



Antidiabetic Effect of *Pleurotus ostreatus* (Jacq.ex Fr) kumm.

Mushroom on Alloxan-induced Diabetic Rats.

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Abstract

Antidiabetic effects of ethanolic extract of *Pleurotus ostreatus* (mushroom) on alloxan-induced diabetic rats was studied. The median lethal dose (LD₅₀) of the extract was determined to be 3,472.14 mgkg⁻¹ and a single dose of 380.0, 760.0 and 1140.0 body weight of the extract were intraperitoneally administered as the treatment dose and the blood glucose levels (BGL) examined for 7 hours and 15 hours (prolonged) at 2 and 4 hours intervals respectively. The extract exhibited significant (p<0.05 and p<0.01) reduction in the blood glucose levels of the albino rats. The extract compared favourably with the standard reference drug (metformin) which all gave their maximum BGL reduction at 5 hours duration. The confirmation of antidiabetic potentials of the *Pleurotus ostreatus* tuber has been justified in this study as claimed by traditional medicine practitioners in Akwa Ibom State.

Keywords: *Pleurotus ostreatus*; diabetic rats; hypoglycaemic activity; ethanolic extract; metformin.

1. Introduction

In traditional practices, medicinal plants are used to control diabetes mellitus in many African countries. This has caused an awareness in the number of experimental and clinical investigations directed towards the validation of the antidiabetic properties, which are empirically attributed to these remedies. The hypoglycemic effects of some edible plants used as antidiabetic remedies have been reported [1, 2] which justify their nutraceutical potentials. These types of plants could be developed for diabetic patients in order to improve their diet and control their diseases. With this type of menu, diabetic patients could potentially reduce

the dose of their orthodox hypoglycemic drugs.

Pleurotus ostreatus, known as oyster mushroom is a macrofungus of the class Basidiomycetes and Family Agaricaceae [3]. The plant is an edible mushroom with excellent flavor which grows on logs or tree stumps in shelf-like layers. They have a very short lateral stalk with an oyster shaped pileus. *Pleurotus ostreatus* has high protein content and may contain other constituents such as Vitamin B₁ and B₂ and low caloric level. This makes it very popular for consumption among people who are dieting [4]. Lovastatin an active chemical principle isolated from the

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oyster mushroom is used in San Francisco to reduce “bad” cholesterol (LDL) in hypercholesterolemic patients [5].

The purpose of this study is to validate the anti-hyperglycemic effect of *Pleurotus ostreatus* mushroom cherished by many Nigerians as a delicacy in soup condiment.

2. Materials and Methods

Plant Materials

Fresh *Pleurotus ostreatus* mushrooms were collected from dead palm trees in Afaha Atan Village, Ibiono Ibom Local Government Area of Akwa Ibom State, Nigeria in September, 2011. The plant species was identified and authenticated by a mycologist; Dr. J. P. Essien in the Department of Microbiology, University of Uyo, Uyo. Herbarium specimen (DBH 52) was deposited at the Department of Botany herbarium. The fresh mushrooms sample were dried on a laboratory table for 2 weeks and reduced to powder by pounding the dried sample with mortar and pestle.

Two hundred grams of the powdered sample was macerated in 95% ethanol (500ml) for 72 hours. The liquid filtrate obtained was concentrated *in-vacuo* at 40°C. They yield was 5.12% w/w. The extract was stored in a refrigerator at 4°C until used for this experiment.

Phytochemical Screening

Phytochemical screening of the extract was carried out according to the standard methods [6, 7].

Experimental Animals

Albino wister rats (101-120g) and albino swiss mice (15-30g) of both sexes were obtained from the University of Uyo animal house. The animals were maintained on standard animal pallets and water *ad libitum*.

Determination of Median Lethal Dose (LD₅₀)

The median lethal dose (LD₅₀) of the *Pleurotus ostreatus* mushroom extract was estimated using albino mice by intraperitoneal (IP) route administration of different doses of 1000-5000mg/kg to five groups of 5 mice per group respectively after starving the animals for 24hours according to the method of Lorke [8]. The IP route was adopted because of its sensitivity and rapid results. The animals were observed from possible manifestation of physical signs of toxicity such as writhing, decreased motor activity, decreased body/limb tones, decreased respiration and finally death. Records on the number of deaths observed were taken in each group within 24hours. The LD₅₀ was calculated as the geometrical means of the maximum dose producing 0% (a) and minimum dose producing 100% mortality (b).

$$LD_{50} = \sqrt{ab}$$

Evaluation of Antidiabetic Activity of the Extract

Induction of Diabetes

Male wister rats were made diabetic by a single dose of intraperitoneal (IP) injection of 150mg/kg body weight of alloxan monohydrate in sterile normal saline. The rats were maintained on 5% glucose solution for next 24 hours to prevent hypoglycaemia. Five days later, blood samples were drawn from tail vein and glucose levels were determined to confirm the development of diabetes (250mg/dl and above). The diabetic rats were divided into five groups, each containing five animals and treated as follows:

- Group I: Diabetic rats administered with 5ml/kg of saline water as the control
Group II: Diabetic rats treated with mushroom ethanol extract (447.21mg/kg/day)
Group III: Diabetic rats treated orally with mushroom extract (894.42mg/kg/day) in aqueous solution for 15-days.
Group IV: Diabetic rats treated orally with mushroom extract (1341.63mg/kg/day) in aqueous solution
Group V: Diabetic rats given 10mg/kg of metformin for 15-days.

Blood samples were collected from the tail vein just prior to and 1h, 3h, 5h and 7h after drug administration for acute study. The effect of the mushroom extract was also tested for a prolonged treatment lasting for 15-days.

The fasting BGL of all the rats were monitored and recorded at regular intervals during the experimental period. For acute study the BGL was monitored after 1, 3, 5 and 7 hour of administration of the extract and at the end of 0, 1, 5, 10 and 15-days for prolonged administration. The blood samples were

collected through the tail just prior to and on the time frames after drug treatment. The blood was dropped on the dextrostix reagent pad, which was insert into microprocessor digital blood glucometer and the readings recorded [9].

Statistical Analysis

The results are expressed as mean S.E.M., the significance of various treatments was calculated using students t-test and were considered statistically significant when $p < 0.05$ [10].

3. Results

The results of phytochemical screening of the extract of *Pleurotus ostreatus* gave positive indications of the presence of alkaloids, saponins, flavonoids and cardiac glycosides.

The mice treated intraperitoneally (IP) with a single dose of 1000-5000mg/kg of *Pleurotus ostreatus* extract after 24-hour starving exhibited physical signs of toxicity in the animals depending on the dose administered with ranged from writing, respiration distress, decreased limb tone, and death within 24 hours post administration of the extract. All the animals given 4000 and 5000mg/kg doses of the extract died. The LD₅₀ of the extract was calculated to be 3,472.14mg/kg.

The anti-diabetic activity of the ethanolic *Pleurotus ostreatus* extract gave a dose-dependent reduction in the BGL of the alloxan induced diabetic

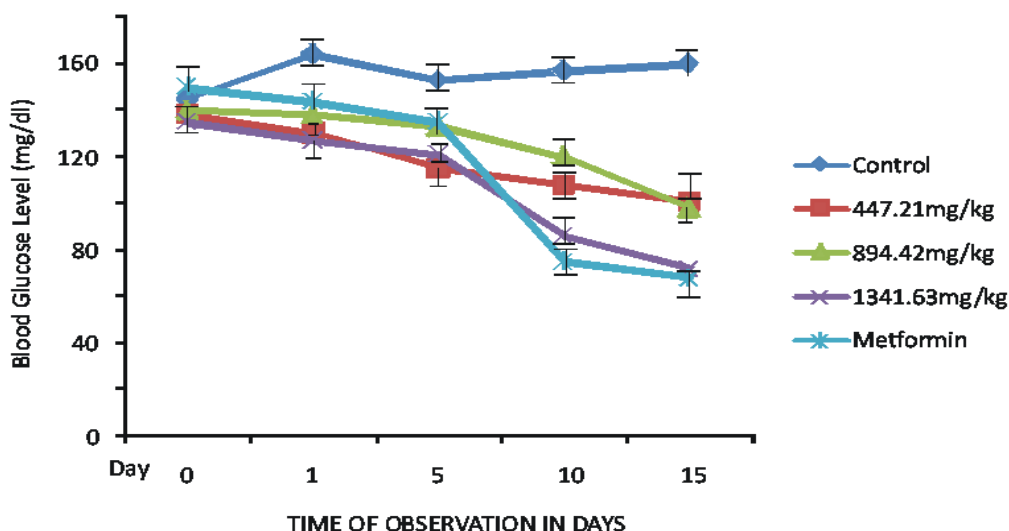
rats. The significant ($P < 0.05$) reduction in the blood glucose level (BGL) of the diabetic rats within the period of acute study was not comparable with that of the control (Table 1). However the highest reduction effect was achieved at 5hour period with the maximum dose of the extract (1341.63mg/kg) giving 83.21 ± 5.2 mg/dl. However the maximum dose less than that of the standard drug 65 ± 3.07 mg/dl metformin (Table 1).

The result of the prolonged treatment (15days) of *P. ostreatus* produced a sustained significant ($p < 0.05$) reduction in BGL of the hyperglycemic rats when compared with that of the control (Figure 1). The potent and progressive reduction activity of BGL of the extract with the pretreatment and treatment values confirms the anti-diabetic potential of *Pleurotus ostreatus*.

Table 1: Antidiabetic activity of *Pleurotus ostreatus* extract during acute study (BGL, mg/dl)

Treatment Dose (mgkg ⁻¹)	0h	1h	3h	5h	7h
5ml/kg saline water (control)	152±4.23	163±3.45	176±4.23	170±8.11	172±6.15
447.21	134±3.06	131±7.28	127±5.51	119±2.31	114±4.21
894.42	130±6.55	126±3.16	121±7.27	110±4.05	103±1.73
1341.63	125±5.64	118±9.30	108±3.54	83±5.20	90±3.51
10mg/kg/metformin (standard drug)	133±5.12	109±4.21	93±2.90	65±3.07	80±4.51

Data are expressed as mean + S. E. M. for five rats per group P<0.05 when compared to control.

**Fig. 1: Antidiabetic activity of *Pleurotus ostreatus* during prolonged study**

4. Discussion

The need for bioprospecting and development of ethnomedicinal plants as hypoglycemic agents is imperative especially now that most diabetic patients in Nigeria find it increasingly difficult to manage hyperglycemic conditions due to high cost of synthetic antidiabetic drugs with their subsequent side effects. This therefore supports this study.

The evaluation of *Pleurotus ostreatus* mushroom extract for its antidiabetic activity in alloxan induced diabetic rats

demonstrated a significant ($p < 0.05$) reduction in hyperglycaemia. The observed hypoglycaemic effects of the mushroom could be implication as a result of the presence of saponins, alkaloids, flavonoids and cardiac glycosides present in the extract. This finding corroborates with the earlier reports of Okokon *et al.* [11] and Odoemena *et al.* [2] on the roles of some of the phytochemical compounds inherent in such plants. These constituents may in part be responsible for the observed significant activity of

this extract either singly or in synergy with one another. The observed reduction in BGL of the diabetic rats by metformin in this study shows a severe state of diabetes. In this study, continuous administration with the mushroom extract for a period of 15 days caused a significant decrease in Blood glucose level of the treated rats compared to untreated diabetic ones. The ethanol extract of *P. ostreatus* reduced the glucose level in the rats to 53.6% in a prolonged treatment study.

Some plant extracts have been reported to exert hypoglycemic action by potentiating the insulin effect, either by increasing the pancreatic secretion of insulin from the cells of islets of Langerhans or its release from bound insulin [12] or corrections of insulin resistance [13]. Another possible mechanism of glucose reduction utilized by the mushroom extract may be due to the action of the extract through extra-pancreatic mechanism by inhibition of hepatic glucose production [1, 13]. Some workers have reported that consumption of fruits and nuts such as apple and walnut, (*Tetracarpidium conophorum*)

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are associated with a lower risk for diabetes [2]. The result of this study clearly indicates that continuous consumption of *Pleurotus ostreatus* mushroom, will always lower the BGL of a diabetic patient and therefore is recommended as a dieting menu for hyperglycemic patients.

5. Conclusion

The significant lowering of blood glucose level shown in the alloxan-induced diabetic rats in this study is good manifestation to show that *Pleurotus ostreatus* is an effective antidiabetic regimen. This result has given credence to the use of the mushroom as a menu for diabetic patient under dieting. Further studies are hereby recommended for the isolation and characterization of the active principles responsible for the action as well as the elucidation of the mechanism of the action.

Conflict of interest statement

We declare that we have no conflict of interest. The authors alone are responsible for the content and writing of the paper.

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