

The Antioxidant Effects of Mushroom Extracts in Boosting Immune System in Mammals using Albino Rats as a Model

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Abstract

Mushrooms have been widely used as medicine in the treatment of several infections and also boosting the immune system. The present study was carried out to ascertain the haematological and the antioxidants properties of aqueous extracts of *Agaricus bisporus* and *Pleurotus tuber-regium*. Twenty five albino rats were grouped into five, each group consisting of five rats of A to E. Group A serves as the control, group B to D were fed with *Pleurotus tuber-regium* 400mg, 600mg and 1000mg concentration respectively while group E was fed with 400mg of *Agaricus bisporus*. On completion of the administration of extracts, the haematological profiles and antioxidant parameters were analysed. The experimental rats showed some little significant increase in both the haematological profile and biomarkers properties with $P < 0.05$. Generally the haematological profiles implies that there was no significant decrease in the level of the experimental rats immunity and also some organs such as the liver and the kidney were intact. SOD, CAT, GSH and MDA are antioxidant enzymes measured to detect toxic consequences of oxidative stress in mammalian systems. The SOD values in rats treated with 400mg/kg/lb of SOD is 113.58mm/mg/protein and rats treated with 1000mg/kg in *Pleurotus tuber-regium*, the values of MDA is 23.32 ± 2.09 (nmol/ml). There was a significant difference in the liver homogenated and kidney homogenated biomarkers in the rats treated with *Pleurotus tuber-regium* and *Agaricus bisporus*. They are cellular and enzymatic defenses against oxidative stress. Oxidative stress causes toxic and adaptive responses within a cell. The importance of an antioxidant defenses in protecting cells and organisms from oxidative damage and toxicity. Further research with higher dosage of the extracts may be required to test on laboratory rats before providing the true haematological and antioxidant properties.

Keywords: Mushroom, Antioxidants and Albino rats

INTRODUCTION

Mushrooms are macro fungi with distinctive fruiting and they are made up of hyphae which forms interwoven web of tissues known as mycelium in the substrates upon which the fungi feed. Most often, their mycelium are buried in the soil around the root of trees beneath leaf litters in the tissues of a tree trunk, on a fallen log of wood or in their nourishing substrates (Ingold, 1993). Mushrooms can be hypogeous or epigeous, large enough to be seen with naked eyes and can be picked up by hands (Chang and Miles, 1992).

Mushrooms are of great economic importance to man; their occurrence is dated back to the time of early man as mushroom appears in traditional Yoruba art works known as (tie and die) which are materials of traditional costumes (Adenle, 1985). They have long been used a valuable food source and as traditional medicine around the world. Many genera of mushrooms are edible and are rich in essential nutrients such as carbohydrates, protein, vitamins, minerals, fats, fibres and various acids (Okwlehie and Odunze, 2004). Records of health promoting properties such as antioxidants, anti-microbial, anti-cancer, cholesterol lowering and immune-stimulatory effect have been reported for some species of mushrooms (Mau *et al.*, 2004). However it must be emphasized that some of these mushroom species are poisonous and may claim lives within few hours after consumption (Philips, 1985). Mushrooms have become attractive as a functional food and as a source for the development of drugs and nutraceuticals (Lakhanpal and Rana, 2005), responsible with their antioxidants, antitumor (Jones and Jonardhanan, 2000) and antimicrobial properties. Mushrooms are becoming more important in our diet due to their nutritional value, related to high protein and low fats/energy contents (Agahar-murugkar and Subbulakshmi, 2005).

Considering mushrooms growth requirements, they grow well on a wide range of lignocellulosic wastes as substrates, has been established that they grow and fruit on various agricultural wastes (Moncalo *et al.*, 2005). Furthermore, some of these mushrooms have been cultivated in the laboratory (Kadiri, 1994; Fasidi, 1995). The substrates could be used in commercial production of mushrooms for food (Fasidi, 1995).

Immunity refers to the protection against infection. The immune system is a collection of cells, tissues and molecules that function to defend our body against infectious microbes. The coordinated reaction of the immune system against infection and other foreign substances is known as immune response. Abnormalities of the immune system that result in defective immune response makes individual susceptible to infection by virus, bacteria, fungi and parasites. The antimicrobial defence function of the immune system is essential for our ability to survive in an environment that is teeming with potentially deadly microbes. However, immune responses are

also capable of causing diseases or damages. Many diseases are caused by uncontrolled and excessive responses (examples include rheumatic fever, asthma, glomerulonephritis).

The immune system of human has the essential function of protecting the body against the damaging effect of microbial agents which is pathogenic. The system comprises of innate immunity (nonspecific), and acquired immunity (specific). Natural killer (NK) cells, complement system, macrophages, antigen presenting cells (APCs) and neutrophils makes up the innate immune response system and mount an immediate nonspecific response to foreign microbial agents. Apart from the natural mechanisms there are additional factors that stimulate and support host immunity. Immunostimulants enhances the overall immunity of the host, and present a nonspecific response against the microbial. They also work to heighten humoral and cellular immune responses by either enhancing cytokines secretion or by directly stimulating B-lymphocyte or T-lymphocyte (Benny and Vanitha, 2004).

Antioxidants have been widely used as food additives to provide protection against oxidative degradation of food by free radicals. Oxidation is essential to many living for the production of energy to fuel biological processes. However, the uncontrolled production of oxygen derived from free radicals is involve in the onset of many diseases such as cancer, rheumatoid arthritis and atherosclerosis as well as in degenerative processes associated with aging. Almost all organisms are well protected from free radicals damage by enzyme such as superoxide dismutase and catalase or compounds such as ascorbic acid, tocophenols and glutathione (Mau *et al.*, 2004).

MATERIALS AND METHODS

Procurement of mushrooms and preparation of aqueous extract

The mushroom (*Agaricus bisporus*) was obtained from Ikorodu, area and also Oyster mushroom (*Pleurotus tuber-regium*) was obtained from Ifo area of Ogun state. They were cut into smaller pieces and shade dried separately for few days and extraction was carried out with a sterile distilled water in a soxhlet apparatus, the residue was filtered and concentrated to dry mass which was weighed and used to prepare the require concentration (400mg, 600mg, and 1000mg).

Experimental design

Male albino rats of Wister strain weighing approximately 310 ± 6 kg were used. A total of thirty Albino male rats were raised in the animal house of Biological science, Yaba College of Technology, Yaba, Lagos. The animals were fed with rat pellets and water was available, *adlibitum*.

The animals were divided into five groups (A, B, C, D, and E). The concentration of the mushroom extract ranges from 400mg/kg bw, 600mg/kg bw, 800mg/kg bw and 1000mg/kg bw. Group A serve as the control while Group E were fed with 400ml/kg bw of *Agaricus bisporus*. The Extract administration were carried out orally using cannulating tube. The rat were sacrificed by the method of cervical dislocation

Blood samples were obtained from orbital plexus venus by means of fine capillary glass tubes in accordance to the method described by Schermer,(1997). The blood samples were placed in a dry and clean centrifuge tubes and 20 μ l liter of anticouagulant was added and mixed properly. Serum was removed using a Pasteur pipette and centrifuged for twenty minutes at 1100xg. The clean supernatant serum was kept frozen until analysed. Body weights of the animals were recorded at the start and the end.

Haematological parameters

Three rats from each of the treatment groups were sacrificed at the end of the 4th week. The animals were anaesthetized with chloroform for 30 seconds. The jugular vein was cut open and blood collected with syringes into bottles containing EDTA and into heparinized bottles. The blood samples were spurned in the centrifuge at 3.000rpm and the haematological indices examined include Red Blood Count (RBC), White Blood Cell (WBC), Packed Cell Volume (PCV), leucocytes, differential count (monocyte, lymphocytes etc.) and haemoglobin concentration (Hb).

Packed Cell Volume (PCV) Determination

This was determined by spinning about 751 of each blood samples in heparinized capillary tube I a centrifuge for 5 minutes.

Erythrocyte (RBC) and Leucocytes (WBC) Counts

Erythrocyte and Leucocytes Counts were determined using Neubaur chamber method as described by (Lamb, 1981). The blood sample collected in each treatment was diluted at a ratio of 1:200 for RBC counter using red cell diluting fluid while a dilution ratio of 1:20 (blood: white cell diluting fluid) was used for WBC count. Samples of RBC WBC count were obtained using the relationship:

$RBC/\mu L = \text{Number of red blood cells counted} \times 5 \times 10^2 \times 200$

WBC/ μ =Number of white blood cells counted x 0.25 x 10x 20

Haemoglobin (Hb) Estimation

Haemoglobin was estimated using cyanomethaemoglobin method. 0.02ml of blood was expelled into 4ml solution. The mixture was allowed to stand for 5 minutes for fully colour development. Sample haemoglobin concentration: 0Sample haemoglobin (g/100ml) = Reading of test x standard haemoglobin concentration.

Biomarkers Test

Biomarkers activities was tested for superoxide dismutase (SOD), Glutathione peroxide, lipid peroxidative and Catalase activity in accordance with zigma procedure of (1978) were carried out in liver and kidney.

Histological Examinations

Small specimens of the organs of liver and kidney 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

Data were analysed as mean \pm standard deviation. Student's "t" test was used to compare the differences between control and experimental groups. Excel software were used to determine the "t" and probability (P) values. P < 0.05 was considered as statistically significant.

RESULTS AND DISCUSSION

TABLE 1: HAEMOGLOBIN CONCENTRATION (g/dl) WITH MEAN \pm SD OF RATS

GROUPS	Concentration	MEAN HB \pm SD	t-VALUE	P-VALUE
A	Control	13.6 \pm 1.03	26.363	.000
B	<i>Pleurotus tuber-regium</i> 400mg	11.4 \pm 0.14	114.000	.006
C	<i>Pleurotus tuber-regium</i> 600mg	9.9 \pm 2.98	5.766	.029
D	<i>Pleurotus tuber-regium</i> 1000mg	12.7 \pm 0.77	33.157	.000
E	<i>Agaricus bisporous</i> 400mg	11.1 \pm 3.35	7.385	.002

Table 2: PACK CELL VOLUME (%) WITH MEAN \pm SD OF RATS

GROUPS	Concentration	MEAN PCV % \pm SD	t-VALUE	P-VALUE
A	Control	41.0 \pm 3.27	25.107	.000
B	<i>Pleurotus tuber-regium</i> 400mg	34.5 \pm 0.707	69.000	.009
C	<i>Pleurotus tuber-regium</i> 600mg	30.7 \pm 9.29	5.717	.029
D	<i>Pleurotus tuber-regium</i> 1000mg	38.5 \pm 2.38	32.346	.000
E	<i>Agaricus bisporous</i> 400mg	33.8 \pm 10.56	7.151	.002

Table 3: WHITE BLOOD CELL COUNT (mm³) WITH MEAN \pm SD OF RATS

GROUPS	Concentration	MEAN WBC % \pm SD	t-VALUE	P-VALUE
A	Control	8600.0 \pm 1622.24	10.603	.002
B	<i>Pleurotus tuber-regium</i> 400mg	2625.0 \pm 742.46	5.000	.126
C	<i>Pleurotus tuber-regium</i> 600mg	7000.0 \pm 4222.26	2.872	.103
D	<i>Pleurotus tuber-regium</i> 1000mg	9537.5 \pm 1949.09	9.787	.002
E	<i>Agaricus bisporous</i> 400mg	6380.0 \pm 2477.55	5.758	.005

Table 4: RED BLOOD CELL COUNT (10⁶/ml) WITH MEAN \pm SD OF RATS

GROUPS	Concentration	MEAN RBC \pm SD	t-VALUE	P-VALUE
A	Control	3.5 \pm 0.13	53.447	.000
B	<i>Pleurotus tuber-regium</i> 400mg	3.9 \pm 1.41	3.900	.160
C	<i>Pleurotus tuber-regium</i> 600mg	3.9 \pm 1.63	4.190	.053
D	<i>Pleurotus tuber-regium</i> 1000mg	4.9 \pm 1.36	7.230	.005
E	<i>Agaricus bisporous</i> 400mg	3.1 \pm 0.81	8.532	.001

Table 5: MEAN CELL VOLUME (fl) WITH MEAN±SD OF RATS

GROUPS	Concentration	MEAN MCV ±SD	t-VALUE	P-VALUE
A	Control	118.9 ± 8.99	26.448	.000
B	<i>Pleurotus tuber-regium</i> 400mg	94.3 ± 32.39	4.118	.152
C	<i>Pleurotus tuber-regium</i> 600mg	81.6 ± 13.96	10.120	.010
D	<i>Pleurotus tuber-regium</i> 1000mg	84.2 ± 30.39	5.538	.012
E	<i>Agaricus bisporous</i> 400mg	112.7 ± 33.72	7.472	.002

Table 6: MEAN CELL HAEMOGLOBIN (pg) WITH MEAN±SD OF RATS

GROUPS	Concentration	MEAN MCH ±SD	t-VALUE	P-VALUE
A	Control	39.2 ± 2.82	27.847	.000
B	<i>Pleurotus tuber-regium</i> 400mg	31.1 ± 10.89	4.039	.155
C	<i>Pleurotus tuber-regium</i> 600mg	26.4 ± 4.55	10.057	.010
D	<i>Pleurotus tuber-regium</i> 1000mg	27.8 ± 9.84	5.657	.011
E	<i>Agaricus bisporous</i> 400mg	36.9 ± 10.76	7.674	.002

Table 7: MEAN CELL HAEMOGLOBIN (g/dl) CONCENTRATION WITH MEAN±SD OF RATS

GROUPS	Concentration	MEAN MCHC ±SD	t-VALUE	P-VALUE
A	Control	32.9 ± 0.28	239.489	.000
B	<i>Pleurotus tuber-regium</i> 400mg	32.9 ± 0.21	219.667	.003
C	<i>Pleurotus tuber-regium</i> 600mg	32.4 ± 0.06	973.000	.000
D	<i>Pleurotus tuber-regium</i> 1000mg	33.1 ± 0.25	262.655	.000
E	<i>Agaricus bisporous</i> 400mg	32.8 ± 0.36	205.250	.000

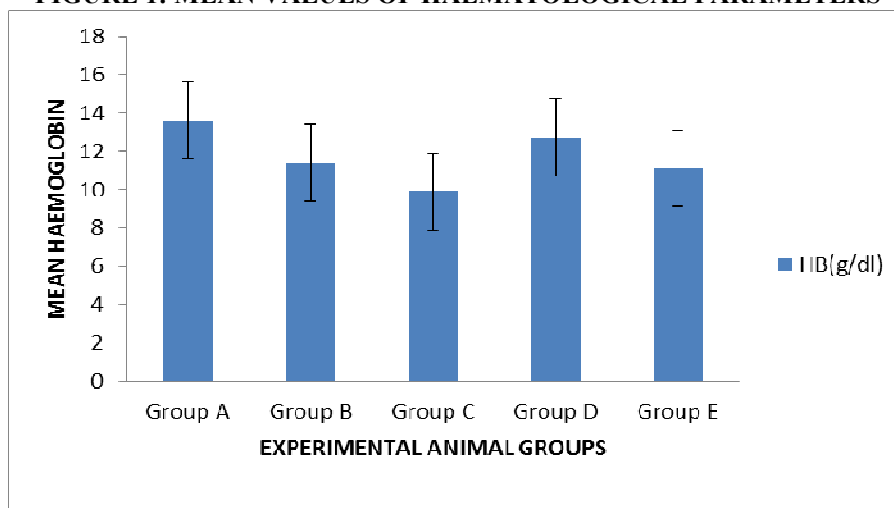
Table 8: NEUTROCYTE COUNT (%) WITH MEAN±SD OF RATS

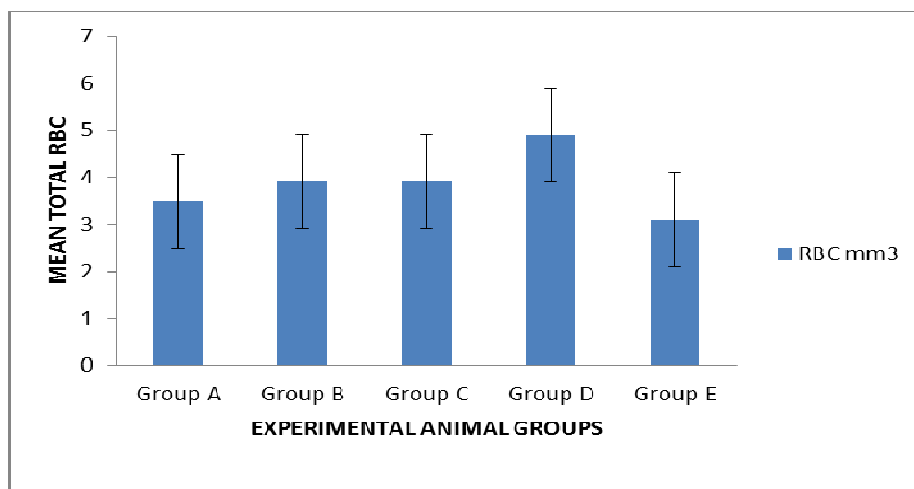
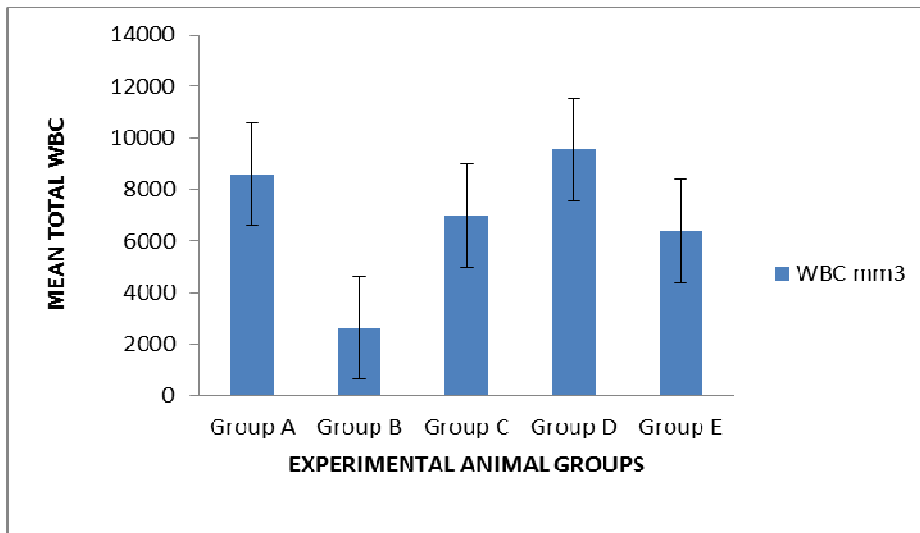
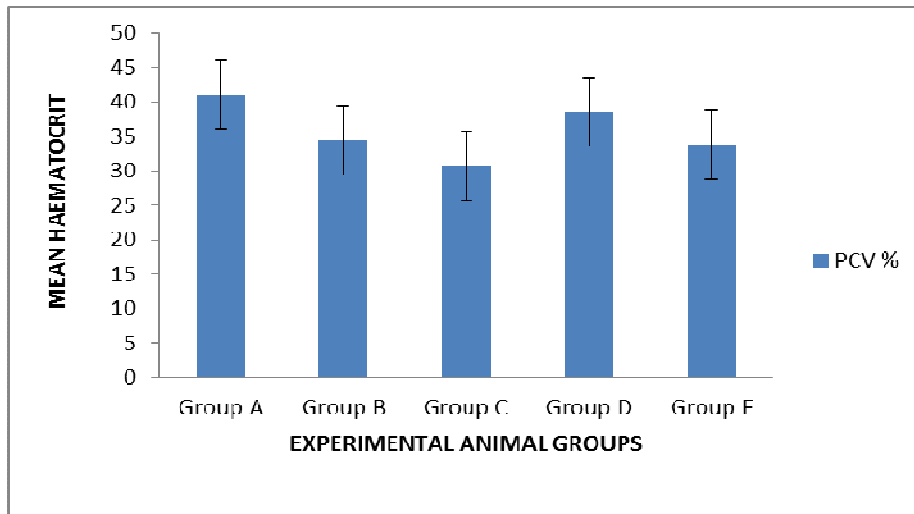
GROUPS	Concentration	MEAN NEUT % ±SD	t-VALUE	P-VALUE
A	Control	70.0 ± 6.68	20.948	.000
B	<i>Pleurotus tuber-regium</i> 400mg	50.3 ± 43.59	2.000	.184
C	<i>Pleurotus tuber-regium</i> 600mg	75.7 ± 3.21	40.770	.001
D	<i>Pleurotus tuber-regium</i> 1000mg	77.0 ± 2.58	59.644	.000
E	<i>Agaricus bisporous</i> 400mg	80.3 ± 2.42	81.241	.000

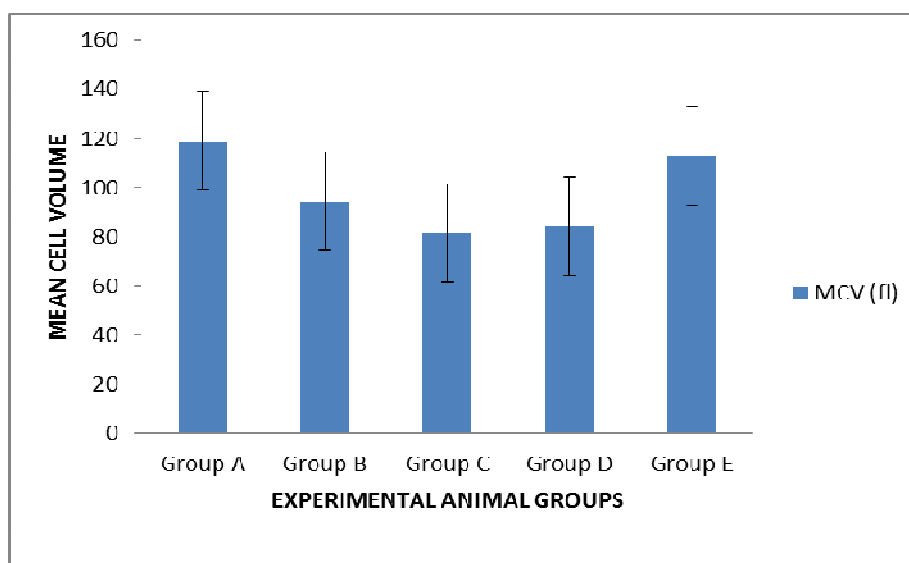
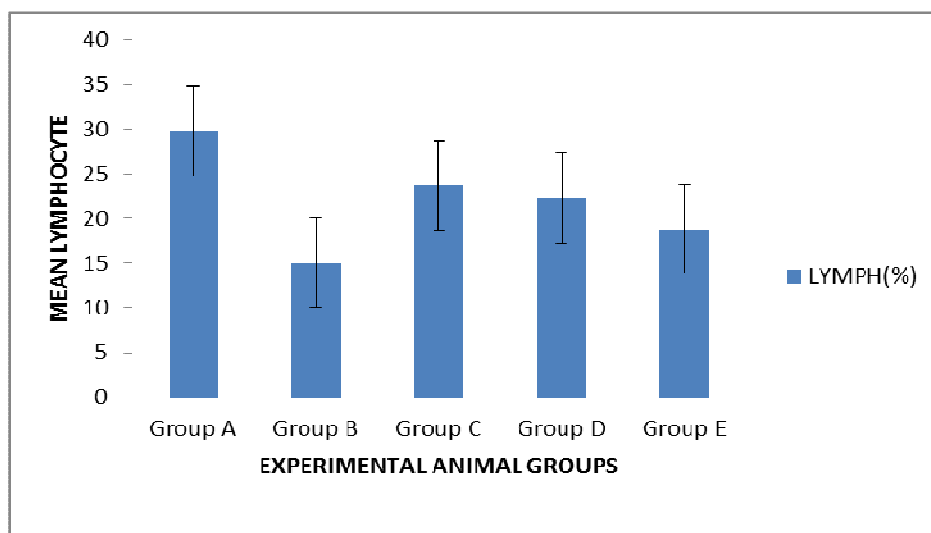
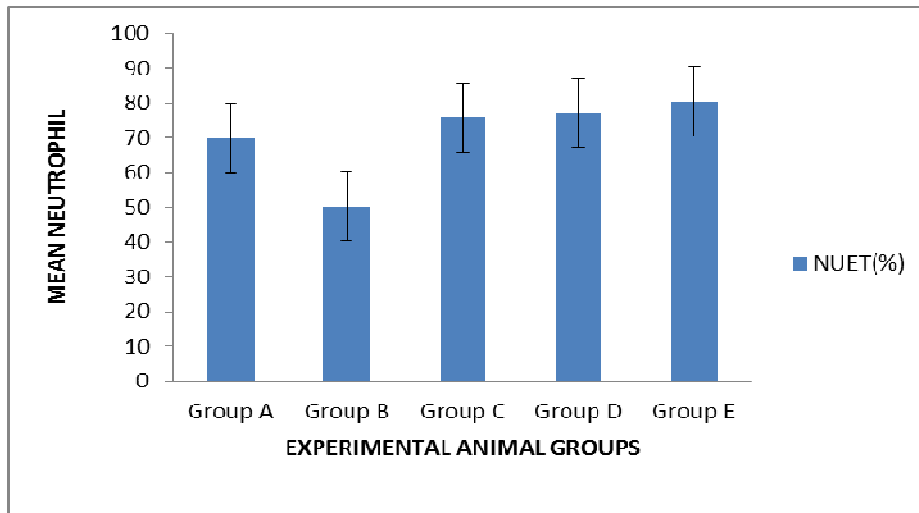
Table 9: LYMPHOCYTE COUNT (%) WITH MEAN±SD OF RATS

GROUPS	Concentration	MEAN LYMPH % ±SD	t-VALUE	P-VALUE
A	Control	29.8 ± 6.80	8.749	.003
B	<i>Pleurotus tuber-regium</i> 400mg	15.0 ± 13.00	1.999	.184
C	<i>Pleurotus tuber-regium</i> 600mg	23.7 ± 2.89	14.200	.005
D	<i>Pleurotus tuber-regium</i> 1000mg	22.3 ± 2.06	21.586	.000
E	<i>Agaricus bisporous</i> 400mg	18.8 ± 2.14	21.588	.000

FIGURE 1: MEAN VALUES OF HAEMATOLOGICAL PARAMETERS







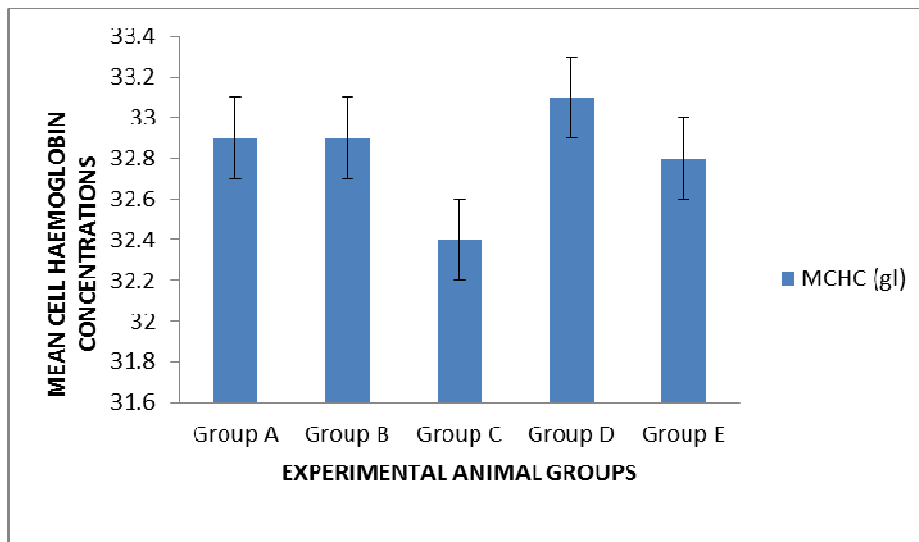
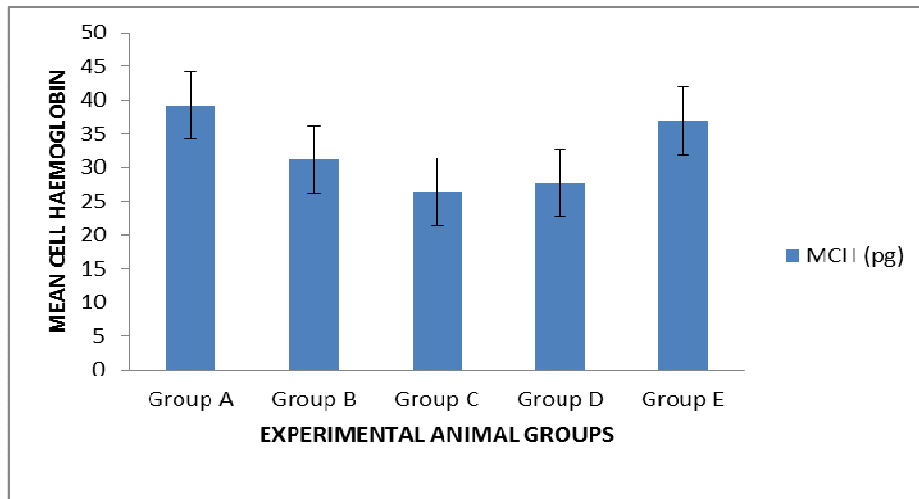


Table 10: Mean \pm SD of Serum Biomarkers

Parameters	Control (A)	<i>Pleurotus tuber-regium</i> 400mg (B)	<i>Pleurotus tuber-regium</i> 600mg (C)	<i>Pleurotus tuber-regium</i> 1000mg (D)	<i>Agaricus bisporus</i> 400mg (E)
Superoxide Dismutase (units/min/mg protein)	74.33 \pm 9.79	79.53 \pm 10.08	55.29 \pm 10.37	79.91 \pm 12.71	113.58 \pm 48.06
Catalase (units/min/mg protein)	0.49 \pm 0.34	0.57 \pm 0.02	0.60 \pm 0.77	0.13 \pm 0.11	0.72 \pm 0.30
Reduce Glutathione (GSH μ mol/ml)	0.743 \pm 0.37	0.90 \pm 0.01	0.94 \pm 0.03	0.72 \pm 0.04	0.74 \pm 0.01
Malondialdehyde (nmol/ml)	27.73 \pm 1.03	27.46 \pm 0.27	28.04 \pm 0.88	32.34 \pm 16.74	23.32 \pm 2.09

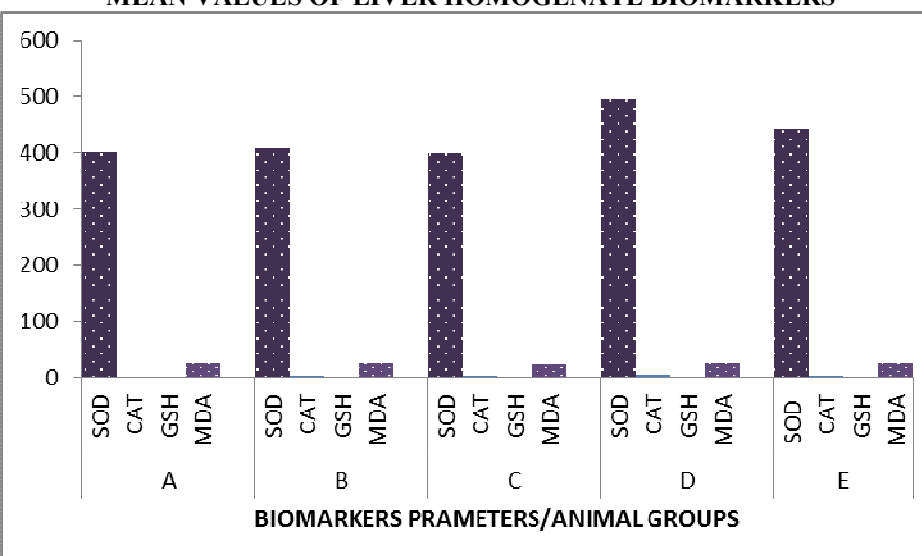
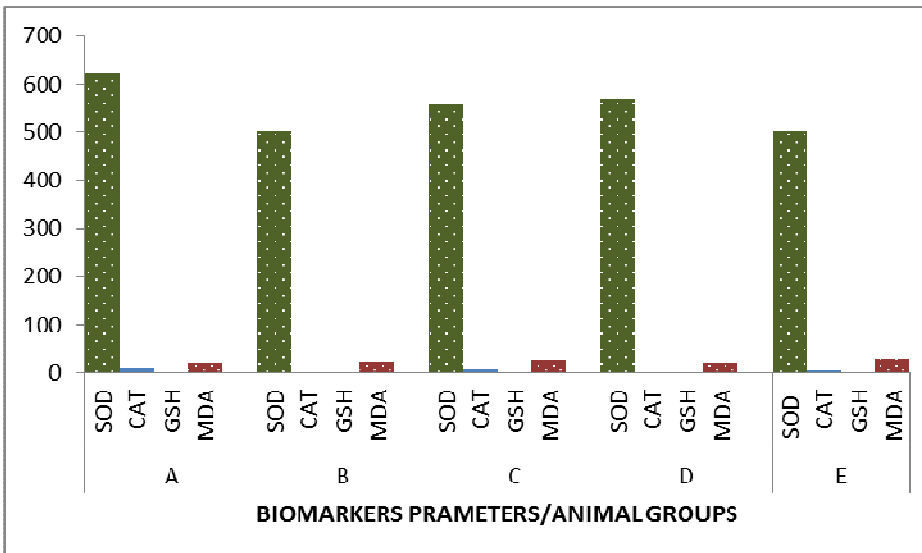
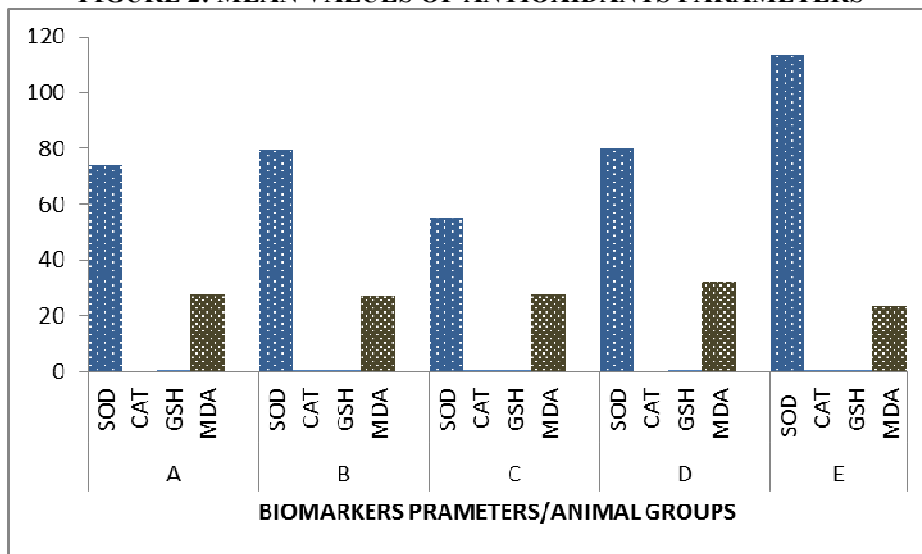
Table 11: Mean \pm SD of Liver Homogenate Biomarkers

Parameters	Control (A)	<i>Pleurotus tuber-regium</i> 400mg (B)	<i>Pleurotus tuber-regium</i> 600mg (C)	<i>Pleurotus tuber-regium</i> 1000mg (D)	<i>Agaricus bisporus</i> 400mg (E)
Superoxide Dismutase (units/min/mg protein)	621.49 \pm 189.53	502.13 \pm 30.82	557.14 \pm 63.02	566.86 \pm 57.45	501.97 \pm 56.10
Catalase (units/min/mg protein)	8.89 \pm 7.99	1.13 \pm 0.44	5.77 \pm 4.43	2.30 \pm 1.32	3.81 \pm 1.73
Reduce Glutathione (GSH μ mol/ml)	0.98 \pm 0.46	0.92 \pm 0.24	0.69 \pm 0.02	0.80 \pm 0.41	0.57 \pm 0.05
Malondialdehyde (nmol/ml)	23.32 \pm 1.87	23.97 \pm 1.35	27.06 \pm 4.15	23.12 \pm 1.09	24.90 \pm 1.89

Table 12: Mean \pm SD of Kidney Homogenate Biomarkers

Parameters	Control (A)	<i>Pleurotus tuber-regium</i> 400mg (B)	<i>Pleurotus tuber-regium</i> 600mg (C)	<i>Pleurotus tuber-regium</i> 1000mg (D)	<i>Agaricus bisporus</i> 400mg (E)
Superoxide Dismutase (units/min/mg protein)	402.08 \pm 34.72	408.93 \pm 26.87	399.96 \pm 56.06	495.41 \pm 42.28	443.00 \pm 69.02
Catalase (units/min/mg protein)	1.99 \pm 1.46	4.53 \pm 0.57	4.23 \pm 1.44	5.68 \pm 3.04	4.50 \pm 2.67
Reduce Glutathione (GSH μ mol/ml)	0.91 \pm 0.13	0.85 \pm 0.05	0.77 \pm 0.04	0.87 \pm 0.06	0.88 \pm 0.06
Malondialdehyde (nmol/ml)	24.67 \pm 1.45	25.85 \pm 1.89	23.33 \pm 0.10	25.17 \pm 0.78	24.88 \pm 2.03

FIGURE 2: MEAN VALUES OF ANTIOXIDANTS PARAMETERS



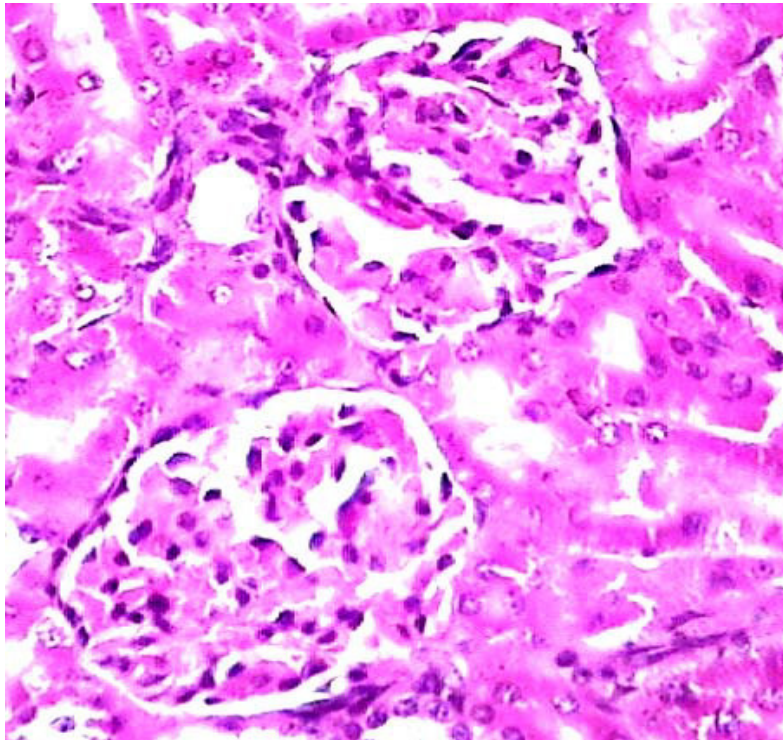


Plate i. Light micrograph of the liver of rat treated with 400 mg/kg bw of *Pleurotus tuber-regium*

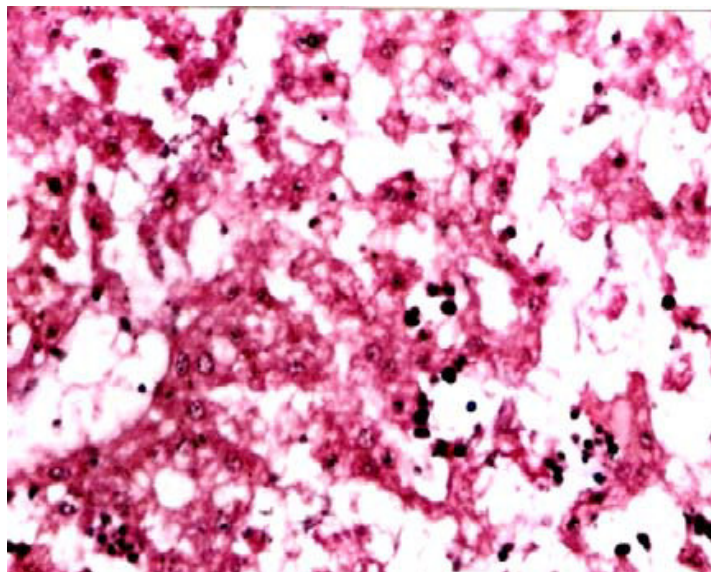


Plate ii. Light micrograph of the kidney of rat treated with 400 mg/kg bw of *Pleurotus tuber-regium*

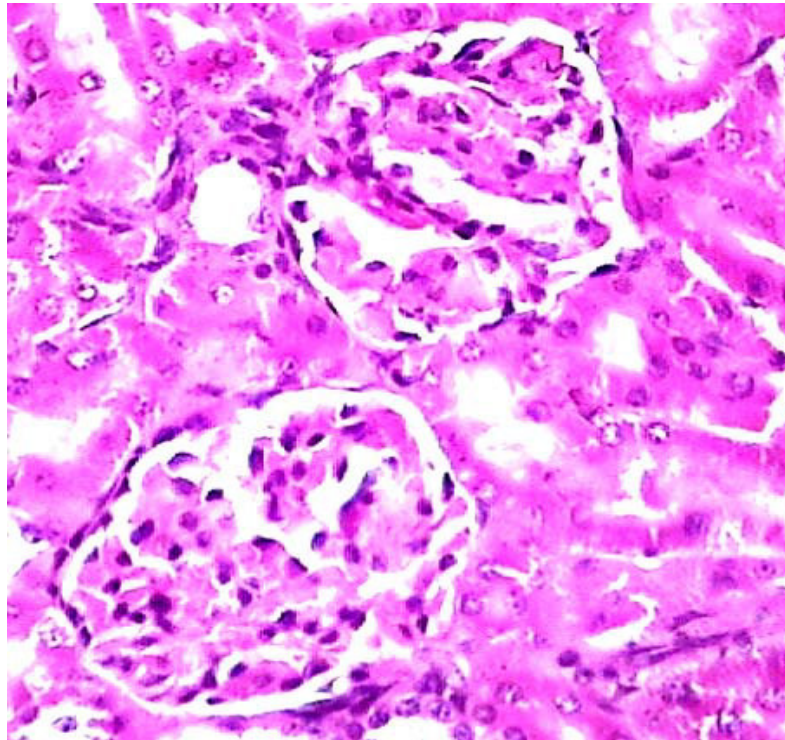


Plate iii. Light micrograph of the liver of rat treated with 400 mg/kg bw of *Agaricus bisporus*

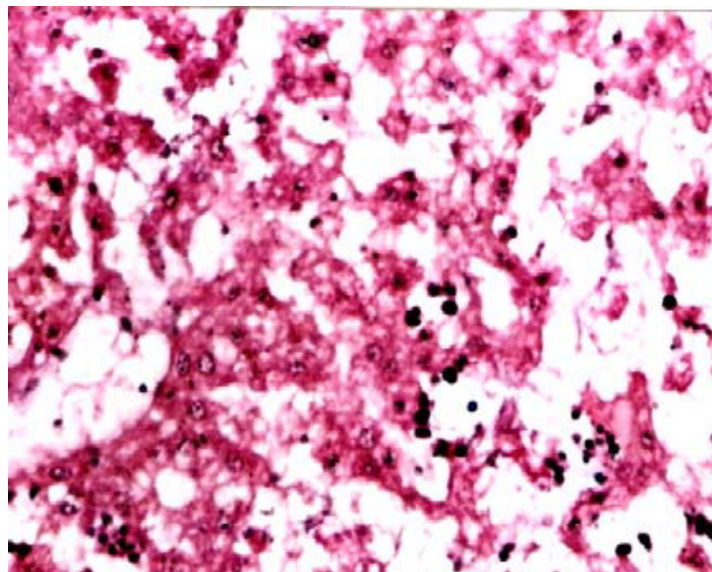


Plate iv. Light micrograph of the kidney of rat treated with 400 mg/kg bw of *Agaricus bisporus*

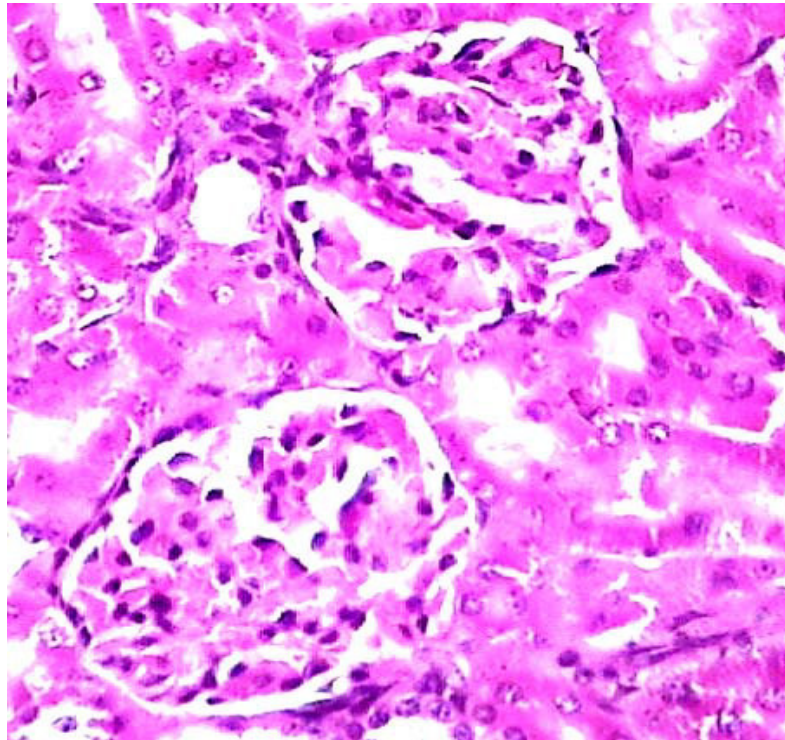


Plate v. Light micrograph of the liver of rat treated with distilled water

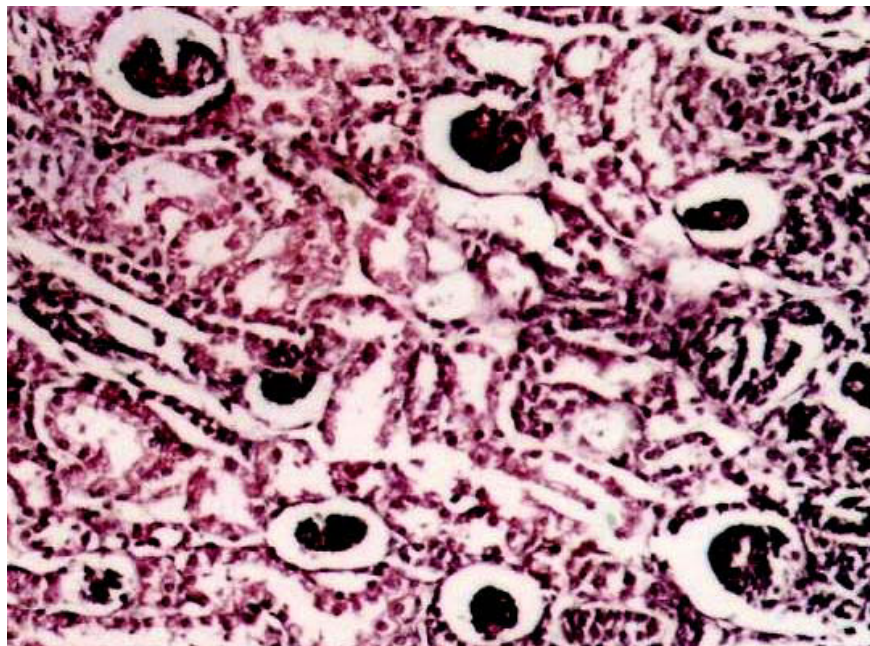


Plate vi. Light micrograph of kidney of rat untreated (control)

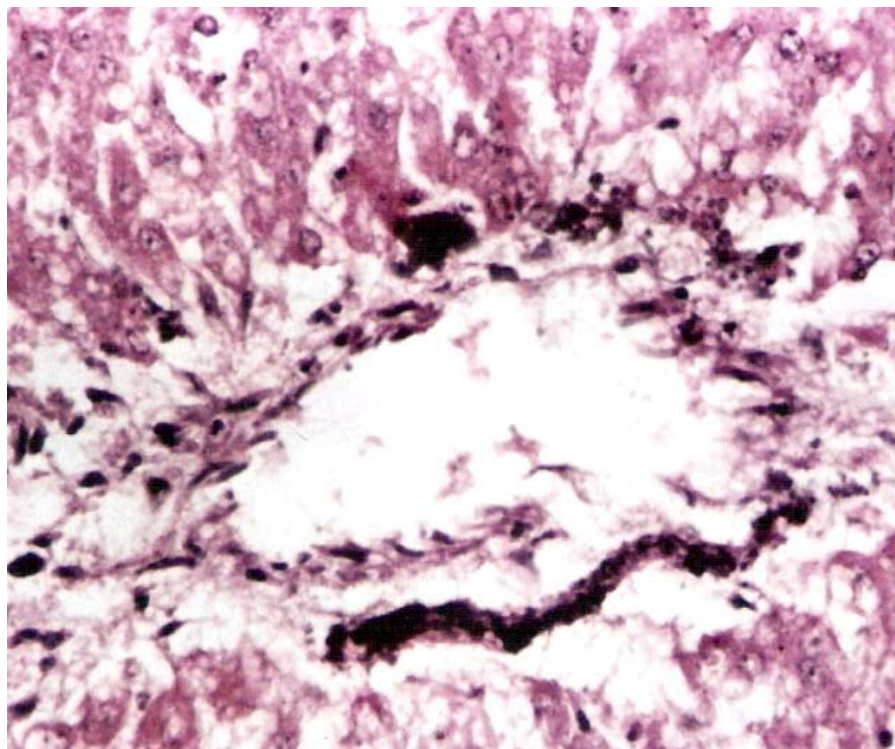


Plate vii. Light micrograph of liver of rat untreated liver

Discussion

Two species of mushroom provided (*Pleurotus tuber-regium* and *Agaricus bisporus*), at the end the 4th week boost the immune system. The mushrooms had immune modulatory properties and also antioxidant properties (Mau *et al.*, 2004).

The effect of the mushroom extract were observed on the haematological parameters as shown in tables 1, 2 3 and 4.

Histopathology examinations review that the rat treated with the two species of mushroom does not show any abnormal growth either at the liver or in the kidney as shown in plates 1-5.

The mean values of Biomarkers Parameters of rats in different treatment regimen. Lipid peroxidation products (MDA), superoxide dismutase (SOD), Catalase Activity (CAT), Glutathione reductase (GSH) are antioxidants enzymes measured to detect toxic consequences of oxidative stress in mammalian systems (Sue *et al.*, 1998). They are cellular and enzymatic defenses against oxidative stress (Winston and Giulio, 1991). Oxidative stress causes toxic and adaptive responses within a cell. Several studies have demonstrated the importance of an antioxidant defenses in protecting cells and organisms from oxidative damage and toxicity (Furono *et al.*, 1996).

The serum biomarkers, superoxide dismutase shows increase in rats administered with 400mg/kg bw and 1000mg/kg bw *Pleurotus tuber-regium* and decrease in rats administered with 600mg/kg bw *Pleurotus tuber-regium* and 400mg/kg bw *Agaricus bisporus* when compared with the control group. The increase are statistically significant with $P < 0.05$. Superoxide dismutases (SODs) are enzymes that scavenge Oxygen (O_2) by a rapid dismutation reaction, the activity of SOD leads to production of hydrogen peroxide that is required by catalase (Iyawe and Onigbinde 2004). These may have explained the observed raise in the activities of these enzymes.

Catalase is a common enzyme found in living organisms. Its function includes catalyzing the decomposition of hydrogen-peroxide to water and oxygen. Activities of SOD can lead to the production of hydrogen peroxide (H_2O_2) required by catalase which invariably lead to the rise of these enzymes in a diseased condition.

Hydrogen peroxide (H_2O_2) is a harmful by – product of many normal metabolic processes. To prevent damage, it must be quickly converted into other, less dangerous substances (Quinlan *et al.*, 1994). To manage this problem, the enzyme catalase is frequently used by cells to rapidly catalyze the decomposition of hydrogen peroxide into less reactive gaseous oxygen and liquid water molecules (Krinsky, 1992).

Glutathione (GSH), a non-enzymatic antioxidant, these enzymes also have high affinity for H_2O_2 and may be important for maintenance of low intracellular levels of H_2O_2 in the cytosol where catalase levels are low. In addition to acting as a reducing agent, (Anderson *et al.*, 1996), GSH also acts as a substrate or co-substrate in

many essential enzymatic reactions, the depletion of GSH during oxidative stress could have a significant impact on the antioxidant pose within a cell (Scarpa *et al.*, 1983).

Catalase and Glutathione shows increase in rats administered with 400mg/kg bw, 600mg/kg bw *Pleurotus tuber-regium* and 400mg/kg bw *Agaricus bisporus* when compared with the control group. The increase are statistically significant with $P < 0.05$. Malondialdehyde increases in all the groups administered with *Pleurotus tuber-regium* but shows decrease in rats administered with *Agaricus bisporus*. The increase are not statistically significant with $P > 0.05$.

The liver homogenate biomarkers, superoxide dismutase decreases in all experimental group compared to the control group and the decrease are statistically significant with $P > 0.05$. Catalase and Glutathione shows decrease in all experimental group compared with control group and the decrease are not statistically significant with $P > 0.05$. Malondialdehyde increases in rats administered with 600mg/kg bw *Pleurotus tuber-regium* and 400mg/kg bw *Agaricus bisporus* and there was decrease in rats administered with 400mg/kg bw and 1000mg/kg bw *Pleurotus tuber-regium* compared with the control group. The decrease are not statistically significant with $P > 0.05$.

The biomarkers of kidney homogenate, superoxide dismutase shows increase in rats with 400mg/kg bw, 1000mg *Pleurotus tuber-regium* and 400mg/kg bw of *Agaricus bisporus* but shows decrease in rats administered with 400mg/kg bw *Pleurotus tuber-regium* when compared with the control group. The increase are statistically significant with $P < 0.05$. Catalase shows increase in all experimental rats when compared with the control group and the increase is statistically significant with $P < 0.05$. Glutathione shows decrease in all experimental rats when compared with control group and the decrease are not statistically significant with $P > 0.05$. Malondialdehyde shows increase in rats administered with 400mg and 1000mg/kg bw *Pleurotus tuber-regium* but shows decrease in rats administered with 600mg/kg bw *Pleurotus tuber-regium* and 400mg/kg bw *Agaricus bisporus* when compared to the control group, the decrease are not statistically significant with $P > 0.05$. Generally, the experimental rats showed less significant increase in the level of antioxidants and further research is needed to be carried out.

CONCLUSION

Medicinal mushroom has been shown to have immunomodulatory properties and antioxidant properties including effects on cancer, infections, allergy, asthma and inflammatory disorder. Mushrooms have been seen to contain proteins, carbohydrates, vitamins, mineral constituents and fats in low amount, all of which are efficient immune developing and body building sources.

From the present study, the two species (*Pleurotus tuber-regium* and *Agaricus bisporus*) of edible mushroom provided has shown their effectiveness on immune system due to the results/analysis. *Agaricus bisporus* should be further tested in higher concentrations.

It can be concluded that the diverse benefits of mushrooms towards human by the words of the father of medicine that is, Hippocrates "Let food be your medicine and medicine be your food". This saying aptly suits mushrooms, as they have tremendous medicinal food, drugs and mineral values; hence they are valuable asset for the welfare of human.

RECOMMENDATIONS

Mushrooms has been used for many centuries, it serves as a major source of medicine for human which has help to influence the immune system. It is therefore recommended that:

- Mushrooms must be well identified depending upon what it is to be treated with.
- Aqueous and ethanol extraction are the two basic extractions solvent for both mushrooms and plant materials, reduced amount of ethanol should be used because it may hamper the immune system.

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