Hepatoprotective Effect of Ethanolic Extract of Oyster Mushroom (*Pleurotus ostreatus*) on acutely Administered Lead Poison in Albino Rats

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ABSTRACT

The hepatoprotective effect of ethanolic extract of Oyster mushroom (*Pleurotus ostreatus*) on acutely administered lead poison in albino rats was studied by assaying the concentration of liver markers enzymes: Alanine Transaminase (ALT), Aspartate Transaminase (AST), Alkaline Phosphate (ALP) and Total Bilirubin (TBL), Total Protein (TP), Albumin (ALB). A total of twenty-four albino rats were distributed into six groups of four animals each. Group 1 (control), group 2 (20mg/kg lead alone), group 3 (40mg/kg lead alone), group 4 (250mg/kg mushroom alone), group 5 (250mg/kg mushroom and 20mg/kg lead), and group 6 (250mg/kg mushroom and 40mg/kg lead). These animal groups were administered their respective treatments for a period of twenty-one days. Thereafter, various tests were carried out on the liver enzymes. The groups administered with lead (low and high dose) showed significant increase in levels of AST, ALT, ALP & TBL but reduced levels of ALB and TP respectively (59.00±1.63, 23.00±1.47, 61.25±2.49, 16.25±0.62, 31.75±1.18, 46.75±0.85, p <0.05), (63.75±0.48, 29.75±0.85, 72.25±1.03, 18.00±0.41, 36.00±1.47, p<0.05) compared to the control with mean values of 37.75±1.88, 15.50±0.64, 46.50±1.55, 11.75±0.48, 39.00±0.71, 71.50±1.32. On the other hand, groups 4, 5 and 6 treated with oyster mushroom extract showed a hepatoprotective effect by maintaining the level of ALT (14.25±1.11, 16.00±0.91, 20.25±0.48, p<0.05), AST (24.25±1.49, 27.25±1.18, 31.25±1.65, p<0.05), ALP (34.25±1.11, 37.50±2.72, 39.50±0.87, p<0.05), TP (78.25±1.37, 72.25±0.47, 74.25±2.56, p<0.05), ALB (38.75±0.48, 40.00±0.41, 43.25±0.63, P<0.05) and TBL (11.00±0.41, 11.75±0.48, 13.25±0.48 p<0.05), around the normal level. Histological examination of the liver tissues showed that mushroom extract was hepatoprotective against the lead induced alterations, notably inflammation, necrosis and vacuolation. This result reveals that aqueous extract from *Pleurotus ostreatus* has significant hepatoprotection against lead induced hepatocellular injury.

Keywords: Lead, *Pleurotus ostreatus*, Hepatoprotection, Hepatotoxicity, AST, ALT, ALP, TP, ALB, TBL

INTRODUCTION

Lead poisoning is a serious and sometimes fatal condition that occurs when lead builds up in the body [1,2,3]. A toxic dose of lead poisoning may result in serious health problems which include; seizures, coma, malignancy, cell death, periportal inflammation, enlargement of the liver cells into hyperplasia (initiating the formation of tumors on the liver) and hepatocyte damage [4,5,6]. Lead is bio-accumulated and is most concentrated in the bone but has more toxic effects in the liver and kidney, and children are at a higher risk of lead poisoning [7,8,9]. The outbreak of lead poisoning was also discovered in 2010 at Zamfara State, Nigeria, which killed at least 400 inhabitants that were mainly children [10,11]. Lead is a common environmental pollutant, and its widespread use has resulted in extensive environmental contamination and adverse health effects, affecting almost every organ and system in the human body [12,13,14].
contaminates the environment through releases from mining industries and factories that use or produce lead, lead alloys, lead compounds, lead-acid batteries, lead wires, pipes and metal recycling [15,16,17]. Lead is readily absorbed by the body via primary routes of entry, inhalation and ingestion [18,19,20]. Some lead compounds are colorful and are widely used in paints [21] and children are at a greater risk as they’re more likely to put objects in their mouth [22]. In adults, exposure at work is a common cause of lead poisoning, with certain occupations at particular risk [23]. Chelation is a method used to treat metal poisoning. Chelating agents are not without side effects and can also remove beneficial metals from the body. Moreover, these chelating agents in turn are potentially toxic [14] and often fail to remove lead burden from body tissues, according to research report on chelators and lead toxicity management. In order to address these problems, natural therapies to promote chelation, detoxification and protection are gaining popularity because of minimal or no side effects [16]. Medicinal properties of plants have been investigated in the light of recent scientific developments throughout the world due to their potent pharmacological activities, low toxicity and economic viability [19]. Thus, there has been an increased interest in the therapeutic potential of plant products and medicinal plants for having beneficial role in reducing lead poisoning in cells, tissues, and organs. Mushrooms are macrofungi that have distinctive basidiomata or ascomata large enough to be seen with the naked eye and to be picked with unaided hand. They are poor in calories, rich in proteins, fibers, carbohydrates and important vitamins and minerals [12]. They are considered as functional foods or nutraceuticals [14]. Liver disease is a serious problem worldwide and several studies have demonstrated the protective effects of herbs against experimentally induced liver injury. Additionally, a number of herbs such as the edible mushroom revealed promising properties which includes antioxidant, anti-cancer, anti-tumor and anti-inflammatory actions [15,17,18]. Therefore, this research is aimed at investigating the hepatoprotective effect of mushroom extract against lead induced hepatotoxicity in albino rats. Liver damage is a widespread disease, which in most cases involves oxidative stress and it’s characterized by a progressive evolution from steatosis to chronic hepatitis, fibrosis, cirrhosis and hepatocellular carcinoma. Recent research on functional foods such as nutraceuticals showed that many natural agents exert protective and therapeutic effects on the liver and some of the other herbal and nutritional supplements also have characteristics that make them beneficial to the liver [16].

**MATERIALS AND METHODS**

This research work was carried out in Animal and Environmental Biology, Animal House in Choba Campus, University of Port Harcourt, Choba, Rivers State, Nigeria. Fresh *P. ostreatus* (Oyster Mushroom) was purchased from the mushroom farm in the University of Science and Technology, Nkpolu-Oroworukwo, Port Harcourt, Rivers
State, Nigeria. Twenty grammes (20g) of water soluble lead nitrate (Pb(NO\textsubscript{3})\textsubscript{2}) was purchased from Geochem store in Choba, Rivers State, Nigeria. Twenty-four (24) albino rats of both sexes weighing 60-100g were obtained from the Department of Animal and Environmental Biology. The animals underwent acclimatization for a period of 7 days prior to the experiment. During the preparation of ethanolic extract of \textit{P. ostreatus}, 870ml of ethanol was poured onto 300g of powdered \textit{P. ostreatus} and then blood samples were collected from the retro-orbital venous plexus under light ether anesthesia. After blood collection, all animals were sacrificed by cervical dislocation, the liver was extracted, rinsed in isotonic sterile saline, blotted and preserved in 10% neutral formalin solution at 4ºC for histological examination. The liver marker enzymes; Alanine Transaminase (ALT), Aspartate Transaminase (AST), Alkaline Phosphate (ALP) and Total Bilirubin (TBL), Total Protein (TP), Albumin (ALB), underwent different procedures for test results. Histological study was carried out on the liver for 5-7 days. Statistical analysis was carried out using the ANOVA. The results were reported as mean ± SEM and considered significantly different at P≤0.05.

**RESULTS**

The serum levels of AST revealed that group 1 to 6 had mean values of 37.75 ± 1.88, 59.00 ± 1.63, 63.75 ± 0.48, 24.25 ± 1.49, 27.25 ±1.18, and 31.25±1.65. Group 3 (lead alone at high dose) had the highest serum level while Group 4 (mushroom extract alone) had the lowest serum level for Aspartate Transaminase (AST). The mean values for ALT levels from Group 1 through 6 were 15.50 ± 0.64, 23.00 ± 1.47, 29.75±0.85, 14.25 ± 1.11, 16.00 ± 0.91 and 20.25 ± 0.48. The result showed that Group 3 had the highest mean serum level while Group 4 had the least mean value for Alanine Transaminase (ALT). The result for ALP levels across the groups from 1 to 6 were 46.50±1.55, 61.25±2.49, 72.25±1.03, 34.25±1.11, 37.50±2.72 and 39.50±0.87. From the result, group 3 had the highest mean value and group 4 had the least mean value for Alkaline Phosphate (ALP). The mean values for serum levels of total bilirubin (TBL) were 11.75 ± 0.48, 16.25 ± 0.62, 18.00 ± 0.41, 11.00 ± 0.41, 11.75 ± 0.48 and 13.25 ± 0.48 across the groups (from 1 to 6) respectively. From the result, TBL had the highest mean value recorded for group 3 and the lowest was recorded for group 4. The serum levels for Total protein (TP) for all groups (1-6) had mean values of 46.75 ± 0.85, 61.25 ± 2.49, 36.00 ±1.47, 78.25 ± 1.37, 72.25 ± 0.47 and 74.25 ± 2.56. Group 4 had the highest mean value for TP while group 3 had the least mean value for TP. The mean values for serum levels of albumin (ALB) across the respective groups (1 to 6) are 39.00 ± 0.71, 31.75±1.18, 27.00 ± 0.71, 38.75 ± 0.48, 28
40.00 ± 0.41 and 43.25 ± 0.63. The overall result for albumin showed that group 6 had the highest mean value while group 3 had the least mean value. The Histopathological analysis carried out showed that group 1 (control) had structural integrity without necrosis, inflammation, or fibrosis. Group 2 (lead alone at low dose) showed inflammation, fatty change, and area of necrotic tissues. Group 3 (lead alone at high dose) showed high inflammation and slightly distorted liver architecture. Group 4 (mushroom alone) showed structural integrity without necrosis and inflammation. Group 5 (mushroom and lead at low dose) showed alteration of portal tract while Group 6 (mushroom and lead at high dose) showed fatty change, vacuolar change and minimal inflammation.

Table 1: The mean response of Liver Marker Enzymes

<table>
<thead>
<tr>
<th>LIVER MARKERS ENZYMES</th>
<th>AST</th>
<th>ALT</th>
<th>ALP</th>
<th>TP</th>
<th>ALB</th>
<th>TB</th>
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<tr>
<td><strong>GROUPS</strong></td>
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<tr>
<td>Control</td>
<td>37.75±1.88</td>
<td>15.50±0.64</td>
<td>46.50±1.55</td>
<td>71.50±1.32</td>
<td>39.00±0.71</td>
<td>11.75±0.48</td>
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<tr>
<td>Group 2</td>
<td>59.00±1.63</td>
<td>23.00±1.47</td>
<td>61.25±2.49</td>
<td>46.75±0.85</td>
<td>31.75±1.18</td>
<td>16.25±0.62</td>
</tr>
<tr>
<td>Group 3</td>
<td>63.75±0.48</td>
<td>29.75±0.85</td>
<td>72.25±1.03</td>
<td>36.00±1.47</td>
<td>27.00±0.71</td>
<td>18.00±0.41</td>
</tr>
<tr>
<td>Group 4</td>
<td>24.25±1.49</td>
<td>14.25±1.11</td>
<td>34.25±1.11</td>
<td>78.25±1.37</td>
<td>38.75±0.48</td>
<td>11.00±0.41</td>
</tr>
<tr>
<td>Group 5</td>
<td>27.25±1.18</td>
<td>16.00±0.91</td>
<td>37.50±2.72</td>
<td>72.25±0.47</td>
<td>40.00±0.41</td>
<td>11.75±0.48</td>
</tr>
<tr>
<td>Group 6</td>
<td>31.25±1.65</td>
<td>20.25±0.48</td>
<td>39.50±0.87</td>
<td>74.25±2.56</td>
<td>43.25±0.63</td>
<td>13.25±0.48</td>
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</table>

Values are presented in mean ± S.E.M (n= 4), p <0.05 "*" means values are statistically significant compared" to the control group.

Figure 1: Mean levels of Liver Marker Enzymes
DISCUSSION

Lead poisoning has been recognized as a major public health issue, particularly in developing countries with children mostly susceptible to it as they’re most likely to put objects in their mouth [3]. The experimental intoxication induced by lead is widely used for modelling liver injury in rats because of the ability of lead to cause severe cell impairment. *P. ostreatus* (oyster mushroom) on the other hand have been reported to possess potent antioxidant, immunomodulatory and anticancer properties [7,9]. Research has also shown it to have potential hepatoprotective effects [11]. From the results shown, the administration of lead alone (20mg/kg and 40mg/kg) in group 2 and 3 increased serum levels of acute liver damage indicators (AST, ALT, ALP, TBL) and decreased protein content indicators (TP, ALB), which was supported by histological examination of the liver section, revealing distorted architecture of hepatocytes and major generalized vacuolations. However, pre-treatment with ethanolic extract obtained from the oyster mushroom was shown to provide protection against hepatic injury caused by lead as seen in groups 5 and 6 which was also supported by histological examination, showing liver with structural integrity and minimal inflammation. The data obtained revealed that the concentration of these parameters were statistically significant (P<0.05) in the different groups pre-treated with mushroom extract. This was result was also supported by the findings of [4]. In the different groups, it could be seen that the liver marker enzymes, total protein and albumin decreased significantly in group 2 and 3, which could be concluded as the inability of the liver to synthesize protein due to liver damage. While group 4, 5, and 6 pre-treated with oyster mushroom extract showed elevations in the total protein and albumin level. Histopathological studies also confirmed the protective effect of *P. ostreatus* extract against lead induced liver damage. The histological appearance in group 4 (mushroom alone) and group 5 (mushroom and lead with low dose) appeared normal with minimal inflammation seen only in group 6 (mushroom and lead with high dose). The other groups (2 & 3) induced with lead alone showed areas of necrosis, fatty changes and scattered lymphocytes, which was reported by [4,7] that lead overdosse causes ultrastructural changes in the liver.

CONCLUSION

It can be concluded that ethanolic extract of oyster mushroom is hepatoprotective against acutely administered lead toxin in albino rats. The serum levels showed a decrease in acute liver damage indicators and the histological studies also showed significant alleviation in the alteration caused by lead intoxication, which proves that oyster mushroom has hepatoprotective effect.

REFERENCES


