PRINT ISSN 1119-8362
Electronic ISSN 1119-8362

Full-text Available Online at
https://www.ajol.info/index.php/jasem
https://www.bioline.org.br/ja

J. Appl. Sci. Environ. Manage.
Vol. 26 (2) 317-322 February 2022

Phytochemistry, and Effects of *Telfairia occidentalis* Leaf Extracts on the Growth and Haematological Properties of Wistar Albino Rats

UDOSEN IR; *OSU SR

Department of Biology, School of Science, College of Education Afaha Nsit, P.M.B. 1019 Etinan, Akwa Ibom State, Nigeria.
**Corresponding Author Email: samuelrobert2007@yahoo.com; Tel: +2348028260825*

ABSTRACT: Standard protocols were used to assess the biochemistry, phytochemistry, and effects of *Telfairia occidentalis* leaf extracts on the growth and haematological parameters of wistar albino rats. Results revealed ascorbic acid and chlorophyll contents of 158.2±0.7mg/100g and 5.8±0.1mg/100g respectively in fresh leaf samples, while plant extracts contained abundant Tannin, Cardiac glucoside and Flavonoids with moderate amounts of Alkaloid, Saponin and Terpene. Acute toxicity of extracts determined by administering crude ethnaolic extracts intraperitoneally to rats at a dose range of 200mg/kg-1000mg/kg revealed an LD_{50} of 480 mg/kg. Three fractions (n-hexane, ethylacetate and n-butanol) were obtained and screened for rat growth promoting potentials. Results of weight, water intake and feed consumption revealed a net gain of 57.7g (22%) and 30.4g (11%) in weight; increased water intake of 3.9ml (19.7%) and 1.3ml (17%) and increased feed consumption of 1.5g (12.9%) and 1.3g (11.6%) in rats after 28 days treatment with n-butanol and ethylacetate fractions respectively. Studies of haematological potentials of n-butanol fraction of *T. occidentalis* leaf extracts showed significant ($P<0.05$) increase in Red Blood cells level ($6.19\pm0.20x10^6$/L), Haemoglobin (18.36±0.84g/dL) and platelet counts ($747.33\pm2.03x10^3$/mL). When compared to other fractions, the butanol fraction had the highest haematological activity, and additional analysis suggested that the bioactive molecule B_2b may be responsible for this. The findings suggest that butanol fraction B_2b is a safe and potent hematinic capable of alleviating anemic conditions, and they also confirm the use of *T. occidentalis* leaf decoction in traditional medicine for the treatment of anemia and other blood-related disorders, suggesting that it could be prescribed as an adjunct to dietary and main anemia therapy.

DOI: https://dx.doi.org/10.4314/jasem.v26i2.20

Impact factor: http://sjifactor.com/passport.php?id=21082

Google Analytics: https://www.ajol.info/stats/bdf07303d34706088ffffbc8a92c9c1491b12470

Keywords: Phytochemistry, Leaf extracts, *Telfairia occidentalis*, Haematological properties, Wistar albino rats.

According to the World Health Organization, four billion people, or almost 80% of the world's population, use plants and plant components in Medicare (WHO, 2004). Following this reliance on plants for therapeutic purposes, there has been a resurgence of interest in plant study and usage in the last several decades. The present prevalent assumption that green medicine is inexpensive, safe, trustworthy, and more accessible than expensive synthetic medications, which may be linked with unpleasant side effects, has sparked an interest in plant-derived pharmaceuticals (Adeneye and Benebo, 2007). Many plants, in addition to providing nutrients, contain a variety of effective bioactive compounds that can produce a variety of physiological effects depending on the mode of application and have been shown to aid in the treatment of tropical diseases such as malaria (Elujoba, Odeleye, and Ogunyemi, 2005), sickle-cell anemia (Okigbo and Menke, 2006), microbial infection (Iwu, Duncan, and Okuriji, 1999), diabetes (Akah *et al*., 2009) and (Imafidon and Okunrobo, 2012). Many edible plants have medical value in addition to being edible (Udofia *et al.*, 2012), and much is known about medicinal plants because they include a large number of therapeutically effective elements that may be used as starting materials for medicines (Sofowora, 1984). The study of medicinal plants used in traditional medicine to alleviate illnesses is gaining popularity. This method was also advised and supported by the World Health Organization, particularly in nations where access to conventional therapy is limited (WHO, 1980). The WHO has also underlined that while choosing herbal medication for use in healthcare, safety should be the most important factor to consider. It has been noted that *Telfairia occidentalis* is used to cure ailments, and certain reports back this up (Asiegbu, 1987; Ajayi *et al.*, 2000; Fasuyi and Nonyerem, 2007). In Nigeria, *T.*

occidentalis (pumpkin) is an extensively utilized indigenous plant. It is mostly used for nutritional benefits, although it has recently been utilized for medicinal purposes. It's been used for centuries as a digestive tonic, an appetizer, and a cure for dizziness and anemia. *T. occidentalis is high in chlorophyll and ascorbic acid, according to the results of this study.* Tannin, cardiac glycoside, and flavonoids are just a few of the phytochemicals found in *T. occidentalis.* The purpose of this study was to see how *T. occidentialis* leaf extracts affected rat growth and hematological parameters.

MATERIALS AND METHODS

Sources of Plant Materials and Animals: Fresh plants (*T. occidentialis)* were obtained from markets in Ikot Ekpene, Eket and Uyo Local Government Areas in Akwa Ibom State early in the morning and transported in moistened jute bags to the Department of Botany and Ecological Studies, University of Uyo for authentication and identification by taxonomists. Voucher specimen was given and specimen deposited at the Herbarium of the Department.

Adult Wistar albino rats (250 - 300g) were obtained from the animal house of Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Uyo, Uyo. Animals were housed in wooden cages with wire gauze under uniform husbandry conditions of day light, night darkness, room temperature with wood shavings as their bedding materials and allowed two weeks acclimation. Ethical approval was obtained from the Animals use and Ethics committee of the Faculty of Pharmacy, University of Uyo, Uyo, Akwa Ibom State.

Processing of Plant Materials: Fresh leaves of *T. occidentialis* were destalked, rinsed in tap water and spread in a single layer thickness on tray and shade-dried between 9am and 5pm. Dehydrated products were pulverized into fine powder using laboratory plant mill (Christy and Norris Ltd., Chemsford, England). About 200g of powder was loaded into a thimble and continuously extracted with 95% ethanol in a soxhlet extractor for 72 hours. The solvent was distilled off in a rotary evaporator to obtain a solid residue of 27g (13.5% $^{W}/_{W}$). The ethanol extract was fractionated by thin layer chromatographic methods using n-hexane, ethylacetate and butanol (Bobbit, 1964; Touchstone, 1992) to obtained n-hexane fraction, ethylacetate fraction and butanol fraction.

Phytochemical Screening: Standard protocols (Odebiyi and Sofowora, 1978; Trease and Evans, 1986; Harbone, 1991) were used in detecting bioactive compounds in ethanol extract such as Alkaloids, Saponin, Tannin, Cardiac glycoside, falconoid, terpene and Authoraguinone.

Chemical Analyses: Chlorophyll content of unextracted, dehydrated plant leaves was analyzed by spectrophotometric method while ascorbic acid was determine by the 2,6-dichlorophenol-indophenol titrimetric method according to AOAC (1984).

The concentration of chlorophyll was calculated using

$$C = log \frac{I}{\frac{T}{ab}}$$

Where C = Concentration (mg/ml); T = Transmittance %; a = Path length (cm); b = wavelength (nm)

Acute Toxicity Determination: Acute toxicity studies (LD_{50}) of ethanol extracts of *Telfairia occidentalis* was carried out in rats of both sexes using the method of Lorke (1983) with slight modifications. Rats were weighed and distributed into two groups of five rats per a group. They were acclimatized for two weeks on animal feed and water *ad libitum*. Before experiment, animals were fasted for 24 hours with free access to water only. Two hours before onset of experiment, the water was withdrawn. The extracts obtained were administered to each rat per group intraperitoneally (i.p) at a dose range of 200mg/kg – 10000mg/kg. They were monitored and observed for physical signs of toxicity and death within 24 hours. The LD_{50} was estimated as a geometric mean of maximum dose producing 0% mortality and minimum dose producing 100% mortality i.e.

$$LD_{50} = ab$$

Where a = maximum dose producing 0% morality; b = minimum dose producing 100% mortality

Haematological Studies: The albino rats were further divided into five groups of six rats per a group. Groups I and II served as positive and negative control respectively. Group I was used as a positive control and was fed with haematinics (folic acid), Group II, negative control, fed normally without any treatment. Butanol fraction, n-hexane fraction and ethylaccetate fractions were prepared as drugs based on LD_{50} and administered to experimental rats. Group III was treated with butanol fraction, Group IV, with n-hexane fraction and Group V with ethylacetate fraction. The treatments were given once daily for 28 days. Rats were monitored weekly for body weight, daily for food and water intake. On day 29, rats were sacrificed and blood collected by cardiac puncture into EDTA bottles

for determination of the following haematological parameters viz, haemoglobin level (Hb), Red blood cell (RBC), white blood cell (WBC), platelet (PLT), heamatocrit (HCT), mean corpuscular volume (MCV) and mean corpuscular haemoglobin (MCH).

Statistical Analysis: Values were expressed as mean ± SEM. Statistical analysis was performed by one-way analysis of variance (ANOVA) to test for difference among treatments group. Fisher's Least Significant Difference (LSD) was used to assess significant difference between the controls and treated group.

RESULTS AND DISCUSSION

Chemical analyses of extracts of *Telfairia occidentalis* leaves revealed Ascorbic acid concentration of 158.2 ± 0.7mg/100g and chlorophyll content of 5.8 ± 0.1mg/100g (Table 1.) while phytochemical screening of *T. occidentalis* leaf extracts showed that the sample contained abundant Tannin, cardiac glycoside and flavonoid with moderate amounts of Alkaloid, Saponin and Terpene (Table 2.). Effect of chemical fractions of *Telfairia occidentalis* extracts based on LD_{50} on body weight of Wister albino rats is presented in Table 3. Results revealed that rats treated with Butanol and Ethylacetate fractions gained 57.7g and 30.4g respectively. This was encouraging when compared to 60.5g body weight gained by rates in positive control group treated with 10mg/kg folic acid. Rats in native control fed normally without treatment gained 2.7g in weight after 28 days observations. Water intake increased in butanol and ethylacetate fraction treated rats to 3.9ml and 3.4ml respectively at the end of 4 weeks (28 days). This was an improvement in water when compared with positive control (1.3ml) (Table 4). Table 5 show incremental feed intake of 1.5g and 1.3g in butanol and Ethylacetate fraction treatments whereas a marginal feed increased (0.7g) was observed in rats within the negative control group.

Results of studies of haematological properties of partitioned *T. occidentalis* leave extracts showed a significant ($P<0.05$) increase in Red Blood Cells (RBC), Haemoglobin concentration (Hb), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin Concentration (MCHC) and Platelet (PLT) (Table 6). Leaf extract fraction treated rats showed elevated RBC level (6.19 ± 0.20 x 10^6/l) and Hb (18.20 ± 0.31g/dl). The MCV increased from 58.20 ± 2.14g/ to 63.43 + 0.52gl in rats treated with 10mg/kg folic acid and butanol fraction respectively. The MCHC significantly ($P<0.05$) increased in rats treated with butanol fraction (39.36 ± 0.84g/dl) and Ethylacetate (31.87 ± 0.58g/dl) respectively.

Table 1: Chlorophyll Content and Ascorbic Acid of Fresh and Dried Fluted Pumpkin Leaves

Treatment	Chlorophyll (mg/100g)	Ascorbic acid (mg/100g)
Sun dried	1.1±0.1	121.0±1.2
Fresh vegetable	5.8±0.1	158.2±0.7

Table 2: Phytochemicals of Leaf Extracts of *Telfairia occidneitalis*

Test	Observation	Inference
Alkaloid	Orange precipitate	++
Saponin	Persistent frothing for over 15min	++
Tannin	Brownish green	+++
Cardiac glycoside	Reddish brown	+++
Flavonoid	Crimoson coloured precipitate	+++
Terpene	Pink colouration at the interface	+
Anthraquinone	No pink, red or violet colouration at the lower phase	-

- Absence; + Trace amount; ++ Moderate amount; +++ Abundant

Table 3: Effect of Fractions (Dose Based on LD_{50}) on Body Weight

Treatment	Dose (mg/kg)	Body Weight (g)		Weight Gained (g)
		wk 1	wk 4	
+ Control	(10mg/kg) folic acid	259.6±6.1	320.1±1.1	60.5
- Control	-	256.5±4.2	259.2±2.0	2.7
n-hexane fraction	380	254.8±2.1	267.1±1.9	12.3
Acetylacetate fraction	350	258.8±2.1	289.1±1.8	30.4
Butanol fraction	250	254.5±1.4	312.2±1.6	57.7

Table 4: Effect of Fractions (Dose Based on LD_{50}) on Water Intake

Treatment	Dose (mg/kg)	Water Intake (ml)		Intake (ml)
		wk I	wk 4	
+ Control	(10mg/kg) folic acid	20.4±o.6	25.5±1.0	5.1
- Control	-	19.2±1.1	20.5±1.9	1.3
n-hexane fraction	380	19.1±1.3	22.4±1.0	3.3
Acetylacetate fraction	350	19.5±1.8	22.9±1.1	3.4
Butanol fraction	250	19.7±0.8	23.6±1.3	3.9

Table 5: Effect of Fractions (Dose Based on LD_{50}) on Feed Intake

Treatment	Dose (mg/kg)	Feed Intake (g)		Feed Increment (g)
		wk I	wk 4	
+ Control	(10mg/kg) folic acid	12.9+09	14.6+1.3	1.7
- Control	-	8.4+1.2	9.1+1.6	0.7
n-hexane fraction	380	10.1+0.1	11.3+1.6	1.2
Acetylacetate fraction	350	11.2+0.3	12.3+1.8	1.3
Butanol fraction	250	11.6+0.4	13.1+0.6	1.5

+ = positive; - = negative

Table 6: Effect of Partitioned Fractions of *Telfairia occidentalis* Leaves on Haematological Parameters in Albino Rats

Group	Treatment	Dose	WBC ($x10^3/1$)	RBC ($x10^6/1$)	HB (g/dl)	HCT (%)	MCV (gl)	MCH (pg)	MCHC (g/dl)	PLT (10^3/ml)
1	Positive control	10mg/kg	14.01±0.98	4.08±0.64	13.60±0.35	42.22±0.38	58.20±2.14	18.00±0.22	29.76±0.64	600.33±2.98
2	Negative control	-	10.26±0.25	3.77±0.13	10.56±0.38	19.00+0.33	40.50±1.12	16.06±0.39	24.30±0.26	622.66±3.35
3	n-hexane	447.22mg/kg	8.43±0.40*	4.05±0.16*	12.03±0.94	28.03+0.94	60.30±0.07*	23.60±0.28	30.63±0.47	730.00±2.13*
4	Ethylacetate	447.22mg/kg	7.20±0.78*	4.41±0.61*	10.03±0.47	24.40±2.60*	59.37±0.49	21.60±0.38	31.87±0.58*	634.33±3.61
5	Butanol	447.22mg/kg	14.81±0.14*	6.19±0.20*	18.20±0.31	39.27+0.37*	63.43+0.52*	23.71±0.35*	39.36±0.84*	747.33±2.03*

Values are expressed as mean ± SEM (n=3). All columns (*) are significant using one-way ANOVA, when compared to positive control, following Fisher's Least Significant Difference (LSD) at P<0.05

***Acronyms**: WBA-White Blood Cell; Hb - Haemoglobin Concentration ; MCV - Mean Corpuscular volume; RBC - Red Blood Cell; HCT – Haematocrit; MCH - Mean Corpuscular Haemoglobin PLT - Platelet*

Table 7: Effect of Purified Butanol Fractions of *Telfairia occidentalis* Leaves on Haematological Parameters in Albino Rats

Group	Code of Compound		WBC ($x10^3/1$)	RBC ($x10^6/1$)	HB (g/dl)	HCT (%)	MCV (gl)	MCH (pg)	MCHC (g/dl)	PLT (10^3/ml)
1	+ control	10mg/kg	18.81±2.10	7.22±0.21	4.59+1.09	18.78±0.85	17.67+1.12	18.82+1.15	10.01±0.37	108.57+0.28
2	- control	-	10.01±0.43	3.77±0.13	3.50±0.09	11.22±0.33	18.10±1.14	12.10±0.02	11.76±0.26	101.23±1.98
3	B_2a	447.22mg/kg	13.30±0.35*	4.97±0.32*	4.97±0.32*	13.83±0.15*	21.11±0.12	15.11±0.15*	13.33±1.15*	157.08±2.02*
4	B_2b	447.22mg/kg	16.20±0.55*	4.41±0.21*	5.91±0.21*	12.40±1.30*	23.15±0.21*	19.41±0.38*	16.97±0.21*	154.32±1.11*

Values are expressed as mean ± SEM (n=3). All columns (*) are significant using one-way ANOVA, when compared to positive control, following Fisher's Least Significant Difference (LSD) at P<0.05

***Acronyms**: WBA-White Blood Cell; Hb - Haemoglobin Concentration ; MCV - Mean Corpuscular volume; RBC - Red Blood Cell; HCT – Haematocrit; MCH - Mean Corpuscular Haemoglobin PLT - Platelet*

Butanol fraction and n-hexane fraction enhanced platelet counts (747.33 ± 2.03 x 10^3/ml and 730.00 ± 2.13 x 10^3/ml respectively) when compared to positive control count of 600.33 ± 2.98 x 10^3/ml. Results further revealed that haematological activity of butanol fraction was highest when compared with other fractions and effect of purified butanol fractions of *T. occidentalis* leaf extracts on haematological properties in albino rats in Table 7 showed that the bioactive component B_2 a and B_2 b were responsible for the enhanced activities.b*Telfairia occidentalis* leaves contain a variety of biochemicals and phytochemicals that are all bioactive. Chlorophyll is a chemical molecule that functions as a medium for the movement of dissolved oxygen. It is found in the blood of most animals and is very similar to haemoglobin. The fundamental structure of chlorophyll is a porphyrin ring, similar to haem in haemoglobin, but with magnesium instead of iron as in haemoglobin; still, chlorophyll has the ability to act in the body as haemoglogbin due to this unique property. In this investigation, chlorophyll was shown to help experimental rats restore and refill their red blood cells. Plant leaf extract also has a high ascorbic acid concentration of 158.2 + 0.7 mg/100g, according to research (Table 1). The Reference Daily Intake (RDI) for man is 60mg (Seely *et al* 2002) and this will be exceeded on consumption of a hundred grammes of plant materials. Ascorbic acid is a water soluble vitamin that functions in general protein metabolism. The antioxidant qualities of this chemical (ascorbic acid) and chlorophyll can prevent oxidation of cell components by giving an electron to free radicals. The haematological activity of *Telfairia occidentalis* lead extracts is thought to be enhanced by these compounds. Tannins, saponins, flavonoids, terpenes, and cardiac glycosides were discovered to be abundant in *Telfairia occidentalis* leaf extracts. Flavonoids have biological effects that include protection from allergens, free radicals, and inflammation (Okwu, 2004; Okwu and Omodamiro, 2005). Flavonoids are water-soluble super antioxidants that protect cells from oxidative damage (Okwu, 2004). This might have had a role in the enhanced hematologic activity of the plant extract under investigation. Aside from flavoniods, additional metabolites such as alkaloids and cardiac glycosides were detected in abundance in the sample plant leaves. When given to animals, alkaloids have been discovered to have significant physiological action, whilst cardiac glycosides have been proven to be effective in the treatment of heart disorders (Trease and Evans, 1989). Butanol and Ethylacetate fractions enhanced weight, water intake, and feed consumption in experimental rats after 28 days of sub-acute research. According to Akah *et al.* (2009), the required renal water loss combined with hypoosmolarity tends to deplete intracellular water, triggering the osmoreceptors of the thirst center of the brain, which leads to increased water consumption (UKPDS, 1998)..

The anabolic effects then take over, leading to an increase in feed consumption and weight gain. In this work, it was discovered that administering fractions of *T. occidentalis* leaf extracts improved various hematological parameters in experimental rats. RBC, Hb, MCHC, PLT, and other parameters all increased dramatically. This might explain the rats' overall happiness, as an increase in weight, water, and feed intake indicates enhanced renal and hepatic functioning. This finding is in line with previous research on the hepatoprotective properties of leaf extracts in mice (Iwalokun *et al.*, 2006). Ingestion of medicinal substances has been demonstrated to change a variety of haematological parameters, according to the literature (Ajagbonna *et al.*, 1999). *T. occidentalis* may have a physio-modulatory impact in rats, as evidenced by the significant rise in blood parameters in treated rats. The presences of diverse phytochemicals that may function as strong antioxidants and heterogeneous phytoconstituents in plant crude extracts have been documented to have a synergistic impact on organ/tissue toxicity (Mazunder *et al.*, 2005). *Conclusion:* The findings of this study support the use of *T. occidentalis* leaf extracts in traditional medicine for the treatment of anemia and other blood diseases. The findings also imply that *T. occidentalis* butanol fraction B_2b is a safe and powerful hematinic capable of boosting anaemic conditions and therefore might be given as an addition to dietary treatment as well as primary therapy for anemia. However, the consumption of leaf extract of *T. occidentalis* enhances various hematological parameters such as chlorophyll content, ascorbic acid, and phytochemicals in the leaf extract and therefore improves the physiological and nutritional status of its consumers.

Acknowledgements: The authors wish to thank the Tertiary Trust Fund **(TET-FUND)** for supporting this research publication during the 2011 – 2014 (Merged) Institution Based Research Intervention with Ref. No. TETFUND/DESS/COE/AFAHA NSIT/VOL.2

REFERENCES

Adeneye, AA; Benebo, AS (2007). Pharmacological evaluation of a Nigerian Poly-herbal Health Tonic Tea in Rat. *Afr. J. Biomed. Res.* 10:249-255.

Ajagbonna, OP; Onifade, KI; Sauleiman, U (1999). Haematological and Biochemical Changes in Rats Given Extract of *Calotropis procera Sokoto J. Vet. Scie.* 1(1): 36-42.

Ajayi, OI; Ajayi, TYC; Omokaro, EU; Halim, KD (2000). Erythropoetic Value of Pumpkin Leaf Extract (*Telfairia occidentalis)* in Rabbits – a Preliminary Study. *Niger. J. Physiol. Scie.* 16(1-2): 1-3.

Akah, PA; Alemji, JA; Salawu, OA; Okoye, TC; Offiah, NV (2009). Effects of *Vernonia amygdalina* on Biochemical and Hematological Parameters in Diabetic Rats. *Asian J. Med. Scie.* 1(3): 108-113.

Akah, PA; Alemji, JA; Salawu, OA; Okoye, TC; Offiah, NV (2009). Effects of *Vernonia amygdalina* on Biochemical and Hematological Parameters in Diabetic Rats. *Asian J. Med. Scie.* 1(3): 108-113.

Asiegbu, JE (1987). Some Biochemical Evaluation of Fluted Pumpkin. Seed Science and Agriculture. 40: 151-155.

Ehiyoba, A; Odeleye, OM; Oguuyemi, CM (2005). Traditional Medicine Development for Medical and Dental Primary Health Care Delivery System in Africa. *Afr. J. Tradit. Complement. Altern. Med.* 21(1): 48-61.

Fasuyi, AO; Nonyerem, AD (2007). Biochemical, Nutritional and Hematological Implication of *Telfairia occidentalis* Leaf Meal as Protein Supplement in Broiler Starter Diets. *Afr. J. Biotechnol.* New York, Reinhold Publishing Corporation: 128-182.

Harbone, JP (1991). A Guide to Modern Technique of Plant Analysis. London, Chapman and Hall: 1880.

Imafidon, KE; Okunrobo, LO (2012). Effects of *Vernonia amygdalina* Del. Extracts on Cholesterol Level and Lipid Peroxidation Status in Rats Given Red Dye Adulterated Palm Oil Diets. *Br. J. Pharm. Res.* 2(2): 98-107.

Iwalokun, BI; Alibi – Sofunde, JA; Odunala, T; Magbagbeola, AO; Akinwande, AI (2006). Hepatoprotective and Anti-Oxidant Activities of *Vernonia amygdalina* on Acetaminophen-Induced Hepatic Damage in Mice. *J. Med. Food.* 9(4): 526-530.

Iwu, MM; Duncan, AR; Okunji, CO (1999). New Antimicrobials of Plant Origin In: Janick, J. (ed) Perspective in New Crop and Uses. Alexandria, ASHS Press: 457-462.

Lorke, D (1983). A New Approach to Practical Acute Toxicity Testing. *Arch Toxicol.* 54 (4): 275 – 287.

Mazunder, UK; Gupta, M; Rajeshhwar, Y (2005). Antihyperglycemic Effect and Antioxidant Potential of *Phyllantus nirur (*Euphorbiaceaea) in Streptozotoc-Induced Diabetic Rats. *Euro. Bull. Drug Res.* 13:15-23.

Odebiyi, OO; Sofowora, EA (1978). Phytochemical Screening of Nigeria Medicinal Plants *Lloydia*, 41:234-237.

Okigbo, RN; Menke, EC (2006). An Appraisal of Phytomedicine in Africa. *KMITL Scie. Technol. J.* 6(2): 83-93.

Okwu, DE (2004). Phytochemicals and Vitamin Content of Indigenous Species of South Eastern Nigeria. *J. Sustain. Agric. Environ.* 6:30-34.

Okwu, DE; Omodamiro, OO (2005). Effeects of Hexane Extract and Phytochemical Content of *Xylopia aethiopia* and *Ocimum gratissimum* on the Uterus of Guinea Pig. *Biol. Res.* 3:41-48.

Seely, RR; Stephens, TD; Tate, P (2002). Essentials of Anatomy and Physiology (4th edn.). New York. McGraw-Hill Higher Education. 643p.

Sofowora, A (1984). Medicinal Plant and Traditional Medicine in West Africa. Ibadan, Spectrum Books Ltd. 153p.

Touchstone, JC (1992). Practice of Thin Layer Chromatography. 3rd (edn.). New York, Wiley – Inter-science Publication 340p.

Trease, GE; Evans, WC (1986). Textbook of Pharmacology, 12th edn. London, Bailliere Tindall: 343-383.

Udofia, GE; Asamudo, NU; Ekong, US; Akpan, MM (2012). Photochemistry and Antibacterial Activity of *Andrographis paniculata* (Vinegar) against Clinical Bacterial Isolates from Diarrheic Patients in Southern Nigeria. *Int. J. Chem. Environ. Pharm. Res.*, 3(2): 158-162.

UKPDS (1998). Intensive Blood Glucose with Sulphonylureas or Insulin Compared with Conventional Treatment and Risk of Complications in Patients with Type 2 Diabetes (UK Prospective Diabetes Study Group 33) *Lancet*, 353:837-853.

WHO (1980). Experts Committee on Diabetes Technical Reports Series. World Health Organization Geneva.

WHO (2004). Guidelines on Development Consumer Information on Proper Use of Traditional, Complementary and Alternative Medicine, Genera: 46-61.