Protective effects of mushroom and their ethyl extract on aging rats compared with L-carnitine

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Abstract: The effects of dried mushroom (10%) and mushroom extract (300 mg/kg body weight/day) on the lipids profile, lipid peroxidation and liver functions of rats were evaluated and compared with L-carnitine. Food intake, body weight and histological examination of liver tissues were also evaluated. Supplementation diet of rats with 10% dried mushroom, 300 mg mushroom extract and 300 and 600 mg L-carnitine resulted in a significant decrease in total lipids, triglycerides, total cholesterol, low-density lipoprotein, very low-density lipoprotein, aspartate amino transferase (AST) enzyme, alanine amino transferase (ALT) enzyme, alkaline phosphatase (AP) enzyme, malonaldehyde (MAD) and body weight compared to control. However, glutathione peroxidase enzyme (GSH) and food intake were significantly increased in rats supplemented with 10% dried mushroom, 300 mg mushroom extract and 300 and 600 mg L-carnitine. Liver tissues of rats were improved by the supplementation with 10% dried mushroom and 300 mg L-carnitine. However, the supplementation with 300 mg mushroom extract and 600 mg L-carnitine were more effective in improving the liver tissues. These results suggest that mushroom and their extract can be improve the antioxidant status during ageing and minimize the occurrence of age-associated disorders associated with involvement of free radicals. [Nature and Science 2010;8(10):26-33]. (ISSN: 1545-0740).

Key words: Dried mushroom, mushroom extract, L-carnitine, lipids profile, liver function, lipid peroxidation

1. Introduction

Aging is associated with biochemical and structural alterations which are thought to result in motor and cognitive impairments and in increased susceptibility to neurodegenerative diseases (Freo et al., 2002; Terry and Buccafusco, 2003). The free radical theory of aging proposed that aging is due to the accumulation of unrepaired damage from free radical attack on cellular components. Modern thinking theory proposes that aging is caused by a shift in the balance between the prooxidative and anti-oxidative processes in the direction of the prooxidative state (Harman, 1992; Beckman and Ames 1998. and Cadenas and Davies 2000). Mushroom had high amounts of proteins, carbohydrates and fibers with low fat contents (Bárbara et al., 2008). Furthermore, mushroom had significant levels of vitamins, namely thiamine, riboflavin, ascorbic acid and vitamin D₂, as well as minerals (Mattila, et al., 2000). Regarding their medicinal value, mushroom is effective as antitumor, antibacterial, antiviral and hematological agents and in immunomodulating treatments (Wasser and Weis, 1999; Yang, et al., **2002**). Mushroom species had been shown to possess antioxidant capacity in *in-vitro* systems (Ribeiro et al., 2006). Seline and Johein. (2007) reported that the l-carnitine concentration in mushroom ranged from 133 to 530 mg/kg DM (mean 320 mg/kg DM).

The free l-carnitine concentration in mushroom ranged between 73 to 383 mg/kg DM (mean 218 mg/kg DM), which represented $65 \pm 8\%$ of total carnitine content. L-carnitine, a nutrient normally synthesized from methionine and lysine in the liver and kidney. L-carnitine transports long-chain fatty acids (LCFA) across the mitochondrial membrane where they undergo beta-oxidation to produce energy. L-carnitine deficiency decreases LCFA availability oxidation, thereby resulting in LCFA for accumulation in the cytosol, and decreased ketone and energy production. Other L-carnitine functions include the maintenance of adequate free coenzyme-A required for various metabolic pathways, the protection of cells against toxic accumulation of acylcoenzyme-A compounds by shuttling acyl groups out of the mitochondria, and the storage and transport of energy (Catherine et al., 2006). Also, L-carnitine supports the immune system and enhances the antioxidant system (Bremer, 1997). The objectives of this study were to evaluate the effects of dried mushroom and mushroom extract on the lipids profile, lipid peroxidation and liver functions of aging rats as compared with L-carnitine.

2. Material and Methods

The mushroom *P. ostreatus* was purchased from Food Technology Research Institute,

Agricultural Research Center. L-carnitine was purchased from Sigma Chemical Co. (St. Louis, MO, USA). Total cholesterol, HDL-cholesterol, total lipids, alkaline phosphatase (AP), aspartate amino transferase (AST), alanine amino transferase (ALT), glutathione peroxidase (GSH) and malonaldehyde (MDA) kits were obtained from Randox Laboratories Ltd, England.

Preparation of the dried mushroom and mushroom extract:-

The mushroom *P. ostreatus* was dried in the shade and then finely powdered. Dried mushroom were ground to pass through a 60 mesh sieve. Fifty grams of dried mushroom were extracted with 150 ml of 95% ethanol using a Soxhlet apparatus. The residue was filtered and concentrated to a dry mass by vacuum distillation and used as mushroom extract. **Experimental Animal:**

Experimental Animal: Male albino rats of Wister strain weighing approximately 310 ± 6 g were used. A total of thirty Albinos male rats were raised the animal house of Food Technology Research. Agriculture research

Food Technology Research, Agriculture research center. Giza, Egypt. The animals were fed a basal diet for 7 days as an adaptation period. The basal diet was formulated according to AIN (1993) and consisted of casein (12%), corn oil (10%), cellulose (5%), salt mixture (4%), vitamin mixture (1%) and starch (68%); Water was available ad libitum. The animals were divided to five groups, the first group (control) fed the basal diet. The second and the third groups fed basal diet + L-carnitine (300 and 600 mg/kg body weight/day, respectively) in 0.89% saline at physiological pH. The fourth group fed dried mushroom diet (10% of dried mushroom was incorporated into the basal diet at the expense of corn starch content) and the fifth group fed basal diet + mushroom extract (300 mg/kg body weight/day) in distilled water. Blood samples were taken at the start and the end of experiment (4 weeks). The blood samples were obtained from orbital plexus venus by means of fine capillary glass tubes according to the method described by Schermer, (1967). The blood samples were placed in dray and clean centrifuge tubes and allowed to clot for 1-2 h at room temperature. Serum was removed using a Pasteur pipette and centrifuged for 20 min at 1100 x g. The clean supernatant serum was kept frozen until analysis. Body weights of animals were recorded at the start and the end of experiment.

Measurements of biochemical variables:-

The serum triglycerides, high-density lipoprotein (HDL), total cholesterol and total lipids were determined according to the methods described by Fossati and Prencipe (1982), Demacker *et al.*, (1980), Richmound, (1973) and Frings and Dunn (1979), respectively. The determination of low density lipoprotein (LDL) and very low density lipoprotein (VLDL) were carried out according to the methods of **Lee and Nieman (1996)** as follows: VLDL=TG/5,LDL=Total cholesterol (HDL+VLDL).

Alanine amino transferase (ALT), aspartate amino transferase (AST) and alkaline phosphatase (AP) enzymes were measured according to the methods described by **Bergmeyer and Harder** (1986), Kachmar and Moss (1976) and Varley *et al.*, (1980), respectively. Glutathione peroxidase enzyme (GSH) and malonaldehyde (MDA) were determined according to the methods described by Hu (1994) and Jentzsch *et al.*, (1996).

Histopathology examinations:-

Small specimens of the organs liver were taken from each experimental group, fixed in neutral buffered formalin, dehydrated in ascending concentration of ethanol (70, 80 and 90%), cleared in zylene and embedded in paraffin. Sections of 4–6 μ m thickness were prepared and stained with hematoxylin and eosin according to **Bancroft** *et al.*, (1996).

Statistical analysis:-

The results recorded as the mean \pm SD of seven replicates. The experimental data were subjected to an analysis of variance (ANOVA) for a completely randomized design using a statistical analysis system (SAS, 2000). Duncan's (1995) multiple range tests were used to determine the differences among means at the level of 5%.

3. Results and Discussion

Effect of dried mushroom, mushroom extract and L-carnitine on lipids profile of aging rats:-

Data presented in Table (1) showed the effect of 10% dried mushroom, 300 mg mushroom extract and L-carnitine on total lipids, triglycerides and total cholesterol. Total lipids was significantly (P ≤ 0.05) reduced in rats supplemented with mushroom and L-carnitine. The reduction in the total lipids was ranged between 6.85 to 13.79%. There was no significant (P > 0.05) difference in total lipids between rats supplemented with 300 mg L-carnitine and those supplemented with 600 mg L-carnitine had a higher (P ≤ 0.05) total lipids than those supplemented with 300 mg mushroom extract.

Supplementation of rats diet with mushroom and L-carnitine resulted in a significant ($P \le 0.05$) decrease in triglycerides and total cholesterol. Triglycerides was reduced by 30.47- 40.92%. However, total cholesterol was reduced by 16.38-27.0%. Supplementation of rats diet with 300 mg mushroom extract and 600 mg L-carnitine were more ($P \le 0.05$) effective in reducing triglycerides and total cholesterol than those supplemented with 10% dried mushroom and 300 mg L-carnitine. On the other hand, supplementation of rats diet with 300 mg mushroom extract and 600 mg L-carnitine were similar (P > 0.05) in reducing triglycerides and total cholesterol. Supplementation of rats diet with 10% dried mushroom and 300 mg L-carnitine were also similar (P > 0.05) in reducing triglycerides and total cholesterol.

Bobek et al., (1994) found that rats fed a semisynthetic diet containing 0.3% cholesterol and supplemented with 5% dried whole oyster mushroom had reduced serum and liver cholesterol levels by 32 and 55%, respectively. Panchamoorthy and Carani, (2007) reported that treated rats with L-carnitine caused a significant reduced in TG as compared to untreated rats. L-carnitine is known to promote the transport of cytosolic long-chain fatty acids into the mitochondrial matrix for β -oxidation, thereby providing mitochondrial energy (Diaz et al., 2000 and Eskandari et al., 2004). L-carnitine may lower plasma TG by increasing the utilization and/or oxidation of fatty acids for energy or possibly by altering very low-density lipoprotein synthesis (Tanaka et al., 2004).

Effect of dried mushroom, mushroom extract and L-carnitine on lipoprotein of aging rats:-

Data presented in Table (2) showed that the high density lipoprotein in rats was not affected (P >0.05) by the supplementation with 10% dried mushroom and 300 mg L-carnitine. However, rats supplemented with 300 mg mushroom extract and 600 mg L-carnitine had a higher ($P \le 0.05$) value of high density lipoprotein than those of the control. High density lipoprotein was increased in these rats by 23.7- 29.09%. Low density lipoprotein was (P \leq 0.05) reduced in rats supplemented with mushroom and L-carnitine by 29.22-54.39%. Supplementation rats with 300 mg mushroom extract and 600 mg Lcarnitine were more ($P \le 0.05$) effective in reducing low density lipoprotein than those supplemented with 10% dried mushroom and 300 mg L-carnitine. On the other hand, supplementation of rats diet with 300 mg mushroom extract and 600 mg L-carnitine were similar (P > 0.05) in reducing low density lipoprotein. Supplementation of rats diet with 10% dried mushroom and 300 mg L-carnitine were also similar (P > 0.05) in reducing low density lipoprotein.

Very low density lipoprotein in rats was ($P \le 0.05$) reduced by the supplementation with mushroom and L-carnitine. Very low density lipoprotein was reduced in these rats by 30.44-40.09%. Supplementation of rats diet with 300 mg mushroom extract and 600 mg L-carnitine were more ($P \le 0.05$) effective in reducing very low density lipoprotein than those supplemented with 10% dried mushroom and 300 mg L-carnitine. Supplemented with 300 mg mushroom extract and 600 mg L-carnitine did not significantly (P > 0.05) differ in their effect on very low density lipoprotein. Also, no significant (P > 0.05) difference was found in very low density lipoprotein between rats supplemented with 10% dried mushroom and those supplemented with 300 mg L-carnitine.

These results are in agreement with those reported by **Sidereal and Volgin**, (**1996**) and **Lofgren** *et al.*, (**2005**) they found that L-carnitine stabilizes the level of lipids peroxidation, decreases concentration of total lipids, triglycerides, total cholesterol, phospholipids, and lipoproteins of low and very low density.

Effect of dried mushroom, mushroom extract and L-carnitine on liver functions of aging rats:-

Data in Table (3) indicated that the aspartate amino transferase enzyme (AST) in rats was significantly (P \leq 0.05) reduced by the supplementation with mushroom and L-carnitine. Mushroom reduced AST enzyme by 37.78-40.33%. However, L-carnitine reduced it by 23-43.86%. Rats supplemented with 300 mg L-carnitine had a higher (P \leq 0.05) AST enzyme than those supplemented with mushroom and 600 mg L-carnitine. Supplemented with 300 mg mushroom extract and 600 mg Lcarnitine were not significantly (P > 0.05) differed in their effect on AST enzyme.

Supplemented rats with mushroom and Lcarnitine had a lower ($P \le 0.05$) alanine amino transferase enzyme (ALT) than that of the control. Mushroom and L-carnitine reduced ALT enzyme by 35.57-44.88% and 20.82-35.40%, respectively. Supplemented rats with 10% dried mushroom, 300 mg mushroom extract and 600 mg L-carnitine were more effective (P > 0.05) in reducing ALT enzyme than those supplemented with 300 mg L-carnitine. No significant (P > 0.05) difference was found in ALT enzyme among rats supplemented with 10% dried mushroom, 300 mg mushroom extract and those supplemented with 600 mg L-carnitine.

The alkaline phosphatase enzyme (AP) in rats was significantly ($P \le 0.05$) reduced by the supplementation with mushroom and L-carnitine. Mushroom reduced AP enzyme by 19.55-31.4%, however, L-carnitine reduced it by 21.17-48.5%. Supplemented rats with 300 mg L-carnitine had a higher (P \leq 0.05) AP enzyme than those with 600 supplemented mg L-carnitine. Supplemented rats with 10% dried mushroom had a higher (P \leq 0.05) AP enzyme than those supplemented with 300 mg mushroom extract. Supplemented rats with 10% dried mushroom and 300 mg L-carnitine were not significantly (P > 0.05) differed in their effect on AP enzyme.

L-carnitine and mushroom restores the changes of ALT, AST and AP activities due to their antioxidant effects and their ability to act as a radical scavenger, thereby protecting membrane permeability. **Augustyniak and Skrzydlewska** (2009) found that ALT and AST after ethanol intoxication their activity increased by about 80%. Lcarnitine partly prevented these changes. It was manifested by a statistically significant decrease in the activity of ALT and AST, by about 20% in comparison with the ethanol group.

Effect of dried mushroom, mushroom extract and L-carnitine on MDA and GSH in aging rats:-

Data presented in Table (4) show that the MDA was ($P \le 0.05$) reduced by 10.4-33.69% in rats supplemented with mushroom and L-carnitine. Supplementation of rats diet with 300 mg mushroom extract and 600 mg L-carnitine were more ($P \le 0.05$) effective in reducing MDA than those supplemented with 10% dried mushroom and 300 mg L-carnitine. On the other hand, supplementation of rats with 300 mg mushroom extract and 600 mg L-carnitine were similar (P > 0.05)in reducing MDA. Supplementation of rats with 300 mg mushroom extract and 300 mg L-carnitine were also similar (P >0.05) in reducing MDA. Rats supplemented with 10% dried mushroom had higher (P \leq 0.05) MDA than those supplemented with 300 mg L-carnitine.

Augustyniak and Skrzydlewska (2009) found that administration of L-carnitine to rats intoxicated with ethanol significantly protects lipids and proteins against oxidative modifications in the serum and liver. The level of MDA was decreased by about 30% in the blood serum in comparison to the ethanol group. Glutathione is a major, non-protein thiol in living organisms which performs a key role in co-coordinating the innate antioxidant defense mechanisms. It is involved in the maintenance of the normal structure and function of cells, probably by its redox and detoxification reactions (Gueeri, 1995).

The GSH in rats was significantly ($P \le 0.05$) increased by the supplementation with mushroom and L-carnitine. Mushroom increased GSH by 57.23-79.23%. However, L-carnitine increased it by 59.1-128.6%. Supplemented rats with 300 mg L-carnitine and 10% dried mushroom had a lower ($P \le 0.05$) GSH than those supplemented with 600 mg Land 300 mg mushroom extract. carnitine Supplemented rats with 10% dried mushroom and 300 mg L-carnitine were not significantly (P > 0.05) differed in their effect on GSH. Supplementation of rats diet with 600 mg L-carnitine was more ($P \le 0.05$) effective in increasing GSH than those supplemented with 300 mg L-carnitine, 10% dried mushroom and

300 mg mushroom extract.

Augustyniak and Skrzydlewska (2009) found that L-carnitine caused a significant increase in the liver and blood serum GSH level by 25%. An increase in the levels of GSH in aged rats treated with mushroom extract as a source of antioxidant has also been reported by Jayakumar, *et al.*, (2006).

Effect of dried mushroom, mushroom extract and L-carnitine on food intake and body weight of aging rats:-

Table (5) showed the effect of dried mushroom, mushroom extract and L-carnitine on food intake and body weight of rats. Either Lcarnitine or mushroom significantly (P ≤ 0.05) increased food intake and reduced body weight in rats. There was no significant (P > 0.05) difference in food intake between rats supplemented with Lcarnitine and mushroom. Supplementation of rats with L-carnitine was more ($\dot{P} \le 0.05$) effective in reducing body weight than those supplemented with mushroom. Supplemented rats with 300 mg Lcarnitine and 600 mg L-carnitine were not significantly (P > 0.05) differed in their effect on body weight. Similar effect was observed in rats supplemented with 10% dried mushroom and 300 mg mushroom extract. The rationale for carnitine supplementation as a weight-loss agent is based on the assumption that regular oral ingestion of the substance increases its intracellular concentration. This would trigger increased fat oxidation and gradual reduction of the body's fat reserves (Barnett et al., 1994 and Villani et al., 2000).

Histopathological examinations

Figure (1) showed the histological examination of liver tissues of aged rats fed basal diet, The examination of rats liver tissues showed congestion of the central vein and infiltration with chronic inflammatory cells. The changes in the liver tissues of rats fed basal diet and supplemented with 300 mg L-carnitine and 10% dried mushroom are showed slight hydropic degeneration of hepatocytes and vacuolations of some hepatocytes and small focal hepatic necrosis. The examination of liver tissues for rats fed basal diet with supplemented by 600 mg L-carnitine and 300 mg mushroom extract indicated apparent normal hepatocytes. These results agree with those reported by Jayakumar et al., (2006).

From the above results, it could be concluded that mushroom and their extract were comparable to L-carnitine in controlling lipids oxidation. Dried mushroom and their extract can improve the antioxidant status during ageing and minimize the occurrence of age-associated disorders associated with involvement of free radicals.

Table (1): Effect of dried mushroom (DM)	, mushroom extract (ME) and L-carnitine	(LC) on lipid profile of aging
rats		

Ground		Total lipids(mg/dl)			Triglycerides(mg/dl)			Total cholesterol(mg/dl)		
Groups	before	after	% changes	before	After	% changes	before	after	% changes	
Aged rats control	482.64±9.34	475.05±4.88 ^a	-1.57	197.6±10.16	165.32±13.18 ^a	-16.33	165.28±4.68	158.74±4.37 ^a	-3.95	
Aged rats with 300 mg LC	480.23±4.98	443.11±6.96 ^b	-8.45	202.5±8.71	140.78±3.93 ^b	-30.47	161.51±3.81	135.05 ^b ±4.67 ^b	-16.38	
Aged rats with 600 mg LC	479.65±5.57	425.73±6.25°	-11.24	195.85±9.64	121.55±5.25°	-37.93	163.25±3.69	119.16±3.82°	-27.00	
Aged rats with 10% DM	475.60±8.23	442.95±6.4 ^b	-6.85	205.30±11.21	135.46±4.44 ^b	-34.0	162.60±4.02	131.47±5.77 ^b	-19.13	
Aged rats with 300mg ME	480.05±7.54	413.82±2.39 ^d	-13.79	202.70±6.34	119.73±1.47°	-40.92	160.52±4.53	120.00±3.65°	-25.24	
LSD	10.16	7.87	-	11.34	12.12	-	5.34	7.76	-	

Values are means \pm SD of 6 rats from each group.

Means in the same column with different superscripts are significantly different (P≤0.05)

Table (2): Effect of dried mushroom (DM), mushroom extract (ME) and L-carnitine (LC) on lipoprotein of aging rats.

	High Den	High Density Lipoproteins(mg/dl)			Low Density Lipoproteins(mg/dl)			Very Low Density Lipoproteins(mg/dl)		
Groups	before	after	% changes	before	After	% changes	before	after	% changes	
Aged rats control	49.18±2.5 6	53.31±4.07°	+8.40	76.58±4. 83	72.37±3.0 3 ^a	-5.49	39.52±2. 54	33.06±2.6 3ª	-16.34	
Aged rats with 300 mg LC	48.36±3.1 4	55.46±5.44 ^{bc}	+14.68	72.65±3. 59	51.42±0.3 7 ^b	-29.22	40.50±1. 93	28.17±0.7 8 ^b	-30.44	
Aged rats with 600 mg LC	49.00±3.5 1	60.61±3.04 ^{ab}	+23.70	75.08±4. 69	34.24±0.1 3 ^c	-54.39	39.17±2. 31	24.31±1.0 5 ^c	-37.93	
Aged rats with 10% DM	47.65±2.7 6	52.73±3.61°	+10.66	73.89±4. 72	51.65±3.1 0 ^b	-30.09	41.06±1. 49	27.09±0.8 8 ^b	-34.02	
Aged rats with 300mg ME	48.53±3.8 5	62.65 ± 2.97^{a}	+29.09	71.45±5. 88	33.40±1.1 8°	-53.25	40.54±1. 67	23.95±1.5 5°	-40.09	
LSD	4.06	6.92		6.28	3.69		2.81	2.55		

Values are means \pm SD of 6 rats from each group.

Means in the same column with different superscripts are significantly different (P≤0.05)

Table (3): Effect of dried mushroom	(DM), mushroom e	extract (ME) and	L-carnitine (LC)	on liver functions of aging rats.

		AST(u/l)			ALT(u/l)			AP(u/l)	
Groups	Before	after	% changes	before	after	% changes	before	after	% change s
Aged rats control	82.65±2.15	76.85±2.47 ^a	-7.02	42.67±1.26	40.21±3.45 ^a	-5.76	62.01±2.35	$62.42{\pm}2.46^{a}$	+0.66
Aged rats with 300 mg LC	81.26±3.46	62.57±3.71 ^b	-23.00	43.52±1.99	34.46±3.31 ^b	-20.82	61.53±1.98	48.50±4.95 ^b	-21.17
Aged rats with 600 mg LC	80.39±3.19	45.13±1.63 ^d	-43.86	41.75±2.39	26.97±1.59°	-35.40	62.37±3.05	32.12±2.14 ^d	-48.5
Aged rats with 10% DM	83.07±2.57	51.69±2.73°	-37.78	41.98±2.96	27.05±1.28°	-35.57	63.40±2.98	51.00±2.82 ^b	-19.55
Aged rats with 300mg ME	82.45±2.31	49.16±2.95 ^{cd}	-40.33	42.16±3.07	23.24±2.07°	-44.88	60.90±3.44	41.72±1.30°	-31.4
LSD	4.16	5.30		3.54	4.26		3.92	4.89	

Values are means \pm SD of 6 rats from each group.

Means in the same column with different superscripts are significantly different (P≤0.05)

Table (4): Effect of dried mushroom (DM), mushroom extract (ME) and L-carnitine (LC) on malonaldehyde (MAD)and glutathione peroxidase enzyme (GSH) of aging rats.

Groups		MDA (n mol/ml)		GSH (U/ml)			
Groups	before	after	% changes	before	after	% changes	
Aged rats control	24.87±2.15	25.32±1.47 ^a	+1.80	11.0±3.56	11.51 ± 2.70^{d}	+1.64	
Aged rats with 300 mg LC	26.37±2.11	19.01±0.53°	-27.9	13.21±3.41	21.86±2.68°	+59.1	
Aged rats with 600 mg LC	25.61±1.68	16.98±1.39 ^d	-33.69	12.14±2.94	31.49±2.97 ^a	+128.6	
Aged rats with 10% DM	23.98±2.52	21.48±1.55 ^b	-10.4	10.28±4.62	19.61±1.67 ^c	+57.23	
Aged rats with 300mg ME	24.45±1.46	18.15±0.90 ^{cd}	-25.7	14.05±4.09	26.03±2.52 ^b	+79.23	
LSD	2.65	2.05		4.76	4.44		

Values are means \pm SD of 3 rats from each group.

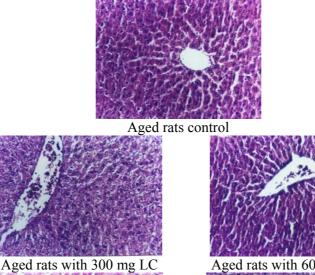
Means in the same column with different superscripts are significantly different ($P \le 0.05$)

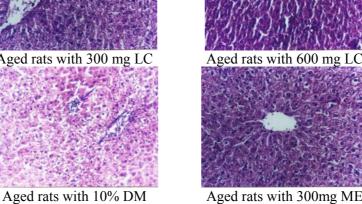
Groups	Food intake(g)	Body weight(g)			
Gloups	rood make(g)	Initial	final		
Aged rats control	9.3 ± 1.14^{b}	312.5 ± 9.84	328.4 ± 8.59^a		
Aged rats with 300 mg LC	17.9 ± 2.13^{a}	313.7 ± 8.37	$275.4 \pm 8.14^{\circ}$		
Aged rats with 600 mg LC	18.1 ± 1.98^{a}	310.8 ± 9.15	$267.9 \pm 7.62^{\circ}$		
Aged rats with 10% DM	17.3 ± 1.85^{a}	312.7 ± 9.46	316.3 ± 8.04^{b}		
Aged rats with 300mg ME	17.5 ± 1.90^{a}	316.9 ± 8.53	312.5 ± 8.16^{b}		
LSD	2.35	7.54	8.95		

Table (5) Effect of dried mushroom (DM), mushroom extract (ME) and L-carnitine (LC) on food intake and body weight of aging rats

Values are means \pm SD of 6 rats from each group.

Means in the same column with different superscripts are significantly different ($P \le 0.05$)





Aged rats with 300mg ME

Fig (1). Effect of dried mushroom (DM), mushroom extract (ME) and L-carnitine (LC) on histological examination of liver tissues of aging rats.

References

- AIN (1993). American Institute of Nutrition (AIN), Purified diet for laboratory rodent. Final Report. ADHOC Writing Diet. J. Nutr., 123: 1939 - 1951.
- Augustyniak, A. and Skrzydlewska, E. (2009). L-Carnitine in the lipid and protein protection against ethanol-induced oxidative stress. Alcohol 43, 217-223.
- Bancroft, D.; Steven, A. and Turner, R. (1996). Theory and Practice of Histological

Techniques, 4th Churchill Livingstone, Edinburgh, London, Melbourne.

- Bárbara, R.; Rosário, L.; Paula, B.; Rosa, M.; Rui, F.; Paula, B. Inès, Q. and Patrícia, V. (2008). Comparative study of phytochemicals and antioxidant potential of wild edible mushroom caps and stipes. Food Chemistry 110, 47-56.
- Barnett, C.; Costill, D. L. and Vukovich, M. D. (1994). Effect of L-carnitine supplementation on muscle and blood

carnitine content and lactate accumulation during highintensity sprint cycling. Int J. Sport Nutr. 4:280-286.

- Beckman, B.; and Ames B. N. (1998). The free radical theory of aging matures. Physiol Rev. 78:547–81.
- Bergmeyer, H. U. and Harder M (1986). A colorimetric method of the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminase Clin. Biochem. 24: 481-486.
- Bobek, P., Ozdín, L. and Kuniak, L., (1994). Mechanism of hypocholesterolemic effect of oyster mushroom (*Pleurotus ostreatus*) in rats: reduction of cholesterol absorption and increase of plasma cholesterol removal. Z. Ernährungswiss. 33, 44–50.
- Bremer, J., (1997). The role of carnitine in cell metabolism. In: De Simone, C., Famularo, G. (Eds.), Molecular Biology Intelligence Unit Carnitine Today (Landes Bioscience Austin, TX, USA, International Copyright). Sringer Verlag, Heidelberg, Germany, pp. 4-37.
- Cadenas, E.; and Davies, K. J. (2000). Mitochondrial free radical generation, oxidative stress and aging. Free Radical Biol. Med. 29:222–230.
- Catherine M. C.; Michael C. S.; Michael L. C.; Charles T. H.; M. Bruce J. and Richard Helms. (2006). A. Carnitine supplementation in premature neonates: Effect on plasma and red blood cell total carnitine concentrations, nutrition parameters and morbidity. Clinical Nutrition. 25, 886-896
- Demacker, P. M.; Von-Janssen, H. E.; Hifman, A. M.; Vant's Lear, A. and Jansen, A. P. (1980). Measurement of high density lipoprotein cholesterol in serum. Comparison of six isolation methods combined with enzymatic cholesterol analysis. Clin. Chem. 26: 1780-1789.
- Diaz, M.; Lopez, F.; Hernandez, F. and Urbina, J. A. (2000). L-carnitine effects on chemical composition of plasma lipoproteins of rabbits fed with normal and high cholesterol diets, Lipids 35 627–632.
- **Duncan, D. B. (1955).** Multiple range and multiple F test Biometrics 11: 1-42.
- Eskandari, H. G.; Burak Cimen, M.Y.; Lulufer Tamer; Arzu Kanik and Ugur Atik. (2004). Short term effects of l-carnitine on serum lipids in STZ-induced diabetic rats Diabetes. Research and Clinical Practice 66 129–132.
- Fossati, P. and Prencipe, I. (1982). Serum

triglycerides determination colorimetrically with an enzyme that produce hydrogen peroxide. Clin. Chem. 28: 2077-2083.

- Freo, U.; Pizzolato, G.; Dam, M.; Ori, C. and Battistin, L. (2002). A short review of cognitive and functional neuroimaging studies of cholinergic drugs: implications for therapeutic potentials. J. Neural Transm. 109, 857–870.
- Frings, C. S. and Dunn RT (1979). Colorimetric method for determination total serum lipids based on the sulphopospho vanillin reaction. Am. J. Clin. Pathol. 53: 89-91.
- Gueeri, H.,(1995). Influence on prolonged ethanol intake on the level and turnover of alcohol and aldehyde dehydrogenase and glutathione. Adv. Exp. Med. Biol. 23, 133– 134.
- Harman, D. (1992). Free radical theory of aging. Mutation Res. 275: 257–66.
- Hu, M. L. (1994). Measurement of protein thiol groups and glutathione in plasma. Methods Enzymol. 233: 380-385.
- Jayakumar, T.; Ramesh, E. and Geraldine, P. (2006). Antioxidant activity of the oyster mushroom, Pleurotus ostreatus, on CCl4induced liver injury in rats. Food and Chemical Toxicology 44: 1989–1996.
- Jentezch, A. M.; Bachmann, H.; Furst, P. and Biesalski H. K. (1996). Improved analysis of malonaldehyde in human body fluids. Free Radic. Biol. Med. 20: 251-260.
- Kachmar, J. F. and Moss D. W. (1976). Enzymes, In: Fundamentals of Clinical Chemistry (edited by Tiez N). pp. 666-672, Philadelphia PA. W.B. Saunders Co.
- Lee. R. and Nieman, D. (1996):Nutrilional. Assessment. 2 nd, Mosby, Missouri, USA
- Lofgren, I.; Zern, T. Herron, K.; West, K.; Sharman, M. Volek, J.; Shachter, N.; and Fernandez, M. (2005). Weight loss associated with reduced intake of carbohydrate reduces the atherogenicity of LDL in premenopausal women. Metabolism. 54(9):1133–41.
- Mattila, P.; Suonpaa, K. and Piironen, V. (2000). Functional properties of edible mushrom. Nutrition, 16, 694–696.
- Panchamoorthy, R.; and Carani, V. (2007). Fructose-induced hepatic gluconeogenesis: Effect of L-carnitine. Life Sciences 80 (2007) 1176–1183.
- Ribeiro, B.; Rangel, J.; Valentáo, P.; Baptista, P.; Seabra, R.; and Andrade, P. (2006). Contents of carboxylic acids and two phenolics and antioxidant activity of dried portuguese wild edible mushrooms. Journal

of Agricultural and Food Chemistry, 54, 8530-8537.

- Richmound, W. (1973). Preparation and properties of cholesterol oxidase from *Nacardia* sp. and its application to enzymatic assay of total cholesterol in serum. Clin. Chem. 19: 1350.
- SAS (2000): Statistics analysis system. SAS Users Guide: Statistics Version 5th Ed., SAS. Institute Inc., Cary N.C.
- Schermer, S. (1967). The Blood Morphology of Laboratry Animals. p. 359. Philadelphia: F. A. Davies Co.
- Seline, K. G. and Johein, H. (2007). The determination of L-carnitine in several food samples. Food Chem., 105: 793 804.
- Sidereal, N. G. and Volgin, D. V. (1996). Effect of L-carnitine on lipid peroxidation and lipid composition in blood serum in hemic hypoxia. Ukr Biokhim Zh ;68(5):54–8.
- Tanaka, Y.; Sasaki, R.; Fukui, F.; Waki, H.; Kawabata, T. and Okazaki, M. (2004). Acetyl-l-carnitine supplementation restores decreased tissue carnitine levels and impaired lipid metabolism in aged rats. J. Lipid Res. 45:729–35.

- **Terry J.; and Buccafusco, J. J.(2003).** The cholinergic hypothesis of age and Azheimer's disease-related cognitive deficits: recent challenges and their implications for novel drug development. J. Pharmacol. Exp. Ther. 306, 821–827.
- Varley, H.; Gewenlock, A. and Bell, M. (1980). Practical clinical biochemistry, Vol. 1, 5th ed. Pp.741:897. London; Williams Heinemen Medical books, Ltd
- Villani, R. G, Gannon, J, Self, M.; and Rich, P. A.(2000). L-carnitine supplementation combined with aerobic training does not promote weight loss in moderately obese women. Int J Sport Nutr Exerc Metab. 10: 199-206.
- Wasser, S. P. and Weis, A. L. (1999). Medicinal properties of substances occurring in higher Basidiomycetes mushrooms: Current perspective (review). International Journal of Medicinal Mushrooms, 1, 31–62.
- Yang, J. H.; Lin, H. C.; and Mau, J. L. (2002). Antioxidant properties of several commercial mushrooms. Food Chemistry, 77, 229–235.