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Phytochemical composition, GC-MS analysis and toxicological profiling of *Gouania longipetala* leaf extract in rats

Kenneth C. Nwachukwu[1,2], Godwin S. Aloh[2], Simeon I. Egbachukwu[2], Solomon N. Ijioma[3], Lukman A. Alli[1], Kingsley C. Ugwuanyi[3], Blessing U. Onwuchekwa[4] and Michael P. Okoh[1]*

[1]Department of Medical Biochemistry, Faculty of Basic Medical Sciences, College of Health Sciences, University of Abuja, FCT-Abuja, Nigeria.
[2]Department of Biochemistry, College of Natural Sciences, Michael Okpara University of Agriculture, Umudike, Nigeria.
[3]Department of Zoology and Environmental Biology, College of Natural Sciences, Michael Okpara University of Agriculture, Umudike, Nigeria.
[4]Department of Science Laboratory Technology, School of Pure and Applied Sciences, Abia State College of Health Sciences and Management Technology, Aba, Nigeria.

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This study investigated the phytochemical composition and toxicity potentials of *Gouania longipetala* leaf extract in rats. Crude extract was prepared from freshly collected leaves of the plant and was subjected to phytochemical analyses including gas chromatography-mass spectrometry (GC-MS) and acute toxicity. 15 rats were used for the sub-acute toxicity evaluation, 5 rats in each group of 3, each were treated such that, group 1 is the control, with groups 2 and 3 administered 400 and 800 mg/kg of the extract orally for 28 days. Results showed the extract contains phenols, saponins, steroids, flavonoids, terpenoids with alkaloids concentration recorded as, 34.30±0.14 mg/100 g, cardiac glycoside 3.89±0.04 mg/100 g being the least. GC-MS chromatogram of the extract showed the presence of 20 compounds with Spartein (24.92%) as the most abundant and ethyl oleate (0.79%), the least. Acute toxicity (LD_{50}) value for the extract was found to be >5000 mg/kg body weight in rats. Sub-acute administration of the extract to rats did not significantly alter values of haematological and liver and renal function parameters when compared with control ($p<0.05$) but lowered levels of cholesterol and low-density lipoprotein cholesterol. Antioxidant parameters like reduced glutathione, catalase and superoxide dismutase were also increased in the extract treated rats ($p<0.05$) while Malondialdehyde concentration was lowered significantly ($p<0.05$). Results therefore showed that *G. longipetala* leaf extract may be a very safe alternative for oral use in the management of hyperlipidaemia and oxidative stress induced diseases.

Key words: *Gouania longipetala* extract, phytochemical, antioxidant, gas chromatography-mass spectrometry, toxicity.

INTRODUCTION

At the moment, alternative medicine involving the use of medicinal plants for the management of diseases appears to have been globally accepted for reasons such as availability, affordability, effectiveness and low side

effects (Ijioma et al., 2019; Okoh et al., 2019; Okoh et al., 2021). Nigeria, and in fact the whole of Africa is host to abundant forests and other natural bodies from which these medicines can be sourced (Sofowora, 1993). It is well established that phytochemical components of plants are responsible for either their healing effects or toxicity potentials (Chukwuma and Ejikeme, 2016). On one hand, the screening of plants for the purpose of providing healing has provided cheap and effective treatment alternatives for man and has in addition identified templates for new drugs discovery, but on the other hand, has exposed the toxicity potentials of plant-based medicines (Oshilonya et al., 2016), thus making toxicity evaluation a key component of medicinal plant research.

Phytochemical agents are referred to as secondary metabolites (Chukwuma and Ejikeme, 2016). Common examples are alkaloids, flavonoids, coumarins, tannins, terpenes, terpenoids, phenols, glycosides, etc. These agents possess different pharmacological activities, which determine their applications in health. For examples phenols and flavonoids are strong antioxidant agents and have been used in the fight against diseases caused by oxidative stress (Ijioma et al., 2016). The activities of flavonoids and phenols also strengthen the body's antioxidant defense line leading to reduced risk of diseases and increased well-being (Kanu et al., 2016). The presence of Vitamins C and E in plants also favors the use of plant-based medicines as promoters of the body's antioxidant defense line. For example, it is well established that vitamin C provides protection against oxidative stress-induced cellular damage by its ability to scavenge reactive oxygen species, while vitamin E plays active role in the inhibition of lipid peroxidation processes (Traber and Stevens, 2011).

Gas chromatography and mass spectrometry (GC-MS) is considered one of the most accurate bio-analytical tools available because it can identify and quantify varying active compounds including those of plant origins such as phytochemical compounds by combining the features of gas chromatography and mass spectrometry within a sample component matrix (Olivia et al., 2021). The technique has the advantage of being able to separate complex mixtures, quantify analytes, identify unknown peaks and determine trace levels of contamination (Olivia et al., 2021). Numerous medicinal plants have been screened for phytochemical composition, pharmacological activities and toxicity potentials. In this study, focus is on the plant, *Gouania longipetala*. *Gouania* is a genus of flowering plants, which belongs to the family Rhamnaceae comprising about 50 to 70 species of which *G. longipetala* is one.

These plants which are mainly shrubs are predominantly found in tropical and subtropical regions of Africa, America, and Southern Asia (Sven et al., 2011). The species *G. longipetala* is characterized by watch-spring tendrils, spike-like thyrsus, a more or less lobed disc, inferior ovary and longitudinally 3-wined septicidal fruits (Buerki et al., 2011). The stems and/or leaves of the plant are employed as alternative medicine across Africa for the management of a myriad of human ailments including but not limited to swelling, pain, edema venomous stings, gout, heart diseases, diabetes mellitus, and malaria (Njamen et al., 2013). Other areas of application of particularly the leaves are in the treatment of constipation, venereal diseases, abdominal pain and stomach upsets, pain, bacterial infections, and inflammations (Ekuadzi et al., 2012, 2014). In Orba village, South Eastern Nigeria (where the plant is referred to as "Asha"), the leaves are used specifically for the treatment of diabetes mellitus (Focho et al., 2009).

Results of scientific studies so far suggest that the plant demonstrated significant antibacterial, antioxidant, anti-inflammatory, anti-diabetic, anti-hyperlipidemic and estrogenic effects (Ezeja et al., 2015). Despite these findings, information on phytochemical composition of *G. longipetala* and its toxicity potentials remain scanty and virtually unavailable, and this is the main stay of our study here.

MATERIALS AND METHODS

Collection of plant materials and authentication

Fresh leaves of *G. longipetala* were collected from a farm settlement in Nsukka town, Nsukka Local Government Area of Enugu State, Nigeria and were authenticated at the Department of Forestry, College of Natural Resources and Environmental Management, Michael Okpara University of Agriculture, Umudike in the month of February, 2021. Dried sample of the material was assigned voucher number MOUAU/ZEB/HERB/21/006 and preserved in the herbarium of the Department of Zoology and Environmental Biology, Michael Okpara University of Agriculture, Umudike.

Preparation of extract

The Soxhlet extraction technique used by Orieke et al. (2019) was adopted with little modifications. Freshly collected leaves of the plant were dried under shade for 14 days and were thereafter pulverized to coarse powder in a manual blender. 80 g of the powdered sample was introduced into the extraction chamber of the Soxhlet extractor for extraction using ethanol as solvent. Temperature was maintained at 65°C through-out the extraction period of 48 h. At the end of the period, the collected extract in ethanol was dried in a hot air oven at 40°C to obtain a brown pasty extract which weighed 8.89 g and represented 11.11% extract yield.

*Corresponding author. E-mail: michael.okoh@uniabuja.edu.ng. Tel: 234-703-668-3068.

Qualitative and quantitative phytochemical analysis of *G. longipetala* leaf extract

Qualitative and quantitative phytochemical analyses of the extract were carried in accordance with the methods outlined by Ezeonu and Ejikeme (2016). The methods were used to test the presence and amounts of alkaloids, phenols, cardiac glycosides, saponins, steroids, flavonoids, terpenoids, and tannins.

Phytochemical analysis of the extract by GC-MS

The GC-MS analysis of the leaf extract was performed using BUCK M910 BUCK M910 Gas chromatography furnished with HP-5MS section (30 m long × 250 µm in width × 0.25 µm in thickness of film). Spectroscopic identification by GC–MS included an electron ionization framework, which used high energy electrons (70 eV). Unadulterated helium gas (99.995%) was utilized as the transporter gas with stream pace of 1 mL/min. The underlying temperature was set at 50 to 150°C with an expanding pace of 3°C min^{-1} and a holding season of around 10 min. At long last, the temperature was expanded to 300°C at 10°C min^{-1}. One microliter of the pre-arranged 1% of the concentrates diluted with particular solvents was infused in a splitless mode. Relative amount of the compounds present in every one of the concentrates was communicated as rate dependent on the top region created in the chromatogram.

Extraction of phytochemicals

In order to extract and separate the phytochemical components of the extract, 1 g of the sample was gauged and moved into a test tube and 15 ml of ethanol was added. The sample inside the test tube was incubated in a water shower at 60°C for 60 min. The reacting sample inside the test tube was moved to a separator pipe, following the progressive washing of the tube's sample effectively with 20 mL of ethanol, 10 mL of cold water, and 10 mL of heated water and 3 mL of hexane, which was totally moved to the channel. The combined resultant extract concentrates were then subjected to the 3 times treatment with 10 mL of 10% v/v ethanol watery arrangement. The arrangement was dried with anhydrous sodium sulfate and the dissolvable was dissipated. The sample was solubilized in 1000 µL of ethyl acetate of which 200 µL was moved to a vial for analysis.

Identification of bioactive components

The distinguishing proof of the constituents of *G. longipetala* leaf extract was accomplished on the premise of comparing the retention index of the mass spectral fragmentation patterns, with those found on the data base of the National Institute Standard and Technology (NIST). In each case the obscure spectra of the mass spectrum was compared with the known component of the NIST database.

Animals

Thirty six matured Wistar rats (120-150 g) obtained from the laboratory animal production unit of the Department of Zoology and Environmental Biology, Michael Okpara University of Agriculture, Umudike were used for the different segments of the study. Twenty-one of the rats were used for acute toxicity evaluation while the remaining fifteen were used for the sub-acute toxicity study. The rats were housed in well ventilated aluminium cages under hygienic conditions and allowed to acclimatize for 14 days before commencement of experiments. Animals were fed with normal rat chore (Chikun Finisher) and water *ad libitum* but were starved for 12 h before each experiment. All experiments were carried out in accordance with International Guidelines and as stipulated by an Ethical Committee in Michael Okpara University of Agriculture, Umudike.

Acute toxicity (LD_{50}) evaluation of *G. longipetala* leaf extract

Acute toxicity value of the extract was determined in albino rats in accordance with a modified new Lorke's method (Lorke, 1983) as was used by Orieke et al. (2019). The test was carried out in two phases. In each first phase, nine randomly selected albino rats were divided into three groups (1, 2 and 3) of three animals each and were administered 10, 100 and 1000 mg/kg body weight of the extract, respectively via the oral route. The rats were thereafter observed within 6 h post administration for signs of toxicity. With zero mortality recorded across the groups at the end of 24 h, the study proceeded into the second phase which also involved the use of a new set of 9 rats also assigned to 3 groups (1, 2 and 3) of 3 rats and administered 1600, 2900 and 5000 mg/kg of the extract, respectively. The treated animals were also observed for toxicity signs and mortalities within 24 h and a further 7 days. The metrical mean of the maximum dose that produced no mortality and the minimum dose that produced 100% mortality was taken as the mean lethal dose (LD_{50}) of the extract.

Sub-acute toxicity evaluation of *G. longipetala* leaf extract in rats

Fifteen matured albino rats assigned to 3 groups of 5 rats each were weighed and put in separate cages and assigned graded treatment with the extract as follows: Group 1: 0.5 ml normal saline (control); Group 2: 400 mg/kg *G. longipetala* leaf extract; Group 3: 800 mg/kg *G. longipetala* leaf extract.

All administrations were via the oral route and lasted for a period of 28 days before the animals were sacrificed by cervical dislocation for cardiac puncture blood collection into EDTA bottles for haematological study and also plain bottles for serum biochemical tests including liver and renal function tests and antioxidant enzymes assays. Body weights of the rats were determined at the beginning and end of treatment.

Determination of haematological, biochemical and *in vivo* antioxidant parameters

Haematological values including red blood cells count (RBCC), packed cell volume (PCV), haemoglobin (Hb), white blood cells count (WBCC), platelets count (PLTC), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were determined for each sample using an automated haematology analyser (BC-2300, Mindray Company, China). Liver function parameters including aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), total protein and bilirubin, and renal function parameters including urea, creatinine, sodium, and electrolytes were all determined using commercial test kits with strict adherence to procedures outlined by the producer Randox Laboratories, UK. Oxidative stress markers including superoxide dismutase (SOD), catalase (CAT), glutathione (GSH), glutathione peroxidase (GPx), and Malondialdehyde (MDA) were determined in serum using commercial test kits as was also used by Kanu et al. (2016).

Statistical analysis

Results were presented as mean values ± standard deviations (mean ± SD). The replicates in each treatment were subjected to

Table 1. Qualitative phytochemical composition of *G. longipetala* leaf extract.

Parameter	Qualitative test	Quantitative test result (mg/100 g)
Saponins	++	18.10±0.05
Tannins	+	9.72±0.25
Phenolics	+++	23.19±0.12
Flavonoids	++	16.49 ±1.08
Steroids	++	13.74±0.19
Terpenoids	+	7.45±0.10
Cardiac glycosides	+	3.89±0.04
Alkaloids	+++	34.30±0.14

+: Present.

one-way analysis of variance (ANOVA) and the difference between the samples' mean was tested by Tukey *post-hoc* test using R-statistics software version 3.03. P-values ≤ 0.05 were considered as being statistically significant.

RESULTS

Results of qualitative and quantitative phytochemical analysis of *G. longipetala*

Phytochemical agents including saponins, tannins, phenolics, flavonoids, steroids, terpenoids, glycosides and alkaloids were found to be present in *G. longipetala* leaf extract following qualitative tests (Table 1). Results of quantitative estimation of the relative amounts of these agents in the extract showed that alkaloids was most in abundance (34.30±0.14 mg/100 g) and was followed by phenols (23.19±0.12 mg/100 g) while cardiac glycosides was the least (3.89±0.04 mg/100 g). Table 1 also shows the relative amounts of identified phytochemical agents in the extract.

Results of GC-MS analysis of *G. longipetala* leaf extract

The GC-MS analysis of the ethanol extract of *G. longipetala* leaf extract as shown in the chromatogram (Figure 1) revealed 20 peaks which represent 20 identified compounds. The compounds with higher availability are Spartein (24.92%), Kamferol (12.97%) and Oleic acid (10.94%) while the compound with the least availability is ethyl oleate (0.79%). A full list of all compounds identified and quantified in the extract by GC-MS is presented in Table 2.

Result of acute toxicity evaluation of *G. longipetala* leaf extract

No death was recorded in any group at all stages of the acute toxicity test, even at the highest dose of 5000 mg/kg administered. Animals instead retained their physical activities, showed no signs of toxicity and survived throughout the 24 h and a further 7 days observation period (Tables 3 and 4). LD_{50} value for the extract was therefore found to be >5000 mg/kg body weight.

Effects of *G. longipetala* leaf extract on haematological parameters in rats

For all haematological parameters including RBC, PCV, Hb, WBC, PLT, MCV, MCH and MCHC studied, no significant difference was observed between values obtained in the test group and the control ($p>0.05$), although slight rise was observed in groups treated with the extract (Table 5).

Effects of *G. longipetala* leaf extract on liver and renal function parameters in rats

Treatment with *G. longipetala* leaf extract did not significantly alter the values of liver and renal function parameters in treated rats when compared with control ($p>0.05$). Values of total protein, AST, ALT, ALP and bilirubin did not change in the test rats when compared with control ($p>0.05$). Same trend of results was obtained for renal function parameters including urea, creatinine and the electrolytes when compared with control ($p>0.05$). Results for liver and renal function parameters are presented in Tables 6 and 7, respectively.

Effects of *G. longipetala* leaf extract on lipid profile parameters in rats

Results obtained for lipid profile in the test rats were significantly different from those obtained in the control group ($p<0.05$). The values of total cholesterol in the groups treated with the extract were lower than the value

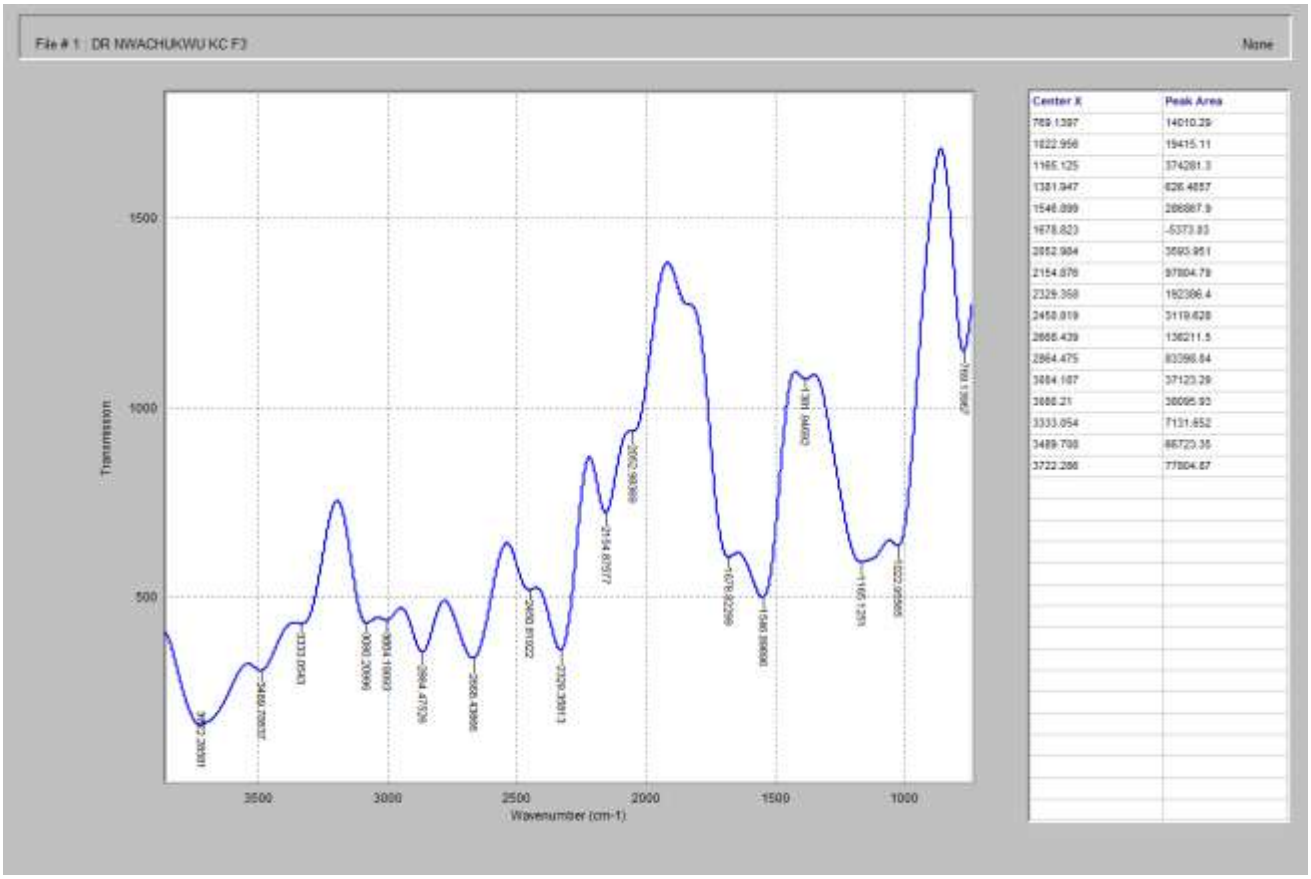

Figure 1. Chromatogram showing peaks for identified compounds in *G. longipetala* leaf extract.

Table 2. Compounds identified in *G. longipetala* leaf extract by GC-MS.

S/N	Name of compound	Concentration (%)
1	2-tetradecanol	1.07
2	Dodecanoic acid	0.98
3	1-octadecene	0.81
4	Dodecanoic acid	2.69
5	5,6-dehydrolupanine	2.11
6	Propanoic acid, 3-chloro-, methyl ester	0.66
7	Luparine	1.44
8	Sapogenine	2.14
9	Catechin	7.12
10	Flavon-3-ol	6.63
11	Anthocyanin	0.88
12	Spartein	24.92
13	Resveratrol	7.73
14	Linoelaidic acid	2.98
15	Aragyrine	4.56
16	Methyl 9,12-heptadecadienoate	2.91
17	Baptifoline	5.76
18	Oleic acid	10.94
19	Kaempferol	12.97
20	Ethyl oleate	0.79

Table 3. Phase 1 result of acute toxicity evaluation of *G.longipetala*leaf extract.

Group	Dose (mg/kg)	No. of death	Observation
1	10	0/3	Animals were active and physically stable. No signs of toxicity were observed
2	100	0/3	Animals were active and physically stable. No signs of toxicity were observed
3	1000	0/3	Animals were active and physically stable. No signs of toxicity were observed

Table 4. Phase 2 result of acute toxicity evaluation of *G.longipetala* leaf extract.

Group	Dose (mg/kg)	No. of death	Observation
1	1600	0/3	Animals were active and physically stable. No signs of toxicity were observed
2	2900	0/3	Animals were active and physically stable. No signs of toxicity were observed
3	5000	0/3	Animals were after administration, calm for about 30 minutes before regaining activity their physical activity

LD_{50}> 5000 mg/kg body weight.

Table 5. Haematological parameters in rats treated with *G. longipetala* leaf extract.

Parameter	Control	GL leaf extract (400 mg/kg)	GL leaf extract (800 mg/kg)
RBC ($\times10^6$ mm^{-3})	7.14±0.18[a]	7.20±0.18[a]	7.29±0.29[a]
PCV (%)	44.67±0.58[a]	44.94±1.16[a]	45.33±1.53[a]
Hb (g/dl)	15.67±0.40[a]	16.10±0.36[a]	16.17±0.15[a]
WBC ($\times10^3$ mm^{-3}))	8.71±0.39[a]	8.86±0.37[a]	8.76±0.54[a]
PLT ($\times10^3$ mm^{-3})	194.73±6.18[a]	198.93±12.79[a]	191.80±8.45[a]
MCV (fl)	62.60±0.98[a]	62.01±0.50[a]	62.23±0.66[a]
MCH (pg)	21.95±0.04[a]	22.35±0.06[a]	22.21±0.86[a]
MCHC (g/dl)	35.07±0.61[a]	36.05±0.27[a]	35.68±1.07[a]

Values are presented as mean ± standard deviation (n = 5). The mean on the same row with different letter superscripts is significantly different (P < 0.05) from any paired value and vice versa.

obtained for the control group. Triglycerides and low-density lipoprotein cholesterol concentrations were also significantly lower than control values (P<0.05). The concentration of very low-density lipoprotein cholesterol was not altered significantly. In the evaluated lipids, only minor difference was observed (Table 8).

Effects of *G. longipetala* leaf extract on some serum antioxidant parameters in rats

Antioxidant enzymes activity levels were significantly increased in the groups treated with the extract when compared with control (p<0.05). Activities of GSH, GPx, SOD and CAT were all higher in the extract treated groups than control, but did not significantly differ at different dose levels of treatment (p>0.05). However, the concentrations of MDA in the extract treated groups are not significantly different from the control value (p>0.05). Results for these antioxidant parameters are presented in Table 9.

DISCUSSION

The significant presence of saponins, tannins, phenols, flavonoids, steroids, terpenoids, glycosides and alkaloids in *G. longipetala* leaf extract attests to the fact that the plant can be harnessed into a veritable healing substance. Phytochemical agents in plants are the reasons for their enormous medicinal potentials and have over the years been implicated in the healing of diseases (Oshilonya et al., 2016). In fact, the roles of the aforementioned phytochemical agents in the healing process have extensively been reported (Agidew, 2022; Ijioma et al., 2016; Aye et al., 2019). For example,

Table 6. Liver function parameters in rats treated with *G. longipetala* leaf extract.

Treatment	Control	GL leaf extract (400 mg/kg)	GL leaf extract (800 mg/kg)
TP (g/dl)	7.11±0.25[a]	7.30±0.09[a]	7.43±0.15[a]
ALT (U/L)	25.00±5.00[a]	27.33±3.06[a]	28.33±3.51[a]
AST (U/L)	30.00±2.00[a]	30.33±3.06[a]	34.33±2.08[a]
ALP (U/L)	84.67±4.16[a]	80.33±2.08[a]	82.33±2.89[a]
T. Bil. (mg/dl)	0.51±0.09[a]	0.55±0.03[a]	0.46±0.12[a]

Values are presented as mean ± standard deviation (n = 5). The mean on the same row with different letter superscripts is significantly different (P < 0.05) from any paired value and vice versa.

Table 7. Renal function parameters in rats treated with *G. longipetala* leaf extract.

Parameter	Control	GL leaf extract (400 mg/kg)	GL leaf extract (800 mg/kg)
Urea (mg/dl)	16.27±1.35[a]	17.27±1.06[a]	17.63±0.76[a]
Creatinine (mg/dl)	0.71±0.03[a]	0.78±0.04[a]	0.75±0.08[a]
Na^+ (mEq/L)	127.50±2.01[a]	128.77±2.17[a]	127.37±2.15[a]
K^+ (mEq/L)	4.66±0.29[a]	4.65±0.23[a]	4.97±0.12[a]
Cl^- (mEq/L)	88.13±2.27[a]	88.23±4.03[a]	88.43±3.44[a]
HCO_3^- (mmol/L)	23.17±5.32[a]	19.47±0.60[a]	19.73±0.75[a]

Values are presented as mean ± standard deviation (n = 5). The mean on the same row with different letter superscripts is significantly different (P < 0.05) from any paired value and vice versa.

Table 8. Lipid profile parameters in rats treated with *G. longipetala* leaf extract.

Parameter	Control	GL leaf extract (400 mg/kg)	GL leaf extract (800 mg/kg)
Cholesterol (mg/dl)	112.27±3.39[b]	104.80±4.77[a,b]	98.47±2.84[a]
HDL (mg/dl)	72.20±1.82[a]	73.63±1.17[a]	73.40±1.15[a]
TAG (mg/dl)	109.80±5.31[a]	108.03±3.71[a]	105.20±1.14[a]
LDL (mg/dl)	18.11±2.33[b]	9.56±4.96[a]	4.03±1.65[a]
VLDL mg/dl)	21.96±1.06[a]	21.61±0.74[a]	21.04±0.23[a]

Values are presented as mean ± standard deviation (n = 5). The mean on the same row with different letter superscripts is significantly different (P < 0.05) from any paired value and vice versa.

flavonoids and phenolics established natural antioxidants, which have been mobilized in the fight against oxidative stress diseases (Orieke et al., 2018; Foresti et al., 2005). Apart from being healing sources, some phytochemicals in plants are known to be toxic to living systems. For example, plant phytotoxin (aristolochic acid) is carcinogenic even at low doses. Some phytochemicals interfere with the absorption of food nutrients while others may be pro-oxidants and therefore may induce oxidative stress via inhibition of the antioxidant mechanisms (Yu et al., 2021), and increased formation of reactive oxygen species (ROS). This makes toxicity evaluation a paramount activity in the ongoing search for new and cheaper treatment alternative from plant sources.

The zero mortality and absence of obvious toxicity signs in this study following acute toxicity testing of *G. longipetala* leaf extract, even at 5000 mg/kg treatment dose suggests that extract from the plant may be safe for oral use in disease management. Existing guidelines for acute toxicity testing had stipulated that mortality is the expected outcome in such tests and that the observance of zero mortality within a population treated with a dose range of the substance at which mortality is expected indicates that the agent is well tolerated or that it may not be toxic (OECD, 2001). This is the basis for our conclusion on the safety of *G. longipetala* leaf extract. Similar conclusion was reached in a prototype study in which the acute toxicity effect of a plant extract was

Table 9. Serum antioxidant parameters in rats treated with *G. longipetala* leaf extract.

Parameter	Control	GL leaf extract (400 mg/kg)	GL leaf extract (800 mg/kg)
GSH (U/L)	58.33±4.51[a]	61.10±1.14[a,b]	64.80±2.26[b]
GPx (U/L)	47.87±2.29[a]	51.40±0.92[a,b]	53.73±2.12[b]
SOD (U/L)	29.67±1.42[a]	31.20±1.97[a]	34.50±0.92[b]
CAT (U/L)	19.73±2.87[a]	23.20±0.70[a]	23.90±2.87[a]
MDA (mg/dl)	0.40±0.02[a]	0.40±0.03[a]	0.41±0.03[a]

Values are presented as mean ± standard deviation (n = 5). The mean on the same row with different letter superscripts is significantly different (P < 0.05) from any paired value and vice versa.

established (Onoja and Anaga, 2013).

Results of the sub-acute toxicity evaluation agree with that of the acute toxicity, and further confirm the non-toxic effect of *G. longipetala* leaf extract. Values of haematological, liver function and renal function parameters in the extract treated groups are not significantly different from those of the control rats and may be an indicator of non-toxicity. Clinically, checking number of red blood cells, packed cell volume and haemoglobin concentrations have been used to assess anaemic conditions and its severity and also to monitor responses of patients to treatment, even as gross elevations in liver function parameters like alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and total bilirubin and renal function parameters including urea and creatinine all indicate that the liver and kidneys are diseased or are being threatened severely (Akomas et al., 2015). However, values obtained for these parameters only suggest that the extract may be safe and poses no toxicity threat to body systems. The decline observed in serum lipids (total cholesterol, triglycerides and low-density lipoprotein cholesterol) suggest that the extract may contain bioactive compounds with hypo-lipidaemic properties, which could be of value in the control of cardiovascular traumas associated with hyperlipidaemia. The results of the present study on phytochemical analysis showed the presence of saponins in *G. longipetala* leaf extract. Saponins reduce serum lipids levels by inhibition of dietary fat absorption and also by inhibiting pancreatic lipase activity (Xu et al., 2018). Improvements observed in the levels of serum antioxidant parameters like reduced glutathione, glutathione peroxidase, superoxide dismutase and catalase may be due to the presence of flavonoids and phenolic compounds in the extract. In addition to that, natural antioxidants like vitamins C and E are also reportedly present in *G. longipetala* leaves since the extract increased the serum levels of these vitamins in treated animals (Ojobo, 2021).

Compounds identified following GC-MS analysis of *G. longipetala* have been fingered in a number of important biological activities. For example, sparteine is well known for its anti-arrhythmic activity and has been used to reduce the incidence of ventricular tachycardia and fibrillation, and also to reduce heart rate and blood pressure (Silva et al., 2014). In the pancreas, sparteine induces insulin and glucagon secretion and by that helps in the regulation of blood sugar (Fornasini et al., 2012). Its protective role against DNA damage in diabetics has also been reported (Farghaly and Hassan, 2012). Other pharmacological effects attributable to sparteine are induction of uterine contractility, and possession of diuretic, anti-inflammatory and antibacterial activities (Flores-Soto et al., 2006). Ethyl oleates are used as plasticizers and lubricants; biological additives and hydraulic fluids (34-35). Kaempferol, catechin, flavon-3-ol, anthocyanins, and other subgroups of the major secondary metabolite flavonoid identified in the extract are undoubtedly natural phenol and antioxidants which in addition to antioxidant effects are also anti-inflammatory, hypocholesterolemic, anti-cancer, nematicide, hepatoprotective, antihistaminic, antieczemic, anti-acne, 5-alpha reductase inhibitor, anti-androgenic, and anti-arthritic agent (Praveen et al., 2010). Oleic acid lowers serum concentrations of bad cholesterol but increases the good one (HDL) (Akoh and Min, 2008), thereby possesses cardio-protective activities (Hazarika et al., 2002; Gillingham, 2011).

Octadecene found in higher percentages in the hexane fraction of facultative marine fungi *Aspergillus ustus* (Oleinikova et al., 2011) and root extract of *Plumbago zeylanica* (Ajayi et al., 2011), in human and animal model studies demonstrated significant anticancer, antioxidant and antimicrobial activities (Mishra and Sree, 2007; Lee et al., 2007; Vinay-Kumar et al., 2011).

Conclusions

Findings here have shown that *G. longipetala* is safe for oral use and poses no threat to body physiology having demonstrated a high margin of safety following acute and sub-acute toxicity evaluations, practically all the parameters evaluated are within normal values. Thus, it is probable that the hypolipidaemic activity maybe

attributable to its abundant saponins composition and the other identified GC-MS compounds; albeit more research will be required to confirm this. The phytochemical components attest to the enormous healing benefits, which the plant possesses. Further studies are required to effectively harness the medicinal benefits of *G. longipetala*.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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