Hypoglycemic Effect of Olive Oil (*Olea europea*) on Alloxan- Induced Diabetic Albino Rats.

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ABSTRACT

This research work is aimed at investigating the hypoglycemic effect of olive oil obtained from the seeds of olive tree (Olea europea,) on alloxan-induced diabetic Albino rats. The results obtained show that olive oil (Olea europea) exhibit hypoglycemic effect on alloxan-induced diabetic Albino rats with the lower dose (150mg/kg/b.wt). as the most effective dose for the significant treatment of type 1 diabetes. Medium and High doses however showed an insignificant effect compared to the control groups. When the, standard drug (positive control) 60 mg/kg/b.wt was administered to the rats , the glucose level of the group 2 alloxan-induced diabetic rats decreased gradually to the range between the normal range 4.9-5.5 mg/dl according to the world health standard (W.H.O, 2014). During treatment, the body weight of the rats was observed to increase as the rats responded to the hypoglycemic effect of the oil treatment. As the treatment continues, the rats look healthier which shows a positive response to the olive oil treatment.

Key words : Hypoglycemic effect, Alloxan- induced Diabetes, Albino rats, Standard Drugs.

INTRODUCTION

Background of Study

Diabetes mellitus is probably the fastest growing metabolic disorder in the world, and it is a major source of morbidity in developed countries. Medicinal plants play an important role in the management of diabetes mellitus, especially in developing countries where resources are meager. Many studies have confirmed the benefits of medicinal plants with hypoglycemic effects in the management of diabetes mellitus. The effects of these plants may delay the development of diabetic

complications and correct the metabolic abnormalities[1].

The olive, known by the botanical name *Olea europea* meaning European olive, is a specie of small tree in the family *Oleaceae*, found traditionally in the Mediterranean basin. The species is cultivated in all the countries of the Mediterranean, such as Australia, New Zealand, North and South America, South Africa and some Arabian countries. *Olea europea* is the type of specie for the genus *Olea*[2].

The Olive's fruits also called olive is of a major agricultural importance in the Mediterranean region as it is a source of olive oil, it is one of the core ingredients in the Mediterranean cuisine. The tree and its fruits give their name to the plant family, which also includes species; like Lilacs, Forsthythia, and the true trees (Fraxinus): It is also known to have varieties of health benefits[3].

During the past few years some of the new bioactive drugs isolated from hypoglycemic plants showed anti-diabetic activity with more efficacy than oral hypoglycemic agents used in clinical therapy. Presently, there is an increased demand to use natural products with anti-diabetic activity due to the side effects associated with the use of insulin and oral hypoglycemic agents[4]. More than 400 plants with glucose lowering effect are known. Also, a number of plants have a hypolipidemic effect. However, there is little information about plants with both hypoglycemic and hypolipidemic effects[5].

The pathogenesis of diabetes mellitus is managed by insulin and oral administration of hypoglycemic drugs such as sulfonyl urea and biguanides. Unfortunately, apart from having a few side effects, none of the oral synthetic hypoglycemic agents have been successful in maintaining and controlling long-term micro vascular and macro vascular complications. Insulin therapy is used for management of diabetes mellitus but there are several drawbacks, which include insulin allergy, insulin antibodies, lipodystrophy, autoimmunity, and other delayed complications like morphological changes in kidney and severe vascular complications. Thus, new relatively non-toxic therapeutic agents are needed to treat hyperglycemia, which also would correct dyslipidemia to reduce the risk of cardio-vascular complications of diabetes[6].

Induction of diabetes in laboratory animals is a convenient and useful strategy in the understanding and treatment of the disease. An appropriate dose of streptozotocin was used to induce experimental diabetes. Streptozotocin selectively destroyed

pancreatic β-cells, resulting in hypoinsulinemia. Streptozocin-treated rats are often used as diabetic animals with insulin-deficiency resulting from damage of beta-cells caused by the drug. These rats are hyperglycemic and have reduced uptake of glucose in skeletal muscles[7].

Justification of the Study

There is increased risk of diabetes in Nigeria and need to provide alternative treatment plans. In 2000, according to the World Health Organization, at least 171 million people worldwide suffer from diabetes, or 2.8% of the population. Its incidence is increasing rapidly, and it is estimated that by 2030, this number will almost double. Olive oil has been known to have different health benefits like reducing the risk of stroke? reduce some forms of cancer, protects against depression. Hence, there is need to carry out anti-diabetic or hypoglycemic effect of the olive oil, to see whether it can lower blood glucose level.

Aim of the Study

The aim the research is to investigate the "hypoglycemic effect of *Olea europea* (olive oil) on alloxan induced diabetic Albino rat" The objectives of the research are as follows:

- To determine the Fasting Blood Glucose (FBG) level in normal rats fed with normal rat feed and water only.
- To determine the Fasting Blood Glucose (FBG) level in diabetic rats treated with standard drug, normal rat feed and water.
- To determine the Fasting Blood Glucose (FBG) level in diabetic rats treated with different doses of olive oil extracts, normal rat feed and water.
- To compare the results and establish whether olive oil extract exhibited hypoglycemic effect on diabetic albino rats.

MATERIALS AND METHODS

Experimental Animals

The albino rats of both sexes were collected from the animal house of biological sciences in Kaduna State University. Their body weight ranges from (160-280)g. They were collected and kept in a well-ventilated cage. The Albino rats were 25 in number, and they were transported to the animal house in Applied Biology Department of Kaduna Polytechnic. They were allowed to acclimatize for a period of two weeks to its new environment.

Animal Grouping(s)

The rats were grouped into five groups of 5 rats per group as follows:

Group 1: Normal control (albino rats were fed with normal rat feed and water *ad libitum*

Group 2: Positive control (Diabetic induced albino rats were fed with normal rat feed and water and then treated with standard drug (Diabetamin 60mg/kg/b.wt)

Group 3: Diabetic induced albino rats were fed with normal rat feed and water, and then treated with lower dose of olive oil (150mg/kgb.wt)

Group 4: Diabetic induced albino rats were fed with normal rat feed and water, and then treated with medium dose of olive oil (300mg/kgb.wt)

Group 5: Diabetic induced albino rats were fed with normal rat feed and water, and then treated with high dose of olive oil (450mg/kgb.wt)

Extraction of Oil

The plant seeds of *Olea europaea* were collected from a chemist in Tudun Wada Kaduna and was identified by a botanist of Applied Biology Department of Kaduna Polytechnic, though no voucher number, since voucher number is not normally given for seeds. They were preserved and properly sealed since they are not grown in Nigeria. *Olea europaea* (olive) are grown in regions with hot weather, and its native range is from the Mediterranean regions. When the seeds were obtained, the seeds were air dried and were pounded to fine powder using a mill grinder and the powdered material was weighed. Soxhlet extraction method was used for the oil extraction. The weight of the powdered sample was recorded as W_1. An empty clean beaker weighed like W_2. Few anti bumping granules were added into 500cm^3 round bottom flask and 300cm^3 of N- hexane was added into the flask. The powdered plant material (200g) was added into an extraction thimble, and the thimble was fixed into a Soxhlet unit. The round bottom flask and a condenser were connected to the Soxhlet extractor, and cold-water circulation was put on. The heating mantle was switched on and the heating rate adjusted until the solvent is refluxing at a steady rate. Extraction was carried out for 6 hours. The solvent was recovered, and the contents of the flask was poured into the already weighted empty beaker. The flask was rinsed with N-Hexane. The solvent was dried in an oven at 70^{0}C for 1hr, until all the solvent evaporates leaving behind an oil residue. The beaker and oil residue were weight as W_3.

. The oil was collected and subsequently used for the experiment.

Induction of Diabetes

Diabetes mellitus was induced in rats by single intraperitoneal injection of 150mg/kg Alloxan monohydrate. 0.5g was dissolved in 10mls of normal saline in a clean beaker after an overnight fast. Induction of diabetes mellitus was confirmed by the presence of high fasting plasma glucose level which was determined 24hours after alloxan administration. Rats with fasting blood glucose higher than 2g/dl were considered diabetic. Body weight, fluid intake and food consumption patterns were monitored at onset and during the 2 weeks' experimentation period.

Animal Feeding /Housing System

The Albino rats were fed with poultry feed (finishers) and the poultry feed was mixed with cereal chaff concentrates. After mixing, a little water was added, and the feed was molded into balls and was given to the rats in a clean container on a regular basis. The cans were ensured to with clean water which was changed regularly.

The inner part of the cages was filled with saw dust which was changed at intervals to maintain a clean environment for the rats.

Collection of Blood Sample to Examine the Blood Glucose Level

All blood samples were collected from the tail artery of the rats and their fasting blood glucose was determined after a 4hour fast Blood Glucose Monitoring system and their results were expressed as mg/dl. As well as their weight in gram(s).

Statistical Analysis

The results were expressed as Mean ± Standard Error of Mean (M ± SEM). Data obtained from biochemical studies were analyzed using one-way ANOVA for comparison between means for treated groups and control groups for statistical difference using SPSS Package for version 15. The values of $p < 0.05$ were considered as significant.

RESULTS AND DISCUSSION

Table 1 : Fasting Blood glucose level of diabetic induced albino rats treated with different doses of olive oil for 14days, and the control groups.

Grouping(s) of Albino Rat	Day 1 (mg/dl)	Day 4 (mg/dl)	Day 8 (mg/dl)	Day 12 (mg/dl)	Day 14 (mg/dl)
Group 1 (Normal Control)	4.90 ± 0.28	4.56 ± 0.44	4.66 ± 0.30	4.60 ± 0.21	4.53 ± 0.16
Group 2 (Positive Control) 60mg/kg/b.wt	9.23 ± 1.86	7.96 ± 2.21	5.46 ± 0.14	5.46 ± 1.55	4.87 ± 1.29
Group 3 (Lower Dose of oil) 150mg/kg/b.wt	8.73 ± 0.75*	8.26 ± 0.79*	8.10 ± 1.72*	7.16 ± 0.66*	6.56 ± 0.34*
Group 4 (Medium dose of oil) 300mg/kg/b.wt	9.16 ± 1.85	7.83 ± 0.21	5.93 ± 1.47	5.36 ± 1.05	4.90 ± 0.97
Group 5 (Higher dose of oil) 450mg/kg	7.83 ± 2.85	5.66 ± 1.21	4.76 ± 1.47	5.10 ± 1.00	4.70 ± 0.80

Values are expressed as Mean ± SEM, n = 3 * Significantly different from the control at p > 0.05

Table 2: Body weights of the albino rats obtained within 12 days of the experiment

Grouping(s) of Albino Rat	Day 1 (g)	Day 3 (g)	Day 6 (g)	Day 9 (g)	Day 12 (g)
Group 1 (Normal Control)	199.59 ± 27.56	199.92 ± 21.33	198.39 ± 27.63	200.43 ± 12.65	209.49 ± 12.57
Group 2 (Positive Control) 60mg/kg/b.wt	188.32 ± 6.45	188.48 ± 12.87	183.409 ± 6.88	190.28 ± 18.06	195.09 ± 17.68
Group 3 (Lower Dose of oil) 150mg/kg/b.wt	232.79 ± 31.88*	222.60 ± 39.69*	231.78 ± 19.10	256.10 ± 47.05	262.36 ± 46.75
Group 4 (Medium dose of oil)	181.65 ± 17.42	182.01 ± 24.46	187.32 ± 23.36*	191.10 ± 28.00*	197.95 ± 23.10*

300mg/kg/b.wt								
Group 5 (Higher dose of oil) 450mg/kg	195.63 ± 14.56	196.78 ± 18.03	199.47 23.52	±	199.98 24.16	±	201.45 21.56	±

Values are expressed as Mean ± SEM, n = 3 *Significantly different from the control at p > 0.05

Discussion

Table 1 shows that the olive oil used for the treatment in lower dose (i.e 150 mg/kg/b.wt) was observed to be significantly different when compared to the normal control glucose level and the positive control. In the case of the medium dose (i.e 300 mg/kglb.wt) it shows a slight hypoglycemic effect on the alloxan-induced rats as it is insignificantly different from the normal and positive control. The higher dose (i.e., 400 mg/kg/b.wt) shows insignificant difference when compared with the control (normal and positive control). This shows a more hypoglycemic effect on the alloxan-induced diabetic albino rats. When the standard drug (positive control) 60 mg/kglb.wt was used the glucose level of the group 2 alloxan-induced diabetic rats decreased gradually to the range between the normal range 4.9-5.5 mg/dl according to the world health standard (W.H.O, 2014)[8].

Table 2 shows the initial significantly lower body weight of the rats before the commencement of treatment. During treatment, the body weight of the rats was observed to increase as the rats responded to the hypoglycemic effect of the oil treatment. As the treatment continues, the rats look healthier, which shows a positive response to the olive oil treatment. From Table 2, the results shows that as the dose of olive oil increased, the animals body weights increased over the 12 days period. The effect of the oil treat is evidenced by the gain in body weight showing that the animals are more tolerant to the carbohydrate feed and the feed are better digested and retained for body growth and hence weight increase.

CONCLUSION

In conclusion, the study shows that olive oil (*Olea europeae*) could play a key role in the management of diabetes. however, an appropriate dosage is required according to the patients' body weight obtained. In addition , lower dose of the oil should be considered for the management of diabetes as it is the one with significant effect when compared with the control groups.

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