycogen (mg/100g tissue)
Negative control	4.11±0.51^{c}	37.22±3.95^{c}	8.11±1.33^{a}
Positive control	8.88±1.21^{a}	50.33±6.17^{a}	4.22±0.55^{c}
Soybean powder	6.11±1.03^{b}	43.41±4.60^{b}	6.71±0.69^{b}
Barley powder	6.41±1.36^{b}	44.21±4.96^{b}	6.55±0.77^{b}
Soybean and barley blend	5.22±0.88^{b}	40.14±5.11b^{c}	7.11±1.03^{a}

The findings presented in Table 4 unveiled a notable elevation in the levels of ALT, AST, ALP, and LDH enzymes in the positive control group when compared to the negative control group. Conversely, the groups that were fed with soybean and barley demonstrated a significant increase in liver function enzyme levels in comparison to the negative control group. However, these levels were significantly lower than those observed in the positive control group, indicating a potential mitigating effect of soybean and barley supplementation against the hypercholesterolemic-induced rise in liver enzyme levels.

In the rat group that consumed a blend of soybean and barley, there was a non-significant increase in ALT, ALP, and LDH levels. Notably, there was a significant increase in AST levels compared to the negative control group. These results suggest that while the combination of soybean and barley may exert a non-significant impact on certain liver function enzymes, it still contributes to maintaining these levels within a range comparable to the negative control group.

The distinct letters (a, b, c, d) representing mean values in each column emphasize the statistically significant differences between the groups, highlighting the potential protective effects of the soybean and barley blend on liver function enzymes when compared to the hypercholesterolemic positive control group.

Table 5 presents significant findings concerning serum MDA and NO levels, as well as the activity of SOD, GPX, and catalase enzymes. The positive control group exhibited a noteworthy increase in serum MDA and NO activity, coupled with a significant decrease in SOD, GPX, and catalase activity compared to the negative control group. This pattern is indicative of increased oxidative stress in the positive control group. In contrast, the soybean group demonstrated a significant reduction in serum MDA and NO activity compared to the positive control group. Interestingly, these values did not significantly differ from those of the negative control group, suggesting a potential protective effect of soybean against hypercholesterolemic-induced oxidative stress.

Furthermore, the soybean group exhibited a significant decline in the activity of SOD, GPX, and catalase enzymes compared to the negative control group. However, these values were significantly higher than those observed in the positive control group. These results imply that while soybean supplementation may lead to a reduction in antioxidant enzyme activity compared to the baseline, it still contributes to maintaining higher antioxidant levels than those induced by hypercholesterolemic conditions.

Similarly, both the barley and blend groups showed a significant decrease in serum oxidant levels (MDA and NO) and a significant increase in the activity of antioxidant enzymes (SOD, GPX, and catalase) compared to the positive control group. These findings indicate an enhancement in antioxidant status, with values approaching those observed in the negative control group.

The distinct letters (a, b, c, d) representing mean values in each column underscore the statistically significant differences between the groups, highlighting the potential antioxidative effects of soybean, barley, and their combination in ameliorating oxidative stress induced by hypercholesterolemic diets.

Table 4. The impact of feeding rats with hypercholesterolemic diets supplemented with 20% soybean or barley or a blend of both on liver function enzymes.

Groups	ALT (U/ML)	AST (U/ML)	ALP (U/ML)	LDH (IU/L)
Negative control	41.44±5.11^{c}	60.33±6.71^{b}	70.81±7.81^{c}	125.41±10.22^{c}
Positive control	70.33±9.03^{a}	101.22±12.11^{a}	113.22±11.21^{a}	195.77±31.41^{a}
Soybean powder	55.14±6.04^{b}	75.21±8.75^{b}	85.14±9.44^{b}	137.81±13.16^{b}
Barley powder	52.77±5.20^{b}	73.81±8.11^{b}	81.31±9.41^{b}	139.77±12.99^{b}
Soybean and barley blend	50.11±5.13^{b}	70.33±7.66^{b}	78.22±8.21^{b}	135.77±13.77^{b}

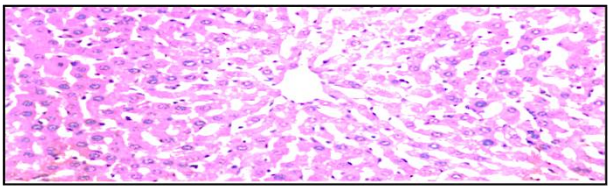
Table 5. Impact of feeding rats on hypercholesterolemic diets supplemented with 20% soybean or barley or a blend of both on serum MDA, NO, SOD, GPX, and catalase activity.

Groups	MDA (nm/mol)	NO (umol/l)	SOD (mmol/dl)	GPX (mmol/dl)	Catalase (mmol/dl)
Negative control	**4.61**±0.40^{b}	20.17±2.11^{b}	45.33±4.18^{a}	63.81±7.17^{a}	35.81±3.41^{a}
Positive control	7.01±1.21^{a}	30.11±2.91^{a}	22.77±2.14^{c}	36.51±4.21^{c}	20.14±2.11^{c}
Soybean powder	5.41±0.33^{b}	23.03±2.11^{b}	39.91±3.16^{b}	57.11±6.17^{b}	29.61±3.10^{b}
Barley powder	5.22±0.44^{b}	23.17±2.19^{b}	40.07±4.22^{ab}	56.14±5.11^{b}	31.14±3.41^{ab}
Soybean and barley blend	5.01±0.43^{b}	22.71±2.31^{b}	41.11±4.30^{ab}	60.41±6.71^{a}	31.61±3.27^{ab}

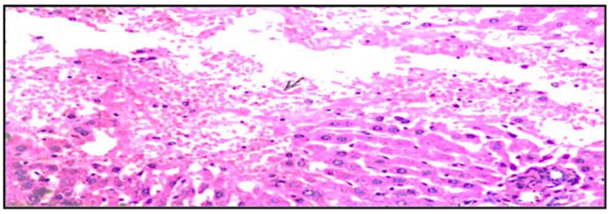
The microscopic examination of liver sections provides crucial insights into the histological changes induced by the experimental diets. In the negative control group (Figure 1a), the hepatic globule exhibited typical histological features, indicating a healthy liver structure. Conversely, livers from the positive control group (Figure 1b) displayed significant pathological alterations, including vacuolar degeneration of hepatocytes and hepatic hemorrhage. These changes are indicative of liver damage and compromised structural integrity.

Intriguingly, the group treated with soybean powder (Figure 1c) exhibited minimal histopathological changes, suggesting a protective effect against severe liver damage. Similarly, livers from the barley powder-treated group (Figure 1d) showed signs of activation of Kupffer cells and dilated hepatic sinusoids. While these changes may indicate a response to dietary interventions, they appear less severe compared to the positive control group.

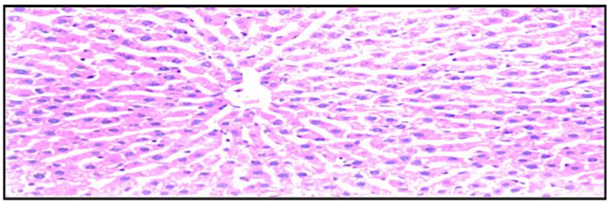
Remarkably, the livers of rats treated with a blend of soybean and barley powder (Figure 1e) displayed histological features resembling a normal hepatic parenchyma. This suggests a potential synergistic or additive effect of combining soybean and barley in ameliorating histopathological alterations induced by hypercholesterolemic diets. The observed protective effects in these groups highlight the significance of dietary interventions in preserving liver histology and function, warranting further investigation into the underlying mechanisms of such effects.



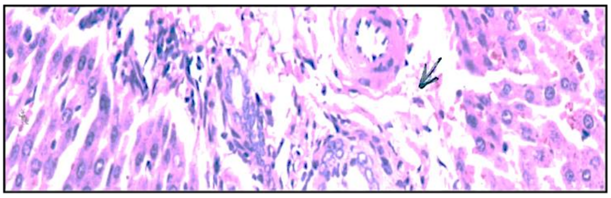
(a): Control (–ve) group revealed the normal histology of hepatic globule.



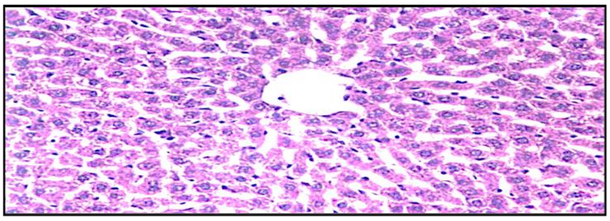
(b): Control (+ve) group revealed vacuolar degeneration of hepatocytes. and hepatic hemorrhage



(c): Liver of rat from soybean powder group showed minor histopathological changes



(d): Liver of rat from barley powder group showed kupffer cells activation and dilatation of hepatic sinusoids



(e): Liver of rats from soybean and barley blend group showed apparent normal hepatic parenchyma

Figure 1. Microscopic analysis of liver sections (from "a" to "e")

4. Discussion

The enhancement observed in the nutritional values of rats consuming a hypercholesterolemic diet supplemented with soybean and barley can be attributed to the rich presence of essential nutrients, minerals, and bioactive compounds in these dietary components. Soybean, recognized for its high protein content, and barley, containing proteins like glutelins and hordeins along with soluble dietary fibers such as ß-glucans, contribute significantly to the overall nutritional improvement [27]. Barley, with its nutritional composition of protein (10-17%), ß-glucan (4-9%), starch (65-68%), free lipids (2-3%), and minerals (1.5-2.5%), stands out as a valuable dietary source [4]. Furthermore, the complex carbohydrates, low fat content, balanced protein, vitamins, minerals, antioxidants (polyphenolics), and soluble and insoluble fiber in barley grain contribute to its high nutritional value [4]. Particularly, dietary fiber plays a pivotal role in retarding gastric emptying, delaying nutrient exposure to digestive enzymes, and altering absorption in the small intestine [28, 29].

The study's findings highlighted that the hyperlipidemic positive control group exhibited elevated serum levels of cholesterol, triglycerides, LDLc, VLDLc, and atherogenic indexes (cholesterol/HDLc & LDLc/HDLc), coupled with reduced HDLc levels. These observations align with previous studies by Ji and Gong, Hamza and Mahmoud, and Amer [30, 31, 32], confirming the association between hyperlipidemia and adverse lipid profiles. The diets supplemented with soybean and barley, as evidenced in Tables 2 and 3, demonstrated hypocholesterolemic effects, and this can be attributed to the fiber content in these ingredients. Numerous studies have shown that soybean protein reduces intestinal cholesterol absorption, enhances bile acid secretion, increases the elimination of LDLc, and reduces liver cholesterol content [33, 34]. These findings are consistent with research by Shimizu et al., indicating that dietary fiber can lower LDLc levels by influencing lipid absorption and transport, thereby altering the site of intestinal absorption [35]. Additionally, Sirtori et al. noted that soybean, rich in high-quality amino acids, directly regulates LDL receptors, contributing to its hypocholesterolemic effect [36]. The components of soybean protein, such as fiber, phytic acid, saponins, and phytosterols, collectively play a role in this cholesterol-lowering effect.

Particularly, soluble fiber plays a vital role in reducing serum cholesterol by increasing the loss of bile acids in feces, promoting bile acid synthesis, and thereby reducing cholesterol availability for lipoprotein synthesis. The amount of fiber intake is correlated with changes in lipid profiles, and higher amounts have been shown to decrease total cholesterol and LDLc without affecting HDLc levels [37, 38, 39]. The hypocholesterolemic effects observed in the soybean and barley blend group align with findings by Tham et al., Chung et al., and Kingsley et al., suggesting that isoflavones in soybeans, structurally similar to estrogens, bind to estrogen receptors, up-regulate LDL receptors, and inhibit endogenous cholesterol synthesis [40, 41, 42].

The study's results also revealed decreased levels of liver cholesterol and total lipids, along with an increased level of liver glycogen in rats fed soybeans and barley compared to the positive control group, as presented in Table 3. These outcomes may be attributed to stimulated cholesterol synthesis and reduced lipoprotein cholesterol output. The increased levels of liver cholesterol and total lipids compared to the negative control group are associated with increased cholesterol synthesis and the activity of microsomal 3-hydroxy-3-methylglutaryl coenzyme A reductase in the liver. Dietary fiber, by reducing gut transit time, interferes with hepatic cholesterol synthesis [9, 43].

Moreover, the study results indicate that the positive control group exhibited higher levels of liver function enzymes, as lipid accumulation in hepatocytes can elevate aminotransferases released from damaged liver cells, serving as a parameter for liver injury [9, 44, 45]. Groups supplemented with soybean and barley showed improvements in serum liver enzymes compared to the positive control group. These findings are consistent with results by Abulnaja and El Rabey, Sofi et al., and Sahajpal et al. [46, 47, 48]. Specifically, β-glucan found in barley has hypolipidemic effects and significantly improves liver enzymes by reducing fat accumulation in the liver. β-Glucan also exhibits antioxidant effects, protecting the epithelium from injury. The oxidative stress caused by hypercholesterolemic diets is associated with atherogenesis. Dietary polyphenols have potent antioxidant activity and an inverse relationship with cardiovascular risk [49, 50]. The protective effects observed in the supplemented groups underscore the potential of soybean and barley in mitigating liver damage associated with hypercholesterolemic diets.

The findings of the current study underscore the synergistic impact of combining soybean protein and barley, showcasing additive effects on cholesterol reduction and antioxidant activity. This aligns with prior research by Iritani et al., Demonty et al., and Yang et al., which emphasized the inhibitory effects of soybean protein on lipid absorption, enhanced lipid excretion, and decreased lipogenic enzyme activities and fatty acid synthesis in the rat liver [51, 52, 53]. Moreover, soybean protein was found to enhance plasma lipoprotein lipase activity more effectively than casein, adding another layer to its lipid-modulating properties. Epidemiological evidence supports the notion that soybeans encompass crucial bioactive components such as globulin proteins (glycinin & beta-conglycinin), phytosterols (sitosterol, sitostanol, campesterol), phytoestrogen oligosaccharides, and antioxidants like isoflavones (daidzen, genistein, glycitein). Isoflavones, specifically, act as functional cellular antioxidants, particularly beneficial under high-fat diet conditions. These components collectively play pivotal roles in lowering cholesterol levels and safeguarding against various hyperlipidemic side effects [54, 55, 56].

Barley, as the other component in the blend, contributes to these additive effects. Rich in polyphenols and total flavonoids, barley exhibits potent antioxidant effects [57, 58, 59]. The polyphenolic compounds in barley are known for their ability to neutralize free radicals and counteract oxidative stress. This antioxidant potential plays a crucial role in protecting cells and tissues from damage caused by hyperlipidemic conditions. The combination of soybean and barley in the diet, as supported by the current study, thus emerges as a promising strategy not only for reducing cholesterol levels but also for enhancing the antioxidant defense mechanisms in the body. The complementary nature of soybean and barley, each bringing its unique set of bioactive compounds to the blend, highlights the potential of this dietary combination in promoting cardiovascular health and mitigating the adverse effects of hyperlipidemia.

5. Conclusions

In summary, this research manuscript highlights the positive impact of introducing soybean and barley at a 20% dietary dose into a hypercholesterolemic diet. The supplementation of these ingredients significantly improved the nutritional profile of the rats by providing essential nutrients, minerals, and bioactive compounds. The high protein content in soybean, combined with barley's proteins like glutelins and hordeins, along with soluble dietary fibers such as ß-glucans, collectively contributed to the overall nutritional enhancement. The hypocholesterolemic effects of soybean and barley were evident, as indicated by reduced serum levels of cholesterol, triglycerides, LDLc, VLDLc, and atherogenic indexes, coupled with increased HDLc levels. The favourable outcomes observed in this study can be ascribed to the rich fiber content present in both soybean and barley, contributing to their interference with cholesterol and bile acid absorption and metabolism. Notably, the liver function of rats that consumed diets supplemented with soybean and barley exhibited notable improvement, as evidenced by reduced liver cholesterol and total lipid levels, coupled with elevated liver glycogen levels. Furthermore, the supplementation resulted in a significant decrease in liver enzyme levels, indicative of reduced liver damage and an overall enhancement in liver health.

The synergistic effects of soybean protein and barley were particularly evident, showcasing additive benefits, especially regarding cholesterol reduction and antioxidant properties. The isoflavones present in soybean acted as potent cellular antioxidants, complemented by the polyphenols and flavonoids found in barley, collectively exerting robust antioxidant effects. This comprehensive analysis underscores the multifaceted advantages of incorporating soybean and barley into the diet, emphasizing their potential not only in improving lipid profiles but also in promoting liver health. The balanced combination of these two ingredients emerges as a promising dietary strategy, offering diverse health benefits that extend beyond lipid modulation, ultimately contributing to overall well-being.

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