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Screening for the Hypoglycaemic Potentials of the Extract of *Vernonia amyggalina*

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Abstract

This work evaluates the hypoglycemic effect of *Vernonia amygdalina* on alloxan induced diabetic rats. Different doses of the aqueous leaf extract of *Vernonia amygdalina* (50mg/kg, 100mg/kg, 200mg/kg and 400mg/kg) were administered to alloxan induced diabetic rats under experimental conditions. The mean fasting blood sugar levels of the groups of diabetic rats increased significantly as it rose from basal blood sugar levels (mg/dl) of 38.0, 39.2, 35.2 and 35.8 to fasting blood sugar levels (mg/dl) of 277.6, 284.8, 256.4 and 265.6. The findings showed that there is a dose dependent reduction in fasting blood sugar in alloxan induced diabetic rats after treatment to 167.5mg/dl, 140.8mg/dl, 104.2mg/dl and 80.4mg/dl respectively.

Key words: *Vernonia amygdalina*, ethnomedicine, diabetes, alloxan.

Introduction

Diabetes mellitus is a diseased condition characterized by high glucose levels caused by an absolute or relative deficiency of insulin, a special sugar regulatory hormone produced by the pancreas, a gland in the body. It can be defined on the basis of laboratory findings as a fasting venous plasma concentration greater than 140mg/dl or 210mg/dl two hours after a carbohydrate meal or to two hours after the oral ingestion of the equivalent of 75mg of glucose.

The disease (DM) is prevalent all over the world. It affects 11-20% of the European and United States of American white population. The highest risk was found in Pima Indians of Arizona and in the urbanized Micronesians of Nauru where up to one half of the population had diabetes (Kings and Rewers, 1991). DM can affect any age, sex, race or socioeconomic group. It was generally considered as disease of the developed countries but recent trends show that it is fast becoming a third world problem (Kings and Rewers, 1991). It has been found that 1-2.6% of the Nigerian population has DM (Kings and Rewers, 1991, Akinkugbe, 1992).

The causes of DM include poor diet, obesity, environmental factors, hereditary factors and drug

usage and symptoms are increased urine production, increased thirst, and sugar in blood, excessive eating, loss of water and loss of memory. Complications of DM include damage to the kidneys, impaired vision, hardening and blockage of blood vessels, infections and impotence (Famuyiwa *et al*,, 1988).

The advantages of medicinal plants over orthodox medicine are cheapness, relative availability, ease of preparation and absence of serious side effects. .It has been estimated that in Nigeria, 80% of rural people depend on herbs for the treatment of various diseases. Extracts of leaves of *Vernonia amygdalina* have been found to possess antimalarial activity against plasmodium (Masaba 2000, Abosi and Roseroka 2003) and active against sexually transmitted diseases (Kambizi and Afolayan, 2001). Chewing sticks of V. amygdalina has been found to have antibacterial activity (Taiwo *et al*., 1999) and water soluble anti cancer agents have been discovered from the plant (Izevbigie, 2003). This study was undertaken to determine the hypoglycemic effect of *Vernonia amygdalina* leave extract.

Materials and Methods

Materials

Fresh plants of *Vernonia amygdalina* were obtained from the International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria. The identification of the plant was carried out at the Herbarium of the Department of Botany and Microbiology, University of Ibadan, Ibadan, Nigeria.

The leaves of the plant were air dried in a shade and later grounded to a powdery form using a blender. 1g of the powdered herb was soaked in 100ml of distilled water to produce a concentration of 10mg/ml of the aqueous extract used for the experiment. The soaking was done overnight (twelve hours) in order to get the aqueous extract.

Experimental animals

Thirty healthy rats weighing between 150g and 250g were obtained from the Veterinary Physiology Animal House, University of Ibadan, Ibadan. Nigeria. Animal feeds were obtained from Ladokun Feed Mill Nigeria Limited, Ibadan. Nigeria.

Alloxan

Ten percent alloxan dissolved in physiological saline was used to induce the diabetic condition in the diabetic rats. This was by weighing 10g of fresh samples of alloxan on a Mettler's balance and dissolving in 100ml of 0.15M physiological saline solutions. The dose administered was a standard dose of 100mg/kg weight of the rats.

The general formula stated below was used to obtain the actual volume of the drug to be administered:

$$\text{Volume to be administered} = \frac{\text{Weight of animal (g) X Dose of drug (mg/kg)}}{\text{1000 X Concentration of drug (mg/ml)}}$$

The remaining extract was stored in a refrigerator at a temperature of -4 degrees centigrade.

Methods

The rats were divided into six groups, each containing five animals. The basal blood glucose levels

of all the rats were obtained after fasting them overnight. This was done before they were injected with alloxan to make them diabetic. The purpose of this is to know their initial blood glucose level before the commencement of the experiment so as to compare the results of the basal blood glucose level when they are not diabetic with the Fasting Blood Sugar (FBS) level when they are made diabetic and this was done by using the glucometer. The procedure involved cutting a bit of their tails for a drop of blood and dropping this blood on a specified space on the glucometer. The FBS result was given in mg/dl.

The rats in Groups 1 – 4 were injected with 100mg/kg alloxan intramuscularly and the Fasting Blood Sugar checked after 48hours.Those with FBS varying between 100 – 500 mg/dl were considered diabetic. These diabetic rats were then treated with 50mg/kg, 100mg/kg, 200mg/kg and 400mg/kg dose of the aqueous extract of Vernonia amygdalina twice daily respectively for two weeks. The administration was done orally with the aid of an oral cannula; the FBS was checked every two days and recorded.

The animals in Group 5 were not given alloxan to induce DM but were administered with 400mg/kg of the plant extract of Vernonia amygdalina and their FBS were also recorded every two days for two weeks. The last group (Group 6) was left as the control of the experiment. They were neither induced with the drug alloxan nor were they administered with the plant extract. They were only fed with the commercial feeds and given clean water daily. Their FBS were also checked every two days and compared with the FBS of the other groups. The whole group of animals offered a commercial feed prepared by Ladokun Feed Mill Nigeria Ltd and the animals were given clean water everyday.

Statistical Analysis

The results are expressed as mean of five readings. The students't-test was utilized to determine the significance of differences between control and treated groups. The level of significance was taken as $p< 0.05$.

Results and Discussion

Table 1 shows the mean values of the fasting blood sugar levels in different groups. After the injection of alloxan, the mean fasting blood sugar levels of the groups of diabetic rats increased significantly because it rose from basal blood sugar levels (mg/dl) of 38.0, 39.2, 35.2 and 35.8 to fasting blood sugar levels (mg/dl) of 277.6, 284.8, 256.4 and 265.6 after the injection of alloxan. This effect was observed two days after the injection of alloxan in the rats.

The mean values of FBS (mg/dl) of diabetic rats following the administration of different doses of *Vernonia amygdalina* in different groups of rats are as shown in Table 2. The results obtained revealed that there was a marked difference between the mean values of the FBS of group 1 which is 167.5mg/dl and the other groups. It took a longer period of time for the fasting blood sugar level of the rats administered with the plant extract *Vernonia amygdalina* to be reduced than it did for the rest of the other groups. Difference between the groups was significant ($p < 0.05$).

Results obtained in Table 3 revealed a significant decrease in the mean fasting blood glucose levels of the normal rats administered with 400mg/kg of *Vernonia amygdalina* which decreased from the mean fasting blood glucose level of 60.0mg/dl to 38.0mg/dl. This difference was statistically significant ($p < 0.05$).

All the rats injected with the alloxan became diabetic as their fasting blood sugar levels exceeded the normal range of 80-100mg/dl. The mean values of the fasting blood sugar of the various groups of diabetic rats were reduced after they were given the plant extracts. Treatment of the rats with *V amygdalina* showed a dose dependent relationship with the period of time it took for the blood of the rats to return to the basal blood sugar levels (Table 2). The longest time was noticed in Group 1 rats (those treated with 50mg/kg which was longest than 14 days. This was followed by Groups 2 and 3 rats. The shortest time of return to the normal blood sugar was seen in the Group 4 rats (those treated with the highest dose - 400mg/dl). It was observed that the blood glucose levels of the rats in group 1 reduced gradually and slowly as compared to the rats in the other groups and some of the rats were still diabetic after the experiment.

It has been seen in this study that *V amygdalina* has hypoglycaemic activity. It has been observed that the dose of the plant extract administered affected the duration of time it took to treat the diabetic condition. Further work needs to be done to ascertain the appropriate dosage and duration of administration. Considering the great antidiabetic properties of the plant, awareness should be created to promote the medicinal advantages of the plant.

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Table 1. Mean values of the Fasting Blood Sugar (FBS) levels in different groups.

	Group 1	Group 2	Group 3	Group 4
Basal Blood Sugar (mg/dl)	38.0	39.2	35.2	35.8
Fasting Blood Sugar after injection of alloxan (mg/dl)	277.6	284.8	256.4	265.6

Table 2. The mean values of Fasting Blood Sugar (mg/dl) in diabetic rats following the administration of different doses of *V. amygdalina* in different groups of rats.

Alternate days that fasting blood sugar was obtained	Group 1 (50mg/kg)	Group 2 (100mg/kg)	Group 3 (200mg/kg)	Group 4 (400mg/kg)
2nd	231.2	226.6	203.2	201.8
4th	237.4	200.0	151.6	94.0
6th	196.0	176.0	94.0	72.0
8th	163.4	132.0	89.0	62.0
10th	142.2	102.0	75.0	50.8
12th	110.4	80.0	61.0	44.0
14th	92.0	68.8	55.8	38.4
Mean FBS	167.5mg/dl	140.8mg/dl	104.2mg/dl	80.4mg/dl

Table 3. The mean values of Fasting Blood Sugar (mg/dl) of the normal (Group 5) rats and the control (Group 6) group rats taken on alternate days.

Number of Days	**Normal Group administered with 400mg/kg of *Vernonia amygdalina***	**Control Group**
2nd	60.0	44.0
4th	65.6	54.4
6th	54.4	46.0
8th	47.6	41.8
10th	42.2	41.0
12th	42.2	40.0
14th	38.0	40.0
Mean	50.0	43.9