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Original Article

Plasma Cholesterol Level Reduction in Albino Rats by β-D Glucan (Pleuran) from *Plurotus ostreatus*

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Abstract

Background: The progression of atherosclerotic lesions and coronary artery disease is known to be accelerated by hypercholesterolemia. **Objective:** This study investigated at the effects of adding an extract of oyster mushroom fruiting bodies to rats with high and normal cholesterol levels on their biochemical and histological changes. This study was aimed to assess the effect of *Pleurotus ostreatus* extraction on hypercholesterolemia in albino rats and show the revelation of the relationship of these with some biochemical parameters. **Materials and Methods:** Three sets of ten albino rats each were formed from 6-week-old rats. To lower plasma total cholesterol, triglycerides, low-density lipoprotein (LDL), total lipid, phospholipids, and LDL/high-density lipoprotein, extracts of *P. ostreatus* fruiting bodies were fed to hypercholesterolemic rats. **Results:** In rats with high cholesterol, feeding oyster mushrooms drastically lowered body weight. The excretion of total lipids and cholesterol in feces was enhanced by feeding mushrooms. **Conclusion:** These findings imply that *P. ostreatus* food supplement improved health by altering hypercholesterolemic rats' atherogenic lipid profiles.

Keywords: β-D glucan, high-density lipoprotein, plasma cholesterol, *Plurotus ostreatus*

Introduction

Pleurotus ostreatus is a genus that is utilized for both food and medicine because of its flavor, scent, and nutritional value.[1] There are around 40 species in the genus *Pleurotus*. Pleurotus is extensively dispersed in Iraq, and rice straw is the main substrate utilized to produce *Pleurotus* species there.[2] There are just a few fruit bodies offered in Iraqi markets, and they are almost usually either Pleurotus or Agaricus mushrooms.[3] Pleurotus fruiting bodies were said to produce at their highest levels in the winter (14–27°C, 70%–80% relative humidity). It has been demonstrated that *P. ostreatus* extracts have antifungal, hypolipidemic, immunostimulant, antiproliferative, and anticancer properties in addition to being anti-inflammatory and antioxidant properties.[4] An edible fungus called *P. ostreatus* has a variety of beneficial components, such as β-glucans. They are soluble fibers that are produced by various bacteria, plants, and fungi in their cell walls. They can reduce blood pressure, the risk of heart attacks, and several metabolic processes including lipid and glucose metabolism. It can also be used in diets to manage body weight (food). According to studies, the fruiting body and mycelium of the oyster mushroom may also be extracted using various solvents to produce a variety of beneficial chemicals (*P. ostreatus*).[5] They can be advantageous in several health-promoting actions, such as probiotic and immunomodulatory ones. Extracts of *P. ostreatus* are used in various solvents and polysaccharides. In animal cells as well as human cells, *P. ostreatus* has demonstrated anti-cancer properties. In 2020, it was revealed that oyster mushrooms (*P. ostreatus*) are a popular nutrient due to its high nutritional content and being readily available. The -glucan, a glucose polymer that binds to the glycoside ring on -1,3-glucan and -1,4-glucan, is among its constituents. A-glucan makes up between 80% and 90% of the fungal cell wall. Studies have demonstrated that β-glucan is utilized as an antioxidant, anti-tumor, anti-cholesterol, anti-aging element, as well as

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to boost the body's immune system.[6-8] Oyster mushrooms, or *P. ostreatus*, are becoming more widely acknowledged as valuable dietary items that play a key role in human nutrition and health.[9] It is well acknowledged that atherosclerosis may be avoided by significantly reducing high plasma cholesterol levels. Because of their high dietary fiber, sterol, protein, and microelement content, oyster mushrooms are an excellent food for both preventing and treating hypercholesterolemia.[5] The common belief that lowering blood cholesterol levels is crucial for lowering the risk of developing atherosclerosis has sparked research into and study of natural compounds having hypocholesterolemic action.[10] Therefore, this study was aimed to assess the effect of *P. ostreatus* extraction on hypercholesterolemia in albino rats and show the revelation of the relationship of these with some biochemical parameters.[11]

Materials and Methods

Pleurotus ostreatus

From several agricultural and woodland locations in Mosul, fresh fruiting bodies of *P. ostreatus* were collected. At the biology department of the University of Mosul in Iraq, a pure culture was added to the collection. 48 h of hot air drying at 40°C for fresh fruiting bodies was followed by pulverization.

Animals

From the College of Veterinary Medicine at the University of Mosul, 40 albino rats weighing 205 ± 20 g were acquired. The animals were housed in normal cages and had full access to food and water (a typical laboratory pellet diet). With a 12-hour light/dark cycle, the temperature in the animal habitat was controlled between 24 and 29°C and then randomly divided into four groups which content each group 10 albino rat as follows:

Group 1 (negative control): 60 days feeding of rats on a typical synthetic diet

Group 2 (hyper-cholesterol): high fat was induced by injecting them with a medical syringe with pure coconut oil prepared by Al-Emad Company at a rate of 1.5 mL/day.

Group 3 (pure D-glucan + coconut oil): the participant rats were given pure coconut oil and D-glucan at a rate of 1.5 mL/day by injecting them with a medical syringe.

Group 4 (standard D-glucan + coconut oil): the participant rats were given pure coconut oil and D-glucan at a rate of 1.5 mL/day by injecting them with a medical syringe.[2]

The investigation's procedure was approved by the institution's animal ethics committee.

Biochemical analysis

Using an automated COBAS INTEGRA 800VR (Roche Diagnostics GmbH, Mannheim, Germany), the biochemical parameters, such as the lipid profile, which included triglycerides, TC (total cholesterol), HDL-C (high-density lipoprotein cholesterol), and LDL-C (low-density lipoprotein cholesterol) were analyzed.[12]

β-D-Glucan assay

The Fungitell test was used to measure the β-D-glucan concentration (Associates of Cape Cod, East Falmouth, MA). Using an Ascent Multiskan equipment, all samples were examined in accordance with the manufacturer's methodology (Thermo Fisher Scientific, Waltham, MA). In accordance with the directions, the tests were carried out twice in the same run. The usage of glucan-free products prevented interference. For the same sample, the mean of the two outcomes was determined. When the coefficient of variation for β-D-glucan concentrations below 200 pg/mL exceeded 20%, the analysis was repeated; when the coefficient of variation for β-D-glucan concentrations over 200 pg/mL exceeded 100%, the analysis was repeated. With a mean β-D-glucan concentration of 211.16 pg/mL, the analysis was repeated in one sample because the coefficient of variance was higher than 100%.[1]

Statistical analysis

Results are presented as mean values and standard deviations. Post-hoc tests were used after a one-way analysis of variance to examine intergroup differences. statistical package for the social sciences (SPSS) version 28 was employed (SPSS Inc., Chicago, IL). Statistics were judged significant at $P < 0.05$.

Ethical consideration

Not Applicable.

Results

The effects of β-D-glucan on rats' body weight among study groups were shown in Table 1. The results revealed that β-D-glucan feeding significantly increased body weight in hypercholesterolemic group and nonhypercholesterolemic groups [see Table 1].

Table 2 shows the effects of β-D-glucan on rats' total cholesterol among study groups. The results revealed that TC level significantly increased in hypercholesterolemic group in all dosage periods (113.45, 112.96 and 110.03) mg/dL, respectively. With β-D-glucan and coconut oil group, the results showed that TC level slightly increased in β-D-glucan and coconut oil group in both 30- and 45-day dosage periods (81.33 and 74.93) mg/dL, respectively, while decrease in 60 days dosage period (67.86 mg/dL). With standard β-D-glucan + coconut oil, the TC level was significantly increased in all dosage periods (86.46, 84.65, and 83.80) mg/dL, respectively, whereas no significant difference was reported in the among groups before starting the dosage [Table 2].

The effects of β-D-glucan from *P. ostreatus* on triglyceride in the blood of rats among study groups were shown

Table 1: Effects of β-D-glucan on rats' body weight among study groups

Study groups	Initial body weight	Body weight after 30 days	Body weight after 45 days	Body weight after 60 days
Control	188.66 ± 3.98a	244.16 ± 2.32a	261.82 ± 2.84a	282.93 ± 3.91a
Hypercholesterol	189.66 ± 4.35a	257.00 ± 5.37b	275.29 ± 4.97b	297.33 ± 5.11b
d-Glucan + coconut oil	190.00 ± 5.20a	229.83 ± 3.36c	247.58 ± 2.85c	258.66 ± 3.06c
Standard d-glucan + coconut oil	193.33 ± 3.59a	232.33 ± 2.51c	256.79 ± 5.46c	269.83 ± 6.09c

Different letters mean difference is significant at 0.05 level (ANOVATest)
Same letters mean difference is non-significant at 0.05 level (ANOVA-Test)

Table 2: The effects of D-glucan extracted from *P. ostreatus* on total cholesterol (mg/dL) in the blood of rats

Study groups	Before injection	After 30 days	After 45 days	After 60 days
Control	67.55 ± 2.46a	84.75 ± 2.95a	87.37 ± 1.62a	91.40 ± 1.17a
Hypercholesterol	66.08 ± 1.75a	113.45 ± 6.06b	112.96 ± 4.75b	110.03 ± 3.43b
β-D-Glucan + coconut oil	68.78 ± 2.98a	81.33 ± 4.61a	74.93 ± 5.89ac	67.86 ± 6.61ac
Standard β-d-glucan + coconut oil	64.78 ± 1.71a	86.46 ± 4.74a	84.65 ± 6.26a	83.80 ± 5.19a

Different letters mean difference is significant at 0.05 level (ANOVA test)
Same letters mean difference is non-significant at 0.05 level (ANOVA test)

Table 3: The effects of D-glucan from *P. ostreatus* on triglyceride in the blood of rats

Study groups	Before injection	After 30 days	After 45 days	After 60 days
Control	75.86 ± 2.97a	77.08 ± 4.19a	87.95 ± 2.39a	100.78 ± 1.70a
Hypercholesterol	70.23 ± 4.01a	119.33 ± 4.23c	108.36 ± 6.86b	120.11 ± 8.14b
d-Glucan + coconut oil	72.13 ± 3.53a	100.93 ± 4.01b	82.97 ± 5.93c	65.85 ± 6.46c
Standard d-glucan + coconut oil	67.80 ± 4.10a	80.15 ± 5.69a	79.04 ± 7.58c	77.46 ± 6.66c

Different letters mean difference is significant at 0.05 level (ANOVA test)
Same letters mean difference is non-significant at 0.05 level (ANOVA test)

Table 4: The effects of D-glucan from *P. ostreatus* on HDL (mg/dL) in the blood of rats

Study groups	Before injection	After 30 days	After 45 days	After 60 days
Control	29.55 ± 1.33a	32.60 ± 1.39a	32.05 ± 0.84a	31.98 ± 069a
Hypercholesterol	28.91 ± 0.86a	23.88 ± 0.79cd	22.98 ± 0.74d	22.63 ± 0.65d
D-Glucan + coconut oil	27.88 ± 1.62a	45.45 ± 2.58bc	45.08 ± 0.73c	44.68 ± 0.77c
Standard D-glucan + coconut oil	26.08 ± 1.18a	44.38 ± 84b	42.89 ± 0.89b	41.86 ± 1.32b

Different letters mean difference is significant at 0.05 level (ANOVA test)
Same letters mean difference is non-significant at 0.05 level (ANOVA test)

in Table 3. The results showed that triglyceride level significantly increased in hypercholesterolemic group in all the dosage periods (119.33, 108.36, and 120.11) mg/dL, respectively with β-D-glucan and coconut oil group. The results showed that triglyceride level slightly increased in β-D-glucan and Coconut Oil group in both 30 and 45 days dosage periods (100.93 and 82.97) mg/dL, respectively, whereas a decrease in 60 days dosage period was noted (65.85 mg/dL).

With standard β-D-glucan + coconut oil, the triglyceride level was significantly increased in all dosage periods (80.15, 79.04, and 77.46) mg/dL respectively. While no significant difference was reported in the among groups before starting the dosage [Table 3].

Table 4 shows the effects of β-D-glucan from *P. ostreatus* on HDL in the blood of rats among the study groups. The results showed that HDL level significantly reduced in hypercholesterolemic group in all the dosage periods (23.88, 22.98, and 22.63) mg/dL, respectively. With β-D-glucan and coconut oil group, the results showed that HDL level significantly increased in β-D-glucan and coconut oil group in all dosage periods (45.45, 45.08, and 44.68) mg/dL, respectively. With standard β-D-glucan and coconut oil group, the results showed that HDL level significantly increased in standard β-D-glucan and coconut oil group in all dosage periods (44.38, 42.89, and 41.86) mg/dL, respectively, than before injection (26.08 mg/dL), whereas a decrease in 60 days dosage period was noted 65.85 mg/dL).

The effects of β-D-glucan from *P. ostreatus* on very low-density lipoprotein (VLDL) in the blood of rats among study groups are shown in Table 5. The results showed that LDL level significantly raised in hypercholesterolemic group in all the dosage periods (62.47, 63.21, and 64.97) mg/dL, respectively.

Table 5: The effects of β-D-glucan from *P. ostreatus* on LDL (mg/dL) in the blood of rats

Study groups	Before injection	After 30 days	After 45 days	After 60 days
Control	22.94 ± 2.51a	43.36 ± 2.77a	38.47 ± 5.94a	36.02 ± 4.76a
Hypercholesterol	23.83 ± 1.69a	62.47 ± 3.15b	63.21 ± 7.78b	64.97 ± 6.46b
β-D-glucan + coconut oil	24.64 ± 2.39a	14.07 ± 2.22c	27.67 ± 5.61a	32.48 ± 4.53a
Standard D-glucan + coconut oil	25.40 ± 1.91a	32.33 ± 5.62c	30.93 ± 6.09a	29.09 ± 6.09a

Different letters mean difference is significant at 0.05 level (ANOVA test)
Same letters mean difference is non-significant at 0.05 level (ANOVA test)

Table 6: The effects of D-glucan from *P. ostreatus* on VLDL (mg/dL) in the blood of rats

Study groups	Before injection	After 30 days	After 45 days	After 60 days
Control	15.090 ± 0.63a	15.40 ± 0.83a	18.62 ± 0.86a	20.15 ± 0.34a
Hypercholesterol	14.047 ± 0.80a	23.86 ± 0.84c	23.99 ± 0.92b	24.02 ± 1.62b
β-D-glucan + coconut oil	14.427 ± 0.70a	20.18 ± 0.80b	16.74 ± 2.06c	13.17 ± 1.29c
Standard D-glucan + coconut oil	13.560 ± 0.83a	16.03 ± 1.13a	15.89 ± 1.81c	15.49 ± 1.33c

Different letters mean difference is significant at 0.05 level (ANOVA test)
Same letters mean difference is non-significant at 0.05 level (ANOVA test)

With β-D-glucan and coconut oil group, the results revealed that LDL level significantly increased in β-D-glucan and coconut oil group in both 45and 60 days dosage periods (27.67 and 32.48) mg/dL, respectively, whereas reduced in 30 days dosage periods (14.07). With standard β-D-glucan and coconut oil group, the results showed that LDL level significantly increased in standard β-D-glucan and coconut oil group in all dosage periods (32.33, 30.93, and 29.09) mg/dL, respectively than before injection (25.40 mg/dL).

The effects of β-D-glucan from *P. ostreatus* on very low-density lipoprotein (VLDL) in the blood of rats among study groups are shown in Table 6. The results documented that VLDL level significantly raised in hypercholesterolemic group in all dosage periods (23.86, 23.99, and 24.02) mg/dL, respectively, compared to before injection (14.047 mg/dL).

With β-D-glucan and coconut oil group, the results revealed that VLDL level significantly increased in β-D-glucan and coconut oil group in both 30 and 45 days dosage periods (20.18 and 16.74) mg/dL, respectively, whereas reduced in 60 days dosage periods (13.17 mg/dL) compared to before injection (14.427 mg/dL). With standard β-D-glucan and coconut oil group, the results showed that VLDL level significantly increased in standard β-D-glucan and coconut oil group in all the dosage periods (16.03, 15.89, and 15.49) mg/dL, respectively than before injection (13.560 mg/dL) [Table 6].

Discussion

The current study showed that rats fed oyster mushrooms had dramatically improved plasma atherogenic lipid profiles compared to rats with experimentally generated hypercholesterolemia.

Rats have a significant ability to regulate their plasma cholesterol levels and are particularly resistant to the development of hypercholesterolemia and atherosclerosis.[13]

Therefore, cholesterol feeding is combined with other additives, such as bile acids and propylthiouracil (an anti-thyroid medication), which improve intestinal absorption of cholesterol, to cause hypercholesterolemia or atherosclerosis in rats. However, because cholesterol feeding itself stimulates bile acid production by around three to fourfold in rats, the current investigation found that adding cholesterol to the baseline diet caused hypercholesterolemia in the animals. The rats in the current investigation had an increase in plasma cholesterol of 30.18%, which was comparable to the value reported by Bobek *et al.*,[14] who supplemented their rats' diets with bile acids (0.5%) and fed them cholesterol (0.3%) found that the cholesterol-fed rats had 1.7 times greater cholesterolemia than the control rats. It is unclear how mushrooms affect plasma lipoprotein levels in HC rats to lower them. Mevnolin, a hypocholesterolemic substance found in mushrooms (also known as monacolin K or lovastatin), may have a role in lowering the activity of the enzyme 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase, which is the rate-limiting enzyme in cholesterol formation. Therefore, by decreasing HMG-CoA reductase activity, eating mushrooms may result in the inhibition of endogenous cholesterol production.[15]

The impact of edible mushrooms on hypercholesterolemia has been shown by a number of authors/researchers. The blood and hepatic cholesterol levels of the male Wistar rats receiving the oyster mushroom diet were noticeably lower.[16] Similar to this, *P. ostreatus* decreased the rat's serum cholesterol concentration.[12] Similarly, eating mushrooms concurrently with a high-fat meal considerably lowers total cholesterol levels.[17] As a result, the hypocholesterolemic

effects seen in the current research *P. ostreatus* were successful in decreasing the blood levels of both total and LDL cholesterol as well as the quantity of total cholesterol in the liver. When feeding the albino Wistar rats a diet, they found a reduction in total cholesterol and total lipid levels with extended treatment time.[12] According to Kadhim and Amin,[21] a rise in plasma high-density lipoprotein concentrations was seen along with a decrease in total cholesterol, LDL, and triglyceride concentration. After a 45- and 60-day feeding study with a mushroom diet, plasma total cholesterol, and low-density lipoprotein cholesterol concentration were found to be considerably lower than control, whereas the HDL level was significantly increased.[11,18] The current results are consistent with these prior publications. In the current study, the *P. ostreatus* treated group of rats showed significantly lower levels of total cholesterol, LDL cholesterol, and HDL cholesterol than the group that just had a high-fat diet. The risk of coronary heart disease in humans and LDL cholesterol concentration in serum have been linked positively in clinical research; therefore, improving the HDL to LDL ratio is crucial for preventing atherosclerosis. The development of coronary heart disease is mostly brought on by excessive levels of cholesterol, particularly LDL cholesterol.[19] In the current study, rats who received varying doses of P. eous food also had higher HDL levels than the control group. In contrast, rats fed a diet high in mushrooms had much lower LDL levels than the control group. Thus, compared to control rats, the rats fed the mushroom diet had reduced LDL/HDL cholesterol ratio, which is regarded to be the atherogenic indicator of lipoproteins.[20] According to Rennerova *et al*.,[1] plasma high-density lipoprotein concentrations significantly increased when total cholesterol, LDL, and triglyceride concentrations decreased. In conclusion, according to the findings of the current study, *P. ostreatus* is a key modulator of the lipid profile of albino rats, decreasing cholesterol, triglycerides, HDL, LDL, and VLDL considerably.[21]

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Nil.

Conflicts of interest
There are no conflicts of interest.

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