

The Potential Effect of Ethanolic Reishi Mushroom (*Ganoderma lucidum*) Extract on Immunity of Hepatic Rats

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مجلة البحوث في مجالات التربية النوعية

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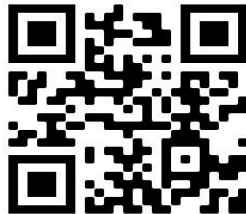
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التأثيرات المحتملة لمستخلص الفطر الريشى على مناعة الفئران المصابة بالتسمم الكبدى

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ملخص البحث

يهدف هذا البحث إلى دراسة التأثيرات المحتملة لمستخلص الفطر الريشى على مناعة الفئران المصابة بالتسمم الكبدى باستخدام ثلاث جرعات من المستخلص الكحولى للفطر الريشى (120 و 240 و 360 ملجم لكل كجم من وزن الفأر) عن طريق الفم. تم استخدام خمسة وعشرون (25) من ذكور فئران الالبينو الناضجة والتي تزن 190 ± 10 جرام. تم تقسيمهم إلى مجموعتين رئيسيتين المجموعة الاولى تحتوى على خمس فئران وعوملت كمجموعة ضابطة سالبة والمجموعة الثانية تحتوى على 20 فأر وقد تم حقنهم برابع كلوريد الكربون مرتين فى الاسبوع لمدة 15 يوم للإصابة بالتسمم الكبدى ثم تم تقسيمهم الى اربع مجاميع تركت المجموعة الاولى كمجموعة ضابطة موجب و تم إعطاء الثلاث مجموعات المتبقية جرعات مستخلص الفطر الريشى بتركيزات 120 و 240 و 360 ملجم لكل كجم من وزن الفار عن طريق الفم لمدة 28 يوم . وقد أظهرت النتائج ارتفاع فى مستوى خلايا الدم البيضاء و الخلايا الليمفاوية والقائلة و الجلوبيولينات المناعية وكذلك مضادات الاكسدة (سوبر اوكسيد ديسميوتيزوالكتاليزيما حدث انخفاض فى مستوى سكر الدم و انزيمات الكبد و عامل نخر الورم والمالونالدهيد وذلك بالمقارنة بالمجموعة الضابطة الموجبة . من خلال النتائج المتحصل عليها توصى الدراسة بضرورة استخدام الفطر الريشى فى وجباتنا وذلك لتحسين المناعة ووظائف الكبد.

الكلمات المفتاحية: الجلوبيولين المناعى ، رابع كلوريد الكربون ، عامل النكرزة الورمى ، مؤشرات وظائف الكبد

The Potential Effect of Ethanolic Reishi Mushroom (*Ganoderma lucidum*) Extract on Immunity of Hepatic Rats

Basma R.M.khateib

Abstract

The purpose of this research was to investigate the effect of ethanolic reishi mushroom (*Ganoderma lucidum*) extract on immunity in hepatic rats at three various doses (120, 240 and 360 mg/kg body weight), orally. Twenty five (25) mature male albino rats weighting 190 ± 10 g were used in this research. The rats were divided into two main groups, the first group (5 rats) still fed on basal diet and the second main group (20 rats) had given CCl_4 twice a week for 15 days to induce liver impaired in rats, then classified into four sub groups (5 rats in each group) one of them left as control positive and other three groups fed on three doses of mushroom extract (120, 240 and 360 mg/kg body weight) orally for 28 days. The obtained results of hepatic rats revealed that reishi mushroom extract showed a significant increase in WBC, lymphocytes, killing cells, IgG, IgM, SOD and CAT but with significant decreases in liver functions markers (ALT, AST and ALP), serum glucose level, TNF- α and MDA as compared with control positive group. In conclusion, the study recommended using reishi mushroom in our daily diets to improve immunity and liver functions.

Key word: IgG - CCl_4 - TNF- α - liver functions markers.

Introduction

Mushrooms are a group of macro-fungi with conspicuous epigenous or hypogenous fruiting bodies (**Chang and Buswell, 1996**). The reishi mushroom belongs to phylum Basidiomycota and Family *Ganodermataceae* of Aphyllophorales (**Yang et al., 2000**). *Ganoderma lucidum* is a medicinal mushroom with fascinating chemical composition imparting nutraceutical benefits and pharmacological properties either as immune suppressors, hypocholesterolemic agents or as coadjuvant treatments in diseases such as cancer and chronic hepatitis (**Zhao et al., 2016**).

Nutritionally, mushrooms give many of dietary benefits found in bean, meat and grains. Vegetarians consume mushroom as a main part of their meal. **Garuba et al., (2020)** reported that the amount of carbohydrate in reishi mushroom was the highest (44.95%), followed by protein (15.75%). The amount of moisture and crude fibre were 12.99% and 14.81% , respectively. Many vitamins have been identified in reishi mushroom , such as vitamins C, D , E , B1, B2, B6 and β -carotene. Moreover, several minerals such as phosphorus , potassium ,calcium, sodium, carbon, iron, magnesium, chromium, zinc, arsenic, manganese, copper, silicon, aluminum, cobalt and lead have been reported from reishi mushroom (**Ahmad, 2018**). About eighteen types of amino acids have been found in reishi mushroom , and leucine was the most abundant amino acid in the mushroom , which has strong antioxidant and hypoglycemic activities (**Zhang et al., 2018a, 2018b**). β -N-Acetylhexosaminidase, endo- β -1,3-glucanase, β -1,3-glucanase, and glutamic protease were isolated from reishi mushroom , and glutamic protease is the major one in the reishi mushroom extract (**Kumakura et al., 2019**). Nutritional composition of reishi mushroom reveals high potential to be used in the design of dietary supplements (**Stojkovic et al., 2014**). Polysaccharides are extracted from the fruit body, mycelium and fermentation liquid of reishi mushroom , The monosaccharides in the mycelium and spores is mainly glucose, while that from the fruiting bodies are mainly glucose and galactose. Polysaccharides extracted from fruiting bodies have anticancer effects via immunomodulation (**Bishop et al., 2015**). More than 200 triterpenes have been

identified from the fruiting bodies, spores, and mycelia of reishi mushroom (Xia *et al.*, 2014; Baby *et al.*, 2015). According to the unctinal groups, reishi mushroom triterpenoids can be divided into compounds such as ganoderiol, ganoderic acid, ganolactone, ganoderal and ganoderone (Baby *et al.*, 2015). More than twenty kinds of sterols have been found in reishi mushroom, and their skeletons can be divided into cholesterol and ergosterols (Baby *et al.*, 2015).

The polysaccharide composition of reishi mushroom involves several monomers that can contribute significantly to its antitumour, antioxidant and antibacterial properties (Ferreira *et al.*, 2015; Liu *et al.*, 2016).

Modern studies have shown that reishi mushroom triterpenoids and polysaccharides which improve immunity are the main contributors to the traditional pharmacological activities of reishi mushroom (Hapuarachchi *et al.*, 2018). Also, reishi mushroom can ameliorate intestinal infections and induce the secretion of immunoglobulin A (Kubota *et al.*, 2018). Therefore, this research aimed to investigate the effect of reishi mushroom extract on immunity in hepatic rats.

Materials and Methods

Plants

Reishi mushroom was obtained from the Agriculture Research Center, Ministry of Agriculture, Cairo, Egypt. Reishi mushroom was grinded to a powder. 10g of the powder was soaked in 90 ml of ethanol alcohol (80%), shaken for 10 minutes and then allowed to stay at room temperature for 72 hours. The mixture was then filtered using a filter paper and the filtrate evaporated to dryness on water bath at 60°C. The ethanolic extract was kept in air tight bottle in a refrigerator at 4°C until use and served as the stock crude extract.

Proximate chemical composition, sodium chloride content and free sugars composition of reishi mushroom

The carbohydrates, proteins, fat, ash and moisture were evaluated according to (AOAC, 2015). The salt concentration in reishi mushroom was determined according to Osailiet *et al.*, (2014). Free sugars were identified according to Fernandes *et al.*, (2016).

Total tocopherols , polysaccharides , terpenoid and triterpenes contents in reishi mushroom

Tocopherols were determined according to **Fernandes *et al.*, (2016)**. Polysaccharides were evaluated according to **Vazirian *et al.*, (2014)**. Total terpenoids and triterpenes were determined according to **Ghorai *et al.*, (2012)**.

Biological experiments

Rats

Twenty five (25) mature male Albino rats weighting 190 ± 10 g were purchased from Medical Insects Research Institute, Dokki, Cairo, Egypt.

Chemicals

The kits for analysis were obtained from Biodiagnostics Company, Cairo, Egypt . Casein, carbon tetrachloride, vitamin and salt mixtures were purchased from El-Gomhoria Company for Trading Drugs, Chemicals and Medicals Instruments, Cairo, Egypt.

Basal diet

The basal diet prepared according to the following formula as mentioned by **AIN, (1993)** as follow: Protein (10%), corn oil (10%), vitamin mixture (1%), mineral mixture (4%), choline chloride(0.2%), methionine (0.3%), cellulose (5%) and the remained is corn starch (69.5%). The used vitamin mixture component was that recommended by **Campbell, (1963)**, while the salt mixture used was formulated according to **Hegsted *et al.*, (1941)**.

Induction of liver damage in rats

Chronic liver damage was induced in normal healthy rats by subcutaneous injection of CCl_4 (0.2mg/kg body weight) twice a week for 15 days according to **Passmore and Eastwood (1986)**.

Experimental design

Twenty five (25) mature male Albino rats weighting 190 ± 10 g were used in this study. The rats were classified into two main groups, the first group (5 rats) still fed on basal diet and the second main group (20 rats) had given CCl_4 twice a week for 15 days to induce chronic liver damage in rats, then divided into sub groups as follow:

- (2): hepatic group fed on basal diet only as a positive control group.
- (3): hepatic group fed on basal diet and treated with ethanolic reishi mushroom extract(120 mg/kg body weight) orally.
- (4): hepatic group fed on basal diet and treated with ethanolic reishi mushroom extract(240 mg/kg body weight) orally.
- (5): hepatic group fed on basal diet and treated with ethanolic reishi mushroom extract (360mg/kg body weight) orally.

Blood sampling collections

At the end of experiment period , the rats were sacrificed after 12 hours fasting .Samples of blood were taken from the portal vein in dry and clean centrifuge tubes. To have the serum, samples of blood centrifuged for 10 minutes at 3000 r.p.m . Serum was frozen at -20°C for analysis (Malhotra, 2003).

biochemical analysis

The serum levels of glucose was determined according to **Kaplan (1984)**. Alanine amino transferase (ALT), Aspartate amino transferase (AST) and alkaline phosphatase (ALP) were estimated according to the method of **Tietz (1976)**, **Henry, (1974)** and **Moss (1982)**, respectively. Malondialdehyde (MDA), Superoxide dismutase (SOD) and Catalase enzyme (CAT) were assayed according to the method of **Ohkawa et al., (1979)**, **Nishikimi et al., (1972)** and **Aebi (1984)**, respectively. Measurement of white blood cells(WBC), lymphocytes, killing cells , Immunoglobulin M (IgM) and Immunoglobulin G (IgG) were assayed according to **Jacobs et al., (2001)**, **Boyum(1968)**, **woldehiwet and Rowan (1990)**, **Burrels and wells (1977)** and **KaisaGranfors (1979)**, respectively. Tumor necrosis factor (TNF- α) was determined by a sandwich enzyme – linked immunosorbent assay (ELISA).

Statistical analysis of data

The results were written as mean \pm standard deviation by ANOVA test according to **Steel and Torrie (1980)**.

Results and Discussion

Proximate chemical composition, sodium chloride content and free sugars composition of reishi mushroom (g/100g) as dry matter

Data in Table (1) show the proximate chemical composition, sodium chloride content and free sugars composition of reishi mushroom. Carbohydrates were the most abundant compounds, followed by proteins, moisture, fiber, fat, ash and salt. The mean values were 70.56 ± 0.60 , 11.9 ± 0.36 , 6.62 ± 0.1 , 5.8 ± 0.26 , 2.76 ± 0.25 , 2.36 ± 0.15 and 0.36 ± 0.09 (g/100g) for carbohydrates, proteins, moisture, fiber fat, ash, and salt., respectively. Concerning the free sugars, fructose was the only one found in reishi mushroom (2.32 ± 0.095 g/100g).

Table (1): Proximate chemical composition, sodium chloride content and free sugars composition of reishi mushroom (g/100g) as dry matter

Component	Mean+SD
Nutritional value	
Crude protein	11.9+ 0.36
Fat	2.76+ 0.25
Carbohydrate	70.56+ 0.60
Moisture	6.62+ 0.1
Crude fiber	5.8+ 0.26
Total ash	2.36+ 0.15
NaCl	0.365+0.09
Free sugars	
Fructose	2.32+0.09

The results agreed with the ones reported by **Stojkovic et al., (2014)**. Also, **Taofiq et al., (2017)** stated that carbohydrate and crude protein are the most abundant nutrient in reishi mushroom. The proximate analysis of reishi mushroom highlighted the importance of it as an edible nutritionally rich mushroom (**Rakhee et al., 2017**). As a result of this, people can use the reishi mushroom as a good dietary supplement (**Salamat et al., 2017**).

On the other side **Cör et al., (2018)** reported that the protein was the most abundant nutrient in the reishi mushroom (10-40%)

followed by carbohydrates (3-28%), fat and fibre .While **Garuba et al., (2020)** reported that the carbohydrate content in the reishi mushroom was greater than protein .

Total tocopherols, polysaccharides, terpenoid and triterpenes contents in reishi mushroom

Data in Table (2) show total tocopherols, polysaccharides, terpenoid and triterpenes contents in reishi mushroom. Regarding the tocopherols, the delta and alpha isoforms were extracted from the reishi mushroom, being delta - tocopherol the most abundant one (127 ± 1.63 mg/100g). Terpenoids , polysaccharides and triterpenoids were evaluated, being terpenoids the most abundant ones, followed by polysaccharides and triterpenoids. The mean values were 27 ± 1 (mg linalool/g) , 16.43 ± 0.5 (mg starch/g) and 5.76 ± 0.25 (mg ursolic acid/g) for terpenoids, polysaccharides and triterpenoids., respectively.

Table (2): Total tocopherols, polysaccharides, terpenoid and triterpenes contents in reishi mushroom

Component	Mean±SD
Tocopherols (mg /100 g)	
alpha-Tocopherol	18.3 ± 0.95
delta-Tocopherol	127 ± 1.63
Total content of biomolecules	
Polysaccharides (mg starch/g)	16.43± 0.5
Terpenoids (mg linalool/g)	27 ± 1
Triterpenoids (mg ursolic acid/g)	5.76 ± 0.25

The present study is in accordance with **Skalicka-Wozniak et al., (2012)** who reported that reishi mushroom contains high amount of polysaccharide (about 18.45 mg glucose equiv/g dw). Polysaccharides in reishi mushroom can make it a good antitumor, antioxidant and antibacterial (**Taofiq et al., 2017**).

Liu et al., (2017) showed that the amount of triterpene in reishi mushroom was about 6 mg ursolic acid equiv/g dw. Also, **Rakhee et al., (2017)** showed that reishi mushroom is rich in flavonoids, phenolic compounds, steroids, saponins and alkaloids .

Effect of reishi mushroom extract on liver functions of hepatic rats

Table (3) show the effect of reishi mushroom extract on serum liver enzymes including AST, ALT and ALP enzymes of hepatic rats. Data in Table (3) indicate that mean values of AST, ALT and Alp enzymes, in hepatic rats (C+ve) group, were 190.60 ± 2.76 , 49.20 ± 1.90 and 314.66 ± 4.12 (u/l), respectively, while in normal rats it was 140.63 ± 2.12 , 23.86 ± 1.56 and 221.33 ± 3.21 (u/l), respectively, with percent of decrease -26.21%, -51.50% and -29.66%, respectively for normal rats as compared to control positive group. Regarding to AST, results showed that reishi mushroom extract can improve AST level in all treated groups. Concerning to ALT and AIP, results indicated that there were significant decreases in ALT and AIP (u/l) in hepatic rats which fed on reishi mushroom compared to control positive group. Group (5) showed the highest decrease in ALT and AIP as compared to all groups and recorded as the best result which reached -44.71% and -27.35% in ALT and AIP, respectively.

Table (3): Effect of reishi mushroom extract on liver functions of hepatic rats

Groups	ALP (U/L) Mean \pm SD	% of change	ALT (U/L) Mean \pm SD	% of change	AST (U/L) Mean \pm SD	% of change
Group (1): negative Control	221.33 ^c ± 3.21	-29.66%	23.86 ^c ± 1.56	-51.50%	140.63 ^d ± 2.12	-26.21%
Group (2) positive control	314.66 ^a ± 4.12	00	49.20 ^a ± 1.90	00	190.60 ^a ± 2.76	00
Group (3):reishi mushroom 120 mg/kgB.Wt.	288.46 ^b ± 2.91	-8.32%	43.16 ^b ± 1.70	-12.27%	186.66 ^a ± 2.08	-2.06%
Group (3):reishi mushroom 240 mg/kg B.Wt.	253.26 ^c ± 3.18	-19.51%	31.46 ^c ± 1.71	-36.05%	178.00 ^b ± 3.05	-6.61%
Group (3):reishi mushroom 360 mg/kgB.Wt.	228.6 ^d ± 3.24	-27.35%	27.20 ^d ± 2.05	-44.71%	158.66 ^c ± 3.51	-16.75%
LSD	6.095	00	3.26	00	5.024	00

Values are expressed as means \pm SD ; means in the same column with different letter are significantly different ($p \leq 0.05$).

These results are in agreement with those found by **Sudheesh et al., (2012)** who reported that the treatment with reishi mushroom extract (100, 250 mg/kg body weight) can reduce the activities of GPT, GOT, and ALP .The decrease of liver functions markers could be attributed to the high amount of

polysaccharide isolated from reishi mushroom which can also increase hepatic glycogen levels in diabetic rats (Zhu *et al.*, 2016).

Also, Chung *et al.*, (2017) showed that reishi mushroom polysaccharides significantly lowered the liver injury biomarkers, triglyceride and cholesterol in plasma and liver. These results are in the same line with Zhong *et al.*, (2018) who reported that *G. lucidum* polysaccharide peptide (GLPP) can decrease ALT and AST in the non alcoholic fatty liver disease (NAFLD) in mice.

Zhao *et al.*, (2019) reported that treatment with reishi mushroom triterpenoids can increase the level of antioxidant enzymes in the alcoholic liver disease mice and reduce the level of ALT enzyme.

Effect of reishi mushroom extract on WBC(k/ul), LYM% and Killing Cells % of hepatic rats

Data listed in Table (4) show the effect of reishi mushroom extract on WBC, lymphocytic and killing cells of hepatic rats. The serum level of the mentioned previously parameters were 10.36 ± 1.10 k/ul , $6.13 \pm 0.32\%$ and $90.66 \pm 0.77\%$ in negative control group rats while in positive control group the mean values were 5.5 ± 1.32 k/ul, $2.83 \pm 0.28\%$ and $66.03 \pm 2.21\%$ respectively. These findings denote that there were significant increases in serum level of WBC, lymphocytic and killing cells of negative control group as compared to positive control group. Group (5) showed the highest significant improvement in the mentioned previously parameters when compared to other tested groups.

Table (4): Effect of reishi mushroom extract on WBC(k/ul), LYM% and Killing Cells % of hepatic rats

Groups	Killing cell % Mean \pm SD	% of change	LYM% Mean \pm SD	% of change	WBC (k/ul) Mean \pm SD	% of change
Group (1): negative Control	90.66^a ± 0.77	37.30%	6.13^a ± 0.32	116.60%	10.36^a ± 1.10	88.36%
Group (2) positive control	66.03^d ± 2.21	00	2.83^d ± 0.28	00	5.5^b ± 1.32	00
Group (3):reishi mushroom 120 mg/kgB.Wt.	73.13^c ± 1.72	10.75%	3.9^c ± 0.36	37.80%	6.16^b ± 0.76	12 %

Group (3):reishi mushroom 240 mg/kgB.Wt.	79.6 ^b ±1.27	20.55%	4.96 ^b ±0.50	75.26%	7.6 ^b ±0.61	38.18%
Group (3):reishi mushroom 360 mg/kgB.Wt.	88.2 ^a ±2.04	33.57%	6 ^a ±0.10	112.01%	9.53 ^a ±0.91	73.27 %
LSD	3.07	00	0.619	00	1.77	00

Values are expressed as means \pm SD ; means in the same column with different letter are significantly different ($p \leq 0.05$).

Effect of ethanolic reishi mushroom extract on IgG, IgM and TNF - α in hepatic rats

Data recorded in Table (5) show the effect of reishi mushroom extract on IgG, IgM and TNF - α in hepatic rats. Control (+ve) group showed significant decrease in IgG and IgM as compared to healthy rats, which were 827.53 \pm 2.83 (mg/dl) and 72.76 \pm 2.85(mg/dl), respectively for hepatic rats as compared to 865.1 \pm 7.63(mg/dl) and 100.4 \pm 2.58(mg/dl), respectively for normal rats.

All groups treated with reishi mushroom extract orally showed significant increases in IgG and IgM as compared to positive control group. As for TNF- α , results show that reishi mushroom extract can decrease TNF- α level in all treated groups . Group (5) recorded as the best treatment.

Table (5): Effect of ethanolic reishi mushroom extract on IgG, IgM and TNF - α in hepatic rats

Groups	TNF α (ng/l) Mean \pm SD	% of change	IgM (mg/dl) Mean \pm SD	% of change	IgG (mg/dl) Mean \pm SD	% of change
Group (1): negative Control	1.66 ^c ±0.14	-38.97%	100.4 ^c ±2.58	37.98%	865.1 ^a ±7.63	4.54%
Group (2) positive control	2.72 ^a ±0.15	00	72.76 ^e ±2.85	00	827.53 ^c ±2.83	00
Group (3):reishi mushroom 120 mg/kgB.Wt.	2.50 ^a ±0.09	-8.08%	88.6 ^d ±2.91	21.77%	836.23 ^b ±2.79	1.05%
Group (4):reishi mushroom 240 mg/kgB.Wt.	2.13 ^b ±0.15	-21.69%	112.53 ^b ±2.55	54.65%	866.83 ^a ±3.42	4.74%
Group (5):reishi mushroom 360	1.88 ^c ±0.07	-30.88%	122.86 ^a ±2.45	68.85%	872.56 ^a ±2.43	5.44%

mg/kgB.Wt.						
LSD	0.234	00	4.84	00	7.79	00

Values are expressed as means \pm SD ; means in the same column with different letter are significantly different ($p \leq 0.05$).

These findings were similar to those of **Habijan et al., (2015)** who reported that reishi mushroom polysaccharides improve immune response. Also, **Lv et al., (2016)** showed the immunostimulation ability of reishi mushroom polysaccharide, they compared reishi mushroom polysaccharides with other herbs to study the effect of reishi mushroom polysaccharides on immunity. **Huang et al., (2018)** have done a new formulation using reishi mushroom microsporum immunomodulatory protein to do apoptosis in cancer cells. Reishi mushroom can stimulate B-cell proliferation and activation, promotes T cell and enhances immunity, This is likely due to the high content of antioxidants in reishi mushroom (**Wang and Lin 2019**).

Effect of reishi mushroom extract on oxidant and antioxidant parameters in liver tissue of hepatic rats

The obtained results from Table (6) showed the effect of ethanloic reishi mushroom extract on oxidant and anti oxidant parameters in liver tissue of hepatic rats. It is clear from Table (6) that in rats intoxicated with CCl_4 without treatment; the enzymatic antioxidant CAT and SOD were 4.46 ± 0.55 (u/mg tissue) and 1.73 ± 0.47 (u/g tissue), respectively, In normal rats, level of the mentioned previously enzymes were 17.86 ± 0.95 (u/g tissue) and 8.53 ± 0.55 (u/mg tissue), respectively, these finding denote that there were significant decreases of enzymatic antioxidants in control positive group. According to CAT and SOD, there were significant differences between all groups treated with reishi mushroom extract and control positive group. Group (5) showed the highest increase in SOD and CAT as compared to all groups. As for MDA, it could be noticed that there was significant increase in group (2) as compared to group (1) also, there was significant decrease between all groups and control positive group.

Table (6): Effect of reishi mushroom extract on oxidant and antioxidant parameters in liver tissue of hepatic rats

Groups	MDA (nmol/g tissue) Mean \pm SD	% of change	SoD (U/mg tissue) Mean \pm SD	% of change	CAT (U/g tissue) Mean \pm SD	% of change
Group (1): negative Control	61.40 ^e +2.05	-30.38%	8.53 ^a +0.55	393.06%	17.86 ^a +0.95	300.44%
Group (2) positive control	88.20 ^a +1.21	00	1.73 ^e +0.47	00	4.46 ^d +0.55	00
Group (3):reishi mushroom 120 mg/kgB.Wt.	83.20 ^b +1.91	-5.66%	3.13 ^d +0.25	80.92%	7.30 ^c +1.51	63.67%
Group (4):reishi mushroom 240 mg/kgB.Wt.	77.03 ^c +1.72	-12.66%	5.3 ^c +0.53	206.35%	11.93 ^b +1.55	167.48%
Group (5):reishi mushroom 360 mg/kgB.Wt.	69.96 ^d +1.46	-20.68%	7.13 ^b +0.30	312.14%	15.9 ^a +1.87	256.50%
LSD	3.090	00	0.799	00	2.494	00

Values are expressed as means \pm SD ; means in the same column with different letter are significantly different ($p \leq 0.05$).

These results are supported by the results published by **Chiu *et al.*, (2017)** who observed that reishi mushroom can improve total thiols, total antioxidant capacity and glutathione content in plasma and increase the levels of antioxidant enzymes. Reishi mushroom extract has antioxidant activities because of the large amount of total polysaccharides; people can use the extract to protect them against oxidative stress and liver damage (**Chen *et al.*, 2018**).

A similar observation was reported by **Sargowoet *et al.*, (2018)** who showed that the level of SOD increased but the level of MDA reduced on treatment with reishi mushroom. This is likely due to high contents of polysaccharides in reishi mushroom which have strong antioxidant activity (**Wu 2018 ; Zenget al. , 2019**). Also, triterpenoids in reishi mushroom not only can reduce the accumulation of cadmium in the liver of chicken, but also significantly can increase the activities of antioxidants (**Li *et al.*, 2019**).

Effect of reishi mushroom extract on serum glucose level of hepatic rats

Data presented in Table (7) show the effect of reishi mushroom extract on serum glucose level of hepatic rats. These findings denote that there were significant increases in mean value of serum glucose of hepatic rats as compared to normal rats, which were 122.60 ± 2.52 and 81.76 ± 3.14 mg/dl, respectively. All variable treatment (G3, G4 and G5) showed significant decreases compared to control (+) group, which were 110.93 ± 1.98 , 95.46 ± 2.60 and 83.33 ± 1.90 mg/dl, respectively. Group (5) recorded highest significant value when compared to hepatic rats (c+ve), also this group showed nonsignificant differences compared to normal rats.

Table (7): Effect of reishi mushroom extract on serum glucose level of hepatic rats

Groups	Serum glucose level (mg/dl) Mean \pm SD	% of change
Group (1): negative Control	$81.76^d \pm 3.14$	-33.31%
Group (2): positive control	$122.60^a \pm 2.52$	00
Group (3): reishi mushroom 120 mg/kgB.Wt	$110.93^b \pm 1.98$	-9.51%
Group (3): reishi mushroom 240 mg/kgB.Wt	$95.46^c \pm 2.60$	-22.13%
Group (3): reishi mushroom 360 mg/kgB.Wt	$83.33^d \pm 1.90$	-32.03%
LSD	4.500	00

Values are expressed as means \pm SD ; means in the same column with different letter are significantly different ($p \leq 0.05$).

These results are supported by the results published by **Xiao et al., (2017)** who reported that reishi mushroom polysaccharide consumption could provide a beneficial effect in terms of lowering the blood glucose levels by inhibition of gluconeogenesis and promotion of glycogen synthesis. Also, Fudan-Yueyang *G. lucidum* (FYGL), a neutral hyperbranched proteoglycan ingredient extracted from reishi mushroom could reduce blood glucose, decrease body weight, and ameliorate insulin resistance in mice (**Chen et al., 2018; Yang et al., 2018; Yang et al., 2019**).

These results are completely in agreement with **Tong et al., (2018)** who suggested that reishi mushroom can reduce the level of blood glucose in patients with type 2 diabetes mellitus. The mechanism may be due to the regulation of adiponectin and leptin levels. So that reishi mushroom has preventive and therapeutic effect on diabetes (**Liu and Tie (2019)**).

In conclusion: From the present results, it can be concluded that reishi mushroom can contribute significantly to the nutrient requirements of man and should be used as a source of nutrients to supplement other major sources and improve immunity.

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