EFFECTS OF CO-ADMINISTRATION OF ETHANOL EXTRACTS OF *GONGRONEMA LATIFOLIUM* AND *TAPINANTHUS BANGWENSIS* LEAVES ON BLOOD GLUCOSE LEVEL, BODY AND ORGANS WEIGHTS IN NORMAL MALE ALBINO WISTAR RATS

ABSTRACT

The effects of co-administration of ethanol extracts of *Gongronema Latifolium* and *Tapinanthus bangwensis* leaves on Blood glucose level, body and organs weights in normal male Albino Wistar rats were evaluated. Groups 1,2 and 3 of four animals each were treated respectively with 200 mg/kg, 300 mg/kg and 400 mg/kg (50:50 dose ratio), while group 4 that served as control was not treated. Each animal in the four groups were allowed free access to commercial rats mash and water for 3weeks. Blood glucose and body weights were determined on day 0, 3, 6, 9, 12, 15 and 21, while organs weights were taken as the animals were sacrificed at the end of the three weeks experimentation. On day 0, 3, 6 and 9 no statistically significant difference ($P<0.05$) was found in the blood glucose levels in the treated groups when compared with the control group. Significant decrease ($P<0.05$) was observed in the blood glucose in the treated groups 2 and 3 compared to the control on days 12, 15, 18 and 21. On day 0, 3 and 6, significant decrease ($P<0.05$) was recorded in body weight of animals in group 2 and 3 when compared to the control group. Groups 2 and 3 animals also recorded significant decrease ($P<0.05$) in body weight when compared to group 1. On day 9, significant decrease ($P<0.05$) was found in groups 2 and 3 when compared to the control and when compared to group 1. On days 12, 15 and 18, body weight in group 3 was significantly decreased ($P<0.05$) when compared to the control and to groups 1 and 2 when compared to the control and with group 1. Body weight on day 21 showed significant decrease ($p< 0.05$) in groups 2 and 3 when compared to the control and with group 1. Liver weight recorded significant decrease ($P<0.05$) in group 3 when compared to group 1. No significant difference ($P<0.05$) was found in the weight of the kidneys in all the treatment groups when compared to the control. The results of this work implies that combined extracts of *T. bangwensis* and *G. latifolium* leaves possess the potentials to reduce blood glucose level in the treatment groups of animals on prolonged treatment, with no marked effects of the extracts on the body and organs weights of the animals. Therefore, the two extracts could be used to manufacture medications that can take care of diabetic condition in human.

Keywords: *Gongronema latifolium*, *Tapinanthus bangwensis*, Blood glucose, Body weight, organs weight, co-administration.

INTRODUCTION

Medicinal plants are plants which contain substances that could be used for therapeutic purpose or which are precursors for the synthesis of useful drugs (Abolaji *et al*., 2007; Akpakpan, *et al*., 2012). Since time immemorial, medicinal plants have been used in virtually all cultures as a

source of medicine. Over 5000 plants are known to be used for medicinal purposes in Africa, but only a few have been described or studied (Evans, 2005). Natural products from plants can be another potent source for the discovery of excellent biological activities (Akpabio and Akpakpan 2012**;** Adebayo *et al*., 2010). Among these medicinal plants are *Gongronema latifolium* and *Tapinanthus bangwensis*.

Gongronema latifolium belongs to the family of Asclepideceae. It is a tropical rain forest plant found throughout Nigeria and other tropical countries such as Guinea-Bissau, Western Cameroon and Sierra Leone. It has been used in the traditional system of medicine for various gastrointestinal disorders such as diarrhea, ulcers and dyspepsia and in the management of diabetes mellitus (Nwinyi *et al*., 2008). The leaves have been reported to possess a hypoglycemic effect (Ogundipe *et al*., 2003) and decreases activity of glucose kinase enzyme and level of hepatic glycogen, hepatic and blood glucose (Eleyinmi, 2007). Ethanol extract of *G. latifolium* has been reported to increase white blood cell count and hemoglobin concentration in normal condition (Antai *et al*., 2009) while the leaves have strong modulatory effect against hepato cellular damage induced by carbon tetrachloride (Etim *et al*., 2008). The plant also has an anti-in-flammatory property (Morebise *et al*., 2002) and also exhibits antimicrobial activities against various microbial pathogens (Osuala *et al*., 2005).

The extract of the leaves may be used to prevent or reduce weight loss, growth depression and haematotoxicity in diabetic subject (Edet *et al*., 2011, Essien, 2022). *Tapinanthus bangwensis* (*Mistletoe*) belongs to the family of Viscaceae. It is well known evergreen parasitic plant, which grows on deciduous trees in ball-like bush (Evans, 2005). It is an excellent medicinal plant. Mistletoe can grow on either edible or non-edible trees, while only those that grow on-visible plants have disease curing specificity, for example, Mistletoe grown on guava, kolanuts and citrus are specific for curing diseases like cancer, hypertension, nervousness and insomnia, while those grown on cocoa is best used for curing diabetes (Akah and Okarfor, 1992).

Tapinanthus bangwensis have been used against a variety of diseases such as disorders in female reproductive system, cancer, arthritis, rheumatism, epithelial tumour, hypertension, asthma, and epilepsy (Evans, 2005; Akah and Okarafor, 1992). *Tapinanthus bangwensis* have been reported to contain the following phytochemicals; alkaloids, flavonoids, saponins, tannin, cardiac glycosides and steroidal aglycon (Iheanacho *et al*., 2008; Olaleye, 2005, Essien, 2023). It also constitutes of phenols, terpenoids, phytate and carbohydrates (Inyang, 2003; Trease and Evans, 2002).

Therefore, the reported medicinal potentials of *G. latifolium and T. bangwesis* leaves motivated the researchers to seek to evaluate the effects of *G. latifolium* and *T. bangwensis* leaves on blood glucose level, body and organs weight in normal male Albino Wistar rats.

2.0 MATERIALS AND METHODS

2.1 Collection and preparation of plants samples

The fresh leaves of *Gongronena latifolium* and *Tapinanthus bangwensis* were collected at different locations within Ikot Ekpene and Ikono Local Government Areas, both in Akwa Ibom State, Nigeria. The two plants were authenticated by a Taxonomist in the Department of Botany and Ecological studies, faculty of Science, University of Uyo, Akwa Ibom State, Nigeria (Essien *et al*., 2023). The two plants leaves were plucked from their stems, washed with distilled water to remove dirt, sliced separately with knife into pieces and dried separately at room temperature for 3 days after which they were separately ground using clean, dry mortar and pistle and 600 g each of the samples were separately soaked in 100ml of 70% ethanol for 72hrs at room temperature (Essien *et al*., 2022) the macerated leaves extracts were differently filtered using Whatman No. 1 filter paper by means of a funnel. The filtrates were separately concentrated for 3 days, after which slurry form of the extracts obtained and preserved in a refrigerator at $4^{o}C$ for further use (Essien *et al*., 2023).

2.2 Experimental Design, Grouping and Treatment of the Animals

Twenty-five (25) healthy adult male albino Wistar rats weighing 135-290g were obtained from the disease-free stuck of the animal house, pharmacy Department, University of Uyo, Uyo, Akwa Ibom State, Nigeria. The animals were housed in a cage with the sizeable compartments of wooden button and were mesh top, randomly assigned 5 animals per four groups. The animals were maintained under standard conditions of temperature and natural light-dark cycle for 7 days acclimatization in the animal houses, Akwa Ibom State Polytechnic, Ikot Osurua. Groups 1 and 2 were respectively treated with 200mg/kg, 300mg/kg *Gongronema latifolium* and *Tapinanthus bangwensis* respectively, whereas group 3 was treated with 400mg/kg combined extracts of the two plants at 50:50 dosage ratio. Group 4 was not treated and served as control. The treatment was done daily via oral route for one month. All animals were allowed free access to commercial rat mesh and with water throughout the treatment period.

3.0 METHODS

3.1 Determination of Blood Glucose

Blood glucose level was determined with a glucometer. A sterilized lancet was used to obtain blood from the tip of the tail of each rat after overnight fasting. The blood was introduced into the instrument by means of a strip, after which the glucose level was read in mg/kg. This was conducted on day 0, 3, 6, 9, 12, 15, 18 and 21 making up three weeks.

3.2 Determination of Body Weight

Each of the animals in each group was weighed using a weighing balance in grams. This was done on day 0, 3, 6, 9, 12, 15, 18 and 21 for the period of three weeks.

3.3 Determination of Organs Weights

Each of the animals in each group was sacrifice after three weeks treatment. The organs (liver and kidney) were obtained from the rats and weighed using a sensitive balance in grams

3.4 Statistical Analysis

Data generated from the tests were subjected to one-way analysis of variance (ANOVA). Bonferroni's multiple range test was used on the results expressed as mean ± standard error of mean (SEM). This was estimated using statistical package for social sciences (SPSS) version 23 significant differences were determined at ($P<0.05$).

4.0 RESULTS

Table 1: Mean Blood glucose levels of male Albino Wistar Rats Co-administered with Ethanol leaves Extracts of *G. latifolium* and *T. bangwensis*.

Blood Glucose level (mg/kg)

Group	Day 0	Day 3	Day 6	Day 9	Day 12	Day 15	Day 18	Day 21
1	82.50±6.29	78.00±5.58	76.75±5.62	70.75±5.34	66.25±4.09	56.75±4.31	58.50±6.13	63.50±6.86
2	82.10±2.60	78.75±2.63	74.50±2.63	67.50±2.90	61.50±2.99	56.75±4.66	62.25±1.60	73.75±4.33
3	83.75±7.69	80.50±3.88	73.75±8.75	70.50±8.02	62.50±4.74	59.25±3.86	58.50±5.38	63.50±5.56
4	87.00±5.67	90.00±1.47	93.25±2.02	82.50±3.50	84.25±2.56	83.00±2.16	85.50±1.04	79.50±2.16

Results presented as mean ± SEM of three triplicates

Table 2: Mean Body weight of Albino Wistar Rats Co-administered with Ethanol leave Extracts of *G. latifolium* and *T. bangwensis*.

Body weight of Rat(s)

Group	Day 0	Day 3	Day 6	Day 9	Day 12	Day 15	Day 18
1	180.50±9.77	183.25±9.07	177.75±5.91	170.25±4.66	165.00±5.07	165.00±5.78	160.75±15.85
2	208.50±8.20	206.50±9.60	202.75±5.54	203.50±8.45	201.00±10.16	198.50±10.08	194.60±9.47
3	249.75±2.51	248.00±12.50	237.00±9.62	233.75±1.67	230.75±12.00	230.75±11.19	218.25±8.19
4	157.5±3.07	159.75±3.47	156.00±8.84	152.50±12.74	154.75±15.38	154.25±13.26	167.50±6.61

Results presented as mean± SEM of 2 duplication

Table 3: Mean Organ weight of Albino Wistar Rats Co-administered with Ethanol leaves Extracts of *G. latifolium* and *T. bangwensis*.

Group	Liver (g)	Kidney (g)
1	5.81 ± 0.42	1.16 ± 0.06
2	6.15 ± 0.14	1.19 ± 0.04
3	7.35 ± 0.02	1.13 ± 0.08
4	6.01 ± 0.38	1.43 ± 0.17

Results present as mean ± SEM of 2 duplicates

DISCUSSION

Recent years have witnessed a renewed interest in plants as pharmaceutically potent products in the Western world. The interest is channeled into the discovery of new biologically-active molecules by the pharmaceutical industry and into the adoption of crude extracts of plants for self-medication by the general public (Summer, 2000). In both of these areas, some attention is being paid to the investigation and use of enthnopharmacology, the traditional use of plants for medical study purpose by particular cultural groups. The present study evaluated the effects of the combined extracts of *Tapinanthus bangwensis* and *Gongronema latifolium* leaves on blood glucose level, body and organs weights in albino Wister rats. The results of the effects of the combined extracts of *T. bangwensis* and *G. latifolium* leaves on blood glucose levels on day 0, 3, 6, and 9 did not show any significant difference in groups 1, 2 and 3 when compared to the control group.

But blood glucose levels on day 12, 15, 18 and 21 recorded significant decreases in all the extracts treated groups when compared to the control group. The decrease was not dose and treatment duration dependent.

Furthermore, the mean blood glucose levels in the extracts treated groups on day 0, 3, 6 and 9 were within the normal range of 70-100mg/dl for human. But, the mean blood glucose levels from day 12 to 21 fell below the normal range. This is in accordance with the report by Ogundipe *et al*. (2003), that the two plants leave extracts possess anti-diabetic potentials as they have exhibited anti-hypergycermic activities in the test groups of animals under study. Hyperglycermia is a condition in which an excessive amount of glucose circulates in the blood plasma. This is generally a blood sugar level higher than 11.1mmol/l (200mg/dl), but symptoms may not start to become noticeable until even higher values such as 15-20mmol/l (≈250-300mg/dl) (Pais *et al*., 2007; Luna and Feinglos, 2001). The observed hypoglycermic effect is

speculated to be due to the phytoconstituents. However, the glucose lowering activity may be due to the alteration in glucose metabolizing machinery via the inhibitory activity of α-glucoside in the intestine which in turn reduces glucose formation. Also, the extracts are suggested to have decrease glucose-6-phosphatase activity, stimulate glucose catabolism and significantly increase insulin levels. The reverse of these occurs when there is an increased blood glucose level above normal range.

However, evaluation of blood glucose (hyperglycemia) is the hallmark of uncontrolled diabetes which is correlated to cardiovascular diseases (Ugochukwu and Babady, 2002; Pais *et al*., 2007). The decrease in the level of blood glucose below the normal range is termed hypoglycemia and it is characterized by headache, confusion, seizures, coma, and if not corrected may lead to death. When these occur, it signifies deprivation of glucose supply to the central nervous system (particularly the brain) and to some extent the erythrocytes and skeletal muscle.

Again, significant decrease in body weight on day 0, 3, 6 and 9 among the animals in the extracts treated groups were recorded when compared to the control group. For day 12, 15, 18 and 21, significant decrease was found in treatment groups 1, 2, and 3 when compared to the control group. Meanwhile, the decrease in body weight in the treated groups was not dose dependent too. From biochemical metabolic analysis, it has been established that fatly acids which constitute the body fat contents, can be synthesized from simple carbohydrates such as glucose (Nelson and Cox, 2005), thus, it is not unexpected for decrease in blood glucose to induce decrease in body mass index (BMI), as increase in glucose level have associated with increase in lipid biosynthesis (Lipogenesis) and hence, an increase in weight (Nelson and Cox, 2005) since BMI is proportional to weight from its standard formula; weight/square height, it is therefore expected that factors such as blood glucose which influence weight will ultimately affect BMI. According to Kahn (1994), a strong correlation has been established between a height, BMI and the development of type II diabetes mellitus from a study of more than 7000 British men (mean follow up of 12 years). However, since *T. bangwensis* efficacy to reduce blood glucose significantly in the treated groups of animals, they also have the potency to reduce their body weights.

Furthermore, liver and kidneys are very important organs in the mammals. These were the organs whose weights were investigated with respect to the effect of the plant extracts of *T. bangwensis* and *G. latifolium* leaves. Significant difference was not found in the weight of liver and kidneys in the treated groups of animals when compared to the control group. Significant increase was only recorded in the weight in the weight of the liver in group 3 when compared to group 1. However, change in body weight is related to change in organ weight depending on the organ involved in the metabolism of a particular substance (Brai *et al*., 2007). Hence, decrease in blood glucose through enhanced insulin sensitivity is associated with reduction in body weight (Akpabio, 2012; Jun *et al*., 2008). Glucose is the major source of energy for cellular activities, a

decrease in glucose results in the catabolism of lipids accumulated in the adipose tissue leading to reduction in the mean body weight.

Contrary to this, obesity acts as a diabetogenic factor and leads to a decrease in insulin receptors on insulin target cell (Satyanarayana and Chakrapani, 2014).

Therefore, *T. bangwensis* and *G. latifolium* leaves extracts can thus be said to possess anti-hyperglycemic potentials and could be employed internally for treatment of diabetes mellitus after their toxicological studies.

CONCLUSION

Based on the results obtained in this work, it could be concluded that the combined leaves of *T. bangwensis* and *G. latifolium* extracts have exerted their hypoglycemic potentials on the male albino Wistar rats under study. However, the plants leave extracts could be of advantage to man in treating diabetes mellitus after full toxicological studies is conducted.

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