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Antihyperglycemic Effect Of Ethanol Extract Of Fruiting Bodies Of Organically Cultivated Pleurotus Ostreatus In High Sucrose High Fat Diet Streptozotocin Induced Diabetes In Rats

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Abstract – The effect of ethanol extract of the fruiting bodies of *Pleurotus ostreatus* (POE)and metformin Hydrochloride on the body weight and blood glucose profile of high sucrose high fat diet streptozotocin induced diabetic rats was investigated. The experimental model was 20% High Sucrose (HS) + 20% High Fat Diet (HFD) + 35mg/kg body weight (via intraperitoneal) streptozotocin (STZ) induced diabetic rat model, with the fruiting body ethanol extracts administered orally at 50, 150 and 300mg/kg .The Metformin HCl and ethanol extract was given once daily by intragastric gavage to the reference and experimental groups respectively at doses 150mg/Kg b.w., 50mg/Kg b.w, 150mg/Kg b.w. and 300mg/Kg b.w. respectively while the normal control received saline solution for 88days . POE caused significantly dose – ($p<0.05$) and time dependent reduction ($p<0.05$) in blood glucose levels of HS-HFD-STZ- induced diabetic rats. The blood glucose concentration of diabetic rats indicated a tendency to normal levels after administration of POE at 300mg/kg and 50mg/kg respectively but metformin HCl, (150mg/kg b.w,) showed a greater blood glucose level reduction effect in 9weeks of treatment. Ethanol extract of *Pleorotus ostreatus* significantly ($p<0.05$), dose and time dependently restored body weights and blood glucose of rats. Results highlighted show that ethanol extracts of organically cultivated mushroom has anti-diabetic properties, suggesting that people may use it in medicinal formulation processes for the management of diabetes mellitus and its associated complications.

Keywords – Body weight, Diabetes mellitus, *Pleurotus ostreatus,* blood glucose, anti-hyperglycemic effect).

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I. INTRODUCTION

Diabetes mellitus is a non-communicable disease which has been singled out as the most outstanding endocrine factor that brings about crisis in the metabolism of macromolecules such as carbohydrates, fats and proteins. Globally, diabetes mellitus has been considered a burden of disorder in the structure and function of biological systems (Lozano *et al.,* 2010). Truly, diabetes mellitus may not be responsible for all deaths connected with non-communicable diseases, yet, when it is not treated properly, may be major contributor to deaths related to degenerative diseases. These diseases include heart disease, chronic kidney disease, liver disease and diseases of large and small blood vessels etc. The World Health Organization (WHO, 2016),highlighted diabetes mellitus as a major reason for impaired vision, stroke, kidney failure and heart attacks as well as the amputation of the legs.

Whiting *et al.*,(2011),indicated that there is already a prediction that the prevalence of diabetes will likely rise to 300million people by 2025. In 2016, the World Health Organization (WHO, 2016) also highlighted that the prevalence of diabetes has been rapidly on the increase particularly among middle- and low- income nations. Mathers and Loncar (2006) had already indicated that WHO highlighted diabetes to be the 7th leading cause of deaths by 2030. This implies that the disease presents a major challenge to researchers and health care systems around the world. Diabetes mellitus is a group of metabolic diseases of endocrine origin indicated when there is high glucose concentration in the blood over a prolonged period and large amount of sugar in urine detected because of complete or relative lack of insulin resulting from the impairment of insulin secretion, insulin action or both (WHO,2014). Its symptoms include osmotic diuresis which eventually results to excessive loss of water from tissues, increased thirst, hunger and high concentration of lipids in the blood (WHO,2013). When diabetes is not treated on time, it causes potential complications at acute level such as diabetic ketoacidosis and nonketotic hyperosmolar hyperglycemic state (Kitabchi *et al.,* 2009). On long term basis, further complications linked with the disease are stroke, heart disease, kidney failure, eye problems, and foot ulcers (WHO,2013).

Presently, the most common treatment employed for diabetes mellitus is insulin therapy .This treatment is supported using a lot of anti -diabetic agents such as sulfonylurea, biguanides, α- glucosidase and thiazolidinedione. Some of these man made drugs may not be effective when the disease seems chronic and at the same time, they may have side effects such as toxicity to the liver, abnormal increase in size of the abdomen, discomfort of the gastrointestinal tract and weight addition (Lee *et al.,* 2012). Recently, nations of the world are developing huge interest on the making of oral blood sugar reducing agents from medicinal plants as well as medicinal macro fungi which must be safe, effective and without side effects. Even the World Health Organization has strongly given recommendation for such drugs (Singh *et.,al.,* 2007)

For many years, humans have utilized mushrooms as source of feeding and for the purpose of healing (Maria *et al.,* 2014). Mushrooms have been taken as small medicinal factories that nature has made. They have been revealed to be rich in immense array of new constituents that humans are yet to tap (Guggenheim, Wright and Zwickey, 2014). At the present time, people use different solvents to extract chemical substances from mushrooms. They trade these substances as supplements in diets because they believe that these substances have properties that may improve the human immune system and may also block the formation of cancer tumor (Guillamon *et al.,* 2010). Mshigeni and Chang (2000) highlighted that people who are oriented towards health now enjoy new foods which come from mushrooms and these edible substances got from macro fungi make up the foods that are growing at a very rapid rate around the globe. According to Mshigeni and Chang(2000),human health challenges as a matter of urgent need in developing and developed nations can be addressed by encouraging the people to engage in massive cultivation of macrofungi.For a period of over ten years ,the interest of people on finding the pharmacological effectiveness of mushroom has risen in an exponential manner (Ferreira *et al.,* 2010).Mushrooms have been indicated to be a good source of numerous compounds which possess nutritional as well as medicinal benefit (Pereira *et al.,* 2012). The term mushroom nutraceutical refers to chemical substances that can be extracted from the mycelium of mushroom or its fruiting body which may be highly nutritious or of medicinal or pharmacological importance (Chang and Buswell, 2008).

There has been a very big sudden strong rise in activities related to the function of mushroom products for medicinal purposes up-to-date (Chang and Buswell, 2008).The hectic work that people do these days is now resulting to great stress to the human body. Stress may bring about a weakening of the immune system and this may lead to numerous new diseases. The reason for the development of these new diseases is because the human body now lacks the capacity to naturally offer resistance to such diseases since immunity has been drastically reduced. Scientific reports have shown that consuming mushrooms as functional foods may help in the treatment of degenerative diseases. Extracts from mushrooms may also contain important bio chemicals

which may serve as supplements in diets to build body immunity and increase body resistance to degenerative medical conditions. These chemical substances extracted from mushrooms possess defined pharmacological and medicinal properties which differ from the man-made drugs that people use today. This is simply because these chemical substances are of very low toxicity, even when people take them at high doses. The use of mushrooms as agents of restoration for a long time is now an outstanding proof that macro fungi can as well greatly improve human health to optimal level (Chang & Buswell, 2008). The use of radiation and chemotherapy in the treatment of diseases may bring about side effects but these side effects may be reduced or eliminated using bio active substances in some mushrooms by a process of cell regeneration mediated through these mycochemicals (Liu, 1999).

The entire globe houses about14, 000 – 15,000 kinds of mushrooms. Out of this range of mushrooms, about1800 of them have been assumed to possess rich pharmacological features yet to be tapped by scientists for health purposes (Miles & Chang, 1997). However, it is an uphill task to differentiate between edible and medicinal mushrooms. The reason is because many of the common macro fungi that are consumable have curative properties and several mushrooms used for therapeutic purposes are also comestible (Guillamon *et al.,* 2010). However, one can use modern research methods through the employment of analytical techniques applied as a scientific base to establish these empirical observations made for the past years about mushrooms (Chang, 2006a).

There are many medicinal functions exhibited by mushrooms. These functions may include treatment of diabetes, enhancement of immunity, management of diseases linked to virus as well as cancer related cases. Mushrooms have antioxidant roles, protect the heart and its associated blood vessels and protect the liver cells. They can as well play detoxification roles in biological systems. When mushrooms are consumed, they may prevent inflammatory diseases. They may also block the formation of cancer tumor (Chang & Wasser, 2012).

The resource mushroom, *Pleurotus ostreatus* belongs the family of mushrooms known as pleurotaceae (Kuo, 2005). The people from Japan call *Pleurotus ostreatus* Hiratake which implies flat mushroom (Hall,2010).The Igbo-speaking people of South-East, Nigeria, call it Ero atakata because it has very tough texture on mastication (Akpaja *et al.,* 2003).

Pleurotus ostreatus is edible, medicinal and also very common. The mushroom has quality nutritional value, numerous medicinal properties and many other beneficial effects. It has been used as food and as means of treating ailments by numerous people all over the globe for many years (Finimundy *et al.,* 2013). *P. Ostreatus* is rich in dietary fiber, sterol, proteins, macro-minerals and trace-elements. The mycochemical composition of these macro fungi has made it a special dietary substance for the prevention and treatment of medical conditions associated with high level of cholesterol in the blood (Hossain *et al.,* 2003). It has also been reported that the macro fungi, due to the presence of mychochemicals in them coupled with their antioxidative properties may be used to cure ailments associated with virus, bacteria, high cholesterol level in blood. It also has hematological characteristics as well as the capacity to enhance immune functions (Finimundy *et al.,* 2013)

It also provides important mineral nutrients such as selenium, potassium, magnesium, copper, calcium, vitamins like riboflavin, niacin, vitamin D, tocopherol, vitamin C, folic acid, vitamin K and dietary fiber to humans (Maria *et al.,* 2014).

The prevalence of diabetes mellitus is a serious threat to the well-being of humans now and in the future. In the bid to lowering the prevalence of this endocrine factor for metabolic crisis and its related health complications, this research was aimed at investigating antihyperglycemic effect of ethanol extract of fruiting bodies of organically cultivated *Pleurotus ostreatus* in high sucrose high fat diet streptozotocin induced diabetes in rats

Fig1: The resource fungi- *Pleurotus ostreatus*

II. MATERIALS AND METHODS

2.1 Materials

Pleurotus ostreatus fruiting bodies were obtained from the samples cultivated using organic supplements at the Research Unit Demonstration Farm of the University of Port Harcourt, Nigeria. The samples were dried and stored in tightly sealed containers for the research.

2.2 Preparation of *P.ostreatus* Ethanol Extract.

The dried macro fungi materials was pulverized with a manual grinder and weighed with an electronic balance to obtain a mass of 944g (ground dry weight sample) which was well packaged and labeled. Ethanol extraction was carried out at the Organic and Inorganic Pharmaceutical Chemistry laboratory**,** University of Port Harcourt. To every mass of 100g of the pulverized macro fungi material, 300ml of ethanol were used for soaking and the bottles were shaken intermittently. First filtration process was done using clean white cotton material already immersed into the ethanol .Second filtration was done using What man No.1 filter paper. The filtrate was concentrated using a rotary evaporator at a temperature of 55^{o}C and the concentrate was subjected to evaporation using a water bath regulated at a temperature of 55^{o}C until a dark brown paste which weighed 54.02g was obtained as extract. The paste was stored in a refrigerator until further experimental use. The percentage yield was 6.0% (w/v). This was calculated as follows:

$$\text{Extract percentage yield (\%)} = \frac{\text{weight of extract}}{\text{weight of dry ground power}} \times \frac{100}{1}$$

2.3 Preparation of High Calorie Density Diet

The high calorie density diet was prepared using normal animal diet, sucrose and lard in the combination ratio of 3:1:1. The basic composition of the High Sucrose-High Fat Diet (HS-HFD) is shown in table 1.

Table 1: High Sucrose-High Fat Diet (HS-HFD %)

Composition	Proportion (%)
Normal diet	60.0
Sucrose	20.0
Lard	20.0
Total	**100.0**

Note: Diet was prepared daily to avoid microbial contamination and fed to the animals ad libitum, throughout the period of the experiment.

2.4 Induction of Diabetes and Determination of Blood Glucose and Body Weights of Rats.

After acclimatization of the animals for a period of 7 days, the nine animals in Normal group were placed on normal diet of guinea growers mash diet while the other rats in the remaining five groups (n=9) were fed with High Sucrose-High Fat Diet (HS-HFD) throughout the experimental period. The forty-five rats (n=9 rats/group) in the other five groups were placed on HS – HFD for 21 days, fasted overnight and induced diabetes using a single intraperitoneal injection of streptozotocin (35mg/kg bw). Stroptozotocin (Sigma, USA) at a dose of 35mg/kg bw was prepared in fresh and cold normal saline solution and administered immediately to the animals. The animals were first weighed using an electronic scale (TH 500) and their base line fasting blood glucose level taken using Fine Test Auto-coding Premium Blood Glucose Monitoring System and Blood Glucose Strips via tail vein cut before they were injected with streptozotocin. The animals in normal control group were injected normal saline alone. After 72hr of streptozotocin administration, the rats were again fasted and blood collected via tail cutting and their fasting blood glucose level were tested which confirmed hyperglycemia. Treatment of the animals with the ethanol extracts of *Pleurotus ostreatus* cultivated by substrate organic supplementation and Metformin HCl reference drug was done immediately after the last streptozotocin injection. Blood samples were drawn after 3rd week, 6th week and 9th week of commencement of treatment during the study. The extracts and metformin HCl (reference drug) were kept in plastic bottles with cap tightly sealed before and after each use, stored in the refrigerator, protected from direct sunlight to prevent spoilage throughout the time of animal treatment.

2.5 Experimental Animals

A total of 54 normoglycemic female wistar albino rats weighing150-180g were used for this research. The animals were purchased from the Animal House, Department of Biochemistry, Faculty of Science, University of Port Harcourt, Port Harcourt , kept and maintained in a house that is well-ventilated, having a 12hour light / 12hour dark cycle in propylene cages, at room temperature. Food and water were adequately given to the animals till the experimental research commenced. The animals were acclimatized to laboratory conditions, 7days prior to starting of experiment.The institutions' guide on the care and use of laboratory animals was followed

Animal experiments were carried out in compliance with applicable laws and regulations. Metformin HCl and the extracts were given (1ml per animal) once daily by intragastric gavage to the experimental groups undergoing treatment while the normal control group were given normal saline only (1ml per animal) once daily. Blood sugar was checked after 3,6 and 9 weeks of treatment.

Table 2 Experimental design for the antidiabetic screening.

S/N	ID	Treatment
1.	Normal control	Normal saline solution + Basal Diet
2.	Diabetic control	Sucrose (20%) + HFD (20%) + Basal Diet (60%) + STZ (35mg/kgbw) + H_2O
3.	Diabetic + *Pleurotus ostreatus* treatment 50mg/kg bw (POE_{50})	Sucrose (20%) + HFD (20%) + Basal Diet (60%) + STZ (35mg/kgbw) + POE (50mg/kgbw) + H_20/DMSO
4.	Diabetic + *Pleurotus ostreatus* treatment 150mg/kgbw (POE_{150})	Sucrose (20%) + HFD (20%) + Basal Diet (60%) + STZ (35mg/kgbw) + POE (150mg/kgbw) + H_2O/DMSO
5.	Diabetic + *Pleurotus ostreatus* treatment 300mg/kgbw (POE_{300})	Sucrose (20%) + HFD (20%) + Basal Diet (60%) + STZ (35mg/kgbw) + POE (300mg/kgbw) + H_2O/DMSO
6.	Diabetic + reference treatment metformin HCl treatment 150mg/kgbw(MET_{150})	Sucrose (20%) + HFD (20%) + Basal Diet (60%) + STZ (35mg/kgbw) + metformin HCl (150mg/kgbw) + H_2O

Statistical Analysis

Experimental data were statistically analyzed by a one way analysis of variance (ANOVA) using SPSS/PC + package. Multiple comparisms of differences between means were conducted by using Fisher's Least Significance Difference(LSD). Significance was accepted at a p-value of less than 0.05 ($p<0.05$).

III. Results

3.1 Effect of ethanol extract of *Pleurotus ostreatus* on body weight in Experimental Groups

The body weights of rats in the six groups were checked during the experimental period of 88 days. As shown in table 3, the body weights of the rats in all the groups were not significantly different ($P>0.05$) on day one before the high sucrose-high fat diet (HS-HFD) treatment to other groups except the normal group. On day 22 (21 days after HS-HFD treatment), there was increase in the body weights of the rats that received HS-HFD but the body weights of these groups were not significantly different ($P<0.05$) from that of the normal group. On day 25 (72 hours after STZ administration), the STZ induced diabetic rats exhibited significant decrease in body weights D group (149.50±1.24g), D+POE_{50} (150.37±11.40g), D + POE_{150} (154.43± 5.4g), D+POE_{300} (158.77±6.79g), D+MET_{150} group(152.50±3.91g) before the initial treatment of POE or metformin HCl ($p<0.05$),as compared to the N group (181.73±6.79g). On day 46 (3rd week of POE and metformin HCl treatment), the body weights of rats in D group (164.17± 5.24g) were lower than those in other groups: D + pOE_{50} group (177.30 ± 9.64g), D + POE_{150} group (174.90±10.43g), D + POE_{300}(187.13±5.06g), D + MET_{150}(182.60 ±4.47) ($p<0.05$), but the body weights of the treated groups were still lower than that of the normal rats (197.20±6.06) ($P<0.05$).

On day 67 (6th week of treatment), there was a significant increase ($P<0.05$) in the body weight of the treated groups compared to that of the diabetic group but the body weights of the treated groups were not significantly different ($P>0.05$) from that of the normal. On the 9th week of POE and metformin HCl administration, the body weights of D + MET_{150} group (244.80±10.62g) was greater than those of the other treated groups although the body weights of both POE and metformin HCl treated groups were much higher than those of the diabetic group ($P<0.05$).POE caused a significant dose dependent($p<0.05$) and time dependent ($P<0.05$) increase in the body weights of the diabetic rats while STZ administration negatively affected the body weights of the non-treated diabetic rats. At all the doses of POE (50mg/kg bw, 150mg/kg bw, 300mg/kg bw) administration, as well as metformin HCl, 150mg/kg bw, there was a significant increase in the body weight ($P<0.05$) of all the treated groups compared to the N group in 88 days of experimental research on the animal subjects.

Table 3 Changes in the body weight(s) of rats in the six groups during the experimental period of 88days.

	Day 1 (before HS/HFD treatment)	Day 22 (21 days after HS-HFD treatment)	Day 25 (72hr after STZ administration)	Day 46 (3rd week of treatment)	Day 67 (6th week of treatment)	Day 88 (9th week of treatment)
N	$152.03±7.45^{a}$	$173.17±6.01^{a}$	$181.73±6.79^{a}$	$197.20±6.06^{a}$	$216.57±1.85^{afgh}$	$232.13±6.06^{aefg}$
D	$149.47±2.10^{a}$	$166.50±3.55^{a}$	$149.50±1.24^{b}$	$164.17±5.24^{bd}$	$181.93±6.15^{bfgh}$	$192.13±9.03^{c}$
D+POE_{50}	$150.03±1.59^{a}$	$168.43±8.31^{a}$	$150.37±11.40^{b}$	$177.30±9.64^{bcd}$	$215.33±30.05^{adfgh}$	$214.73±14.43^{bdef}$
D+ POE_{150}	$150.47±0.97^{a}$	$171.90±4.22^{a}$	$154.43±4.22^{a}$	$174.90±10.43^{bd}$	$205.6±14.44^{abdfgh}$	$224.00±7.13^{adef}$
D+POE_{300}	$153.63±7.30^{a}$	$171.80±10.88^{a}$	$158.77±6.58^{b}$	$187.13±5.06^{acd}$	$203.57±10.81_{abdfgh}$	$228.70±7.92^{adefg}$
D+MET_{150}	$151.90±3.87^{a}$	$174.73±6.94^{a}$	$152.50±3.91^{b}$	$182.60±4.47^{bcd}$	$205.80±2.25^{abdfgh}$	$244.80±10.62^{afg}$

Values are means ± SD for 9 rats in each group of triplicate determination Values in the same column with different superscripts are significantly different at $p<0.05$

Values in the same column with different superscripts are significantly different at $p<0.05$

3.2 Effect of Ethanol extracts of *Pleurotus ostreatus* on Blood Glucose Profile of Rats

The HS-HFD-STZ- induced diabetic rats exhibited a significant increase in fasting blood glucose, D group (321.60±46.95mg/dL), D+ POE_{50} group (210.60±61.41mg/dL), D+POE_{150} (320.40±52.20mg/dL), D+ POE_{300} group (210.60±113.16), D+ MET_{150} group (233.40±40.46) as compared to non- HS-HFD-STZ-treated rats (74.93±15.04mg/dL) before the initial treatment of POE or metformin HCl ($P<0.05$).

After POE and metformin HCI treatment, the changes of blood glucose levels in different experimental groups are shown in table 4. After the 3 weeks of POE and metformin treatment (i.e, on Day 46 from Day 25 of diabetes establishment), the level of blood glucose in D group (219.00±29.60mg/dL), D+POE_{50} (133.80±22.51mg/dL), D+POE_{150} group (133.80±28.71mg/dL), D+POE_{300} group (115.80±25.22mg/dL), D+MET_{150} group (154.53±27.61mg/dL) were higher than N group (79.73±9.87mg/dL) ($P<0.05$). Also, on Day 46 of POE and metformin treatment, the blood glucose levels of D+POE_{300} group (115.80±25.22mg/dL), D+POE_{150}(133.80±28.71mg/dL),D+POE_{50} group(154.53±27.61mg/dL), were lower than the D group (219.00±29.60mg/dL) ($P< 0.05$) and POE at the highest dose (300mg/kgbw) had the greatest effect. On day 67 (i.e. after the 2nd three weeks of treatment), the administration of POE or metformin HCl in D+POE_{50} group (132.60±8.51mg/dL), D+POE_{50} group (132.60±8.51mg/dL), D+POE_{150} group (129.60±17.17mg/dL), D+POE_{300} group (122.40±40.89mg/dL), D+ MET_{150} group (106.80±19.25mg/dL) caused a significant decrease in blood glucose levels when compared with D group (222.60±41.41mg/dL) ($P<0.05$). However, all POE-treated groups retained high blood glucose (>100mg/dL) after the first three weeks and second three weeks of extract administration.

On day 88, the concentrations of glucose in the blood of the rats treated with POE and metformin HCl, D+POE_{50}group (100.00mg/dL), D+POE_{150} group (111.40±17.82mg/Dl), D+POE_{300}group (102.53±17.18mg/dL), D+MET_{150} (92.40±19.91mg/dL) were not significantly different from N group (79.8±16.72) ($P>0.05$) but these values were significantly different from the blood glucose level of D group (287.40±139.83mg/dL). POE caused a significantly dose- ($P<0.05$) and time dependent reduction ($p<0.05$) in blood glucose levels of HS-HFD-STZ-induced diabetic rats. The blood glucose concentration of diabetic rats indicated a tendency to normal levels after administration of POE at 300mg/kg bw and 50mg/kg bw respectively but metformin HCl, 150mg/kg bw showed a greater blood glucose level reduction effect in 9 weeks of treatment (day 88).

Table 4 : Effect of ethanol extract of the fruiting bodies of *Pleurotus ostreatus* on blood glucose levels (mg/dL) of rats in the six groups during the experimental period of 88days.

Treatment group	Day 22 (before HS/HFD treatment)	Day 25 (72hr after STZ treatment)	Day 46 (3rd week POE treatment)	Day 67(6th week POE treatment)	Day 88 (9th week POE treatment)
N	73.2±9.24^{a}	74.93±15.04^{b}	79.73±9.87^{b}	84.60±4.76^{c}	79.8±16.72^{b}
D	70.80±2.75^{a}	321.60±46.95^{a}	219.00±29.60^{a}	222.60±41.41^{a}	287.40±139.83^{a}
D+POE_{50}	68.40±17.17^{a}	210.60±61.41^{a}	133.80±22.51^{ca}	132.60±8.51^{bd}	100.00±9.90^{b}
D+ POE_{150}	75.60±10.80^{a}	320.40±52.20^{a}	133.80±28.71^{da}	129.60±17.17^{cd}	111.40±17.82^{b}
D+POE_{300}	67.80±22.36^{a}	210.60±113.16^{a}	115.80±25.22^{ba}	122.40±40.89^{cd}	102.53±17.18^{b}
D+MET_{150}	77.40±12.60^{a}	233.40±40.46^{a}	154.53±27.61^{ea}	106.80±19.25^{cd}	92.40±19.91^{b}

Values are means ± SD for 9rats in each group. of triplicate determinations.

Values in the same column with different superscript are significantly different at $p<0.05$

IV. Discussion

The intention of this research work was to investigate the effects of *Pleurotus ostreatus* ethanol extract on body weight and blood glucose in diabetic animal model. High sucrose-high fat diet-streptozotocin induced diabetic rats were used as a major experimental model as it established diabetes properties in obese animals [26](Lenzen, 2008). Metformin HCl was used as a reference treatment because it is well established as one of the standard drugs used for the management of diabetes mellitus. Biochemical parameters used to justify the extract therapeutic effects were monitored at 3 weeks, 6 weeks and 9 weeks intervals. In this study, Table 1. Indicates the changes in the body weight of normal and HS-HFD-streptozotocin-induced diabetic rats.

HS-HFD-streptozotocin induced diabetes mellitus [26](Lenzen, 2008) highlights features such as lack of insulin, high level of blood sugar, increased triglycerides, high plasma total cholesterol and reduced HDL-cholesterol. Diabetes mellitus caused by the administration of streptozotocin is associated with loss in body weight. The loss in body weight may be due increase in muscle wastage coupled with loss of tissue proteins [27](Swanston-Flatt *et al.,* 1990). The induced diabetic rats exhibited significant decrease in body weight but treatment with POE and metformin HCl brought about a reverse in the trend by causing a dose and time dependent increase in the body weights of the diabetic rats while STZ administration negatively affected the body weight of the non-treated diabetic rats. The improvement in the body weights of the treated animals showed that the extracts prevented muscle tissue damage caused by the condition of high blood sugar level. However, it was observed that the body weights of rats on metformin treatment were higher than those on POE treatment on the 9th week indicating that metformin had greater positive effect on reducing blood sugar than the extracts at all doses. Diabetes mellitus induced by high sucrose-high fat diet and low STZ injection [26](Lenzen, 2008) has the hallmark of insulin insensitivity,relativelack of insulin, high level of blood sugar. In this study, high sucrose-high fat diet and low STZ injection at 35mg/kg caused diabetes after 72 hours. This may be due to insulin insensitivity and the destruction of the β-cells of the islets of Langerhans [28](Kavalali *et al.,* 2003). The ethanol extract (POE) used in the treatment of the animals caused a significant dose and time dependent reduction in blood glucose levels of the diabetic rats particularly the rats that were given POE at 300mg/kgbw and 50mg/kgbw respectively. Table 4.15 indicated that their blood glucose levels were tending towards normal even more than metformin after 9 weeks. This shows that the fruiting bodies of *Pleurotus ostreatus* may be used in the management of diabetes mellitus. This study has therefore revealed that *Pleurotus ostreatus* may have hypoglycemic effect on diabetic rats. The reason why plasma blood glucose reduced may be either due to a rise in plasma insulin level in the diabetic rats which may stimulate the β-cells of the islets of Langerhans to secrete pancreatic insulin. Another reason may also be because the transport of blood glucose to peripheral tissues may have been improved due to the enhancement of insulin sensitivity. This finding is in line with the work of [29]Agrawal *et al.,* (2010) who examined *Pleurotus ostreatus* and indicated that it has beneficial effect on sugar control and lipid composition. The hypoglycemic effects of *Pleurotus ostreatus* extracts may be due to the presence of dietary fiber. Dietary fiber has been revealed by science as an agent that reduces glucose absorption. It also decreases the rate of gastric emptying [30](Chen and Raymond, 2008) and binds to cholesterol to eliminate it, hence the mushroom is good for diabetic patients. The presence of thiamine HCl in *Pleurotus ostreatus* could also be a reason for the reduction of blood sugar level in the diabetic animals that were treated since thiamine acts as an essential cofactor in glucose metabolism [31](Arora *et al.,* 2006) and therefore may modulate diabetic complications by controlling glycemic status in diabetic patients [32](Thornally, 2005). Impaired glucose tolerance [32](Thornalley, 2005) and β-cells malfunction has been indicated in the lack of thiamine. The presence of terpenoids [33](Keeling and Bohlmann, 2006) or alkaloids [34](Qiu *et al.,* 2014) may also be the reason for the reduction in blood glucose level on administration of *Pleurotus ostreatus* ethanol extracts since these bioactive compounds are known as mycochemicals with anti-hyperglycemic properties.

V. CONCLUSION

In summary, ethanol extract of organically cultivated *Pleurotus ostreatus* possesses antihyperglycemic activities in HS-HFD-Streptozotocin-induced diabetic rats suggesting that people may use it in medicinal formulation processes for the management of diabetes mellitus and its associated complications.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest

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