

**Tropical Journal of Phytochemistry & Pharmaceutical Sciences**Available online at <https://www.tjpps.org>**Original Research Article****In-vitro Anti-inflammatory and Antioxidant Potentials of Methanol Extract of *Uvaria chamae* (Bush Banana) Leaves**Godfrey R. Kweki<sup>1,\*</sup>, Anthony Orhu<sup>2</sup>, Ugochukwu Uzuegbu<sup>1</sup>, Okeroghene S. Iwhiwhu<sup>2</sup>, Oghenenyore A. Ohwokevw<sup>3</sup><sup>1</sup>Department of Medical Biochemistry, Delta State University, P.M.B. 1, Abraka<sup>2</sup>School of Science and Technology, Delta State School of Marine Technology, Burutu,<sup>3</sup>Department of Biochemistry, Delta State University of Science and Technology, P.M.B. 5, Ozoro**ABSTRACT**

Plants existence is pivotal for human survival, they do not only serve as a source of foliage and economic value but also provide an alternative source of medicine in combating various metabolic disorders. The preliminary investigation of the bioactive composition, *in-vitro* antioxidant, and anti-inflammatory properties of methanol extract of *Uvaria chamae* leaves was evaluated in this study. The antioxidant activities were assessed using four various assays; Nitric oxide (NO) and 2, 2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activities, Reducing power (RP), and Total antioxidant capacity (TOAC). *In-vitro* anti-inflammatory potentials were quantified using membrane stabilization, albumin denaturing test, and protease inhibitor test in addition to qualitative and quantitative plant phytochemical assays. The result obtained detected the presence of phenol, flavonoids, alkaloids, terpenes, and tannins from the qualitative phytochemistry. However, Flavonoid recorded the highest concentration of 1.21mg/gCAE while the least was alkaloid 0.41mg/gATE. The *in-vitro* antioxidant and anti-inflammatory activities of the extract showed a significant ( $p < 0.05$ ) increase in the activities of the extract in a concentration-dependent manner, while the respective IC<sub>50</sub> values showed that the extracts obtained higher values in comparison ( $p < 0.05$ ) to the respective standard for the antioxidant activities. Conversely, the extract IC<sub>50</sub> values for the anti-inflammatory activities were lower when compared ( $p < 0.05$ ) to the aspirin. The results of these investigations, suggest that *Uvaria chamae* leave methanol extracts have antioxidant and anti-inflammatory properties. The mechanism of action may be due to its high content of flavonoids, which are known to inhibit membrane lysis, denaturation of albumin, and the activity of proteases. This study supports the utilization of *Uvaria chamae* in folk medicine.

**Keywords:** Phytochemical, Antioxidant, Anti-inflammatory, free radicals, *U. chamae*

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**Copyright:** © 2024 Kweki *et al.* This is an open-access article distributed under the terms of the [Creative Commons Attribution License](#), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.**Introduction**

Herbal plants are the pivot for healthcare delivery in low-tier countries. The vitality of plants in addressing ailing conditions dates back to time in memorial and even the present day.<sup>1,2</sup> Existing literature has it, that the use of synthetic drugs has its setback (adverse effects) pharmacologically and also their high cost has limited their affordability among the common populace. Food nutrients are needed for general body function and maintenance, which can be sourced from plants.<sup>3,4</sup> As a byproduct of cellular metabolic pathways, reactive oxidants of both oxygen and nitrogen origin are generated.<sup>5,6</sup> The activities of this endogenous free radical mediate cellular aberrations in cells resulting to disruption of the cell membrane. The disturbance of the cell layer engenders the growth of materials all through the cell without severe metabolic guidelines, accordingly uncovering these essential biomolecules (protein, sugars, lipids, and nucleic acids) to oxidations.<sup>7</sup>

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The interaction of the oxidative products (DNA adducts, carbonyl formation, ketoamines and ketoaldehydes, aldehydes and malonyl aldehydes, 4-hydroxy-2-nonenal) of these biomolecules with other cellular components of the cell further result in the cellular distortion of its equilibrium leading to health conditions such as diabetes, cancer, tumor, cataract, asthma, hypertension, anemia, atherosclerosis, etc.<sup>8</sup> Reports have stated that reactive species activity is an underlying factor in almost all uncontrolled conditions, which is mediated by the activities of free radicals.<sup>9</sup> Cellular equilibrium distort favorably towards pro-oxidant inferring depletion in the cell protective guards. Invariably during pathological conditions, one major target is restoring cellular normalcy, is the enhancement of the antioxidant levels in the system.<sup>10</sup> Enhancing the antioxidant levels of the cells are easily and economically achieved in less developed country via the supplementation of bioactive ingredients from plant origin.<sup>11</sup> Literature has revealed that these compounds mediate free radicals scavenging, inhibitory, break bond chain reactions etc. and such they successfully anneal the activities of these free radicals within the cell system.<sup>12,13</sup> *Uvaria chamae* is a shrubby plant common in the Niger Delta region and belongs to the family of *Annonaceae*.<sup>14,15,16</sup> The plant is aromatic with several traditional folklore attached to it.<sup>15</sup> The plant has the common name "bush banana". Local ethnic groups in Nigeria such as the Igala named it as Ayiloko, Yoruba: Okooja, Hausa: Kaskaifi, and Ghana: Akotompo.<sup>17</sup> Among the Urhobo ethnic group, is called Akpata, while the leaves are referred to as Ebe re Akpata. However, studies have shown that seed of *U. chamae*. possess anti-radical activities *in-vitro* against 1,1-diphenyl-2-picrylhydrazyl (DPPH), 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid) (ABTS), metal chelating activities and reducing power using various solvent (aqueous, methanol and ethanol).<sup>18</sup> The anti-inflammatory, anti-diabetic, anti-

microbial, anti-malarial and hepato-protective activity, anti-venom activity, etc. of the seed and roots are well documented using albino Wistar rats and albino mice.<sup>14,15,19,20, 16</sup> Conversely, there are no scientific investigations on the leaves, thus this study aimed to evaluate biological activities of the methanol extract of *U. chamae* leaves, this is geared to bring to lime-light the proper utilization of the leaves as a source of foliage and folk medicine, as the plant species is endangered and one the many under-utilize plants.

## Materials and Methods

### Materials

All reagents used for this study were of analytical grade.

### Collection of Plant Sample and Identification

Fresh leaves of *U. chamae* were harvested from the bush of Urhovie-Abraka (Latitude 5° 47' 21.9552' N Longitude: 6° 6' 8.4492' E) in Ethiopia, East Local Government, Delta State Area in February 2022. The botanical identification and authentication were done in the Plant Biology and Biotechnology Department of the University of Benin by Dr. Akinnibosun Henry Adewale. The voucher number of the specimen was assigned as UBH-U353 and deposited at the Department of Plant Biology and Biotechnology Herbarium. University of Benin

### Preparation of extract

Fresh Leaves of the study plant were thoroughly washed in clean water to remove debris. They were air-dried at 25°C for a period of four weeks. The dried leaves were pulverized to powder using a manual grinder. Cold maceration for 72 hours was used to extract one hundred gram (100g) of coarsely crushed leaves with 400ml (70% v/v) of methanol. The extract was then filtered through cheesecloth with fine pores, and the filtrate was filtered for the second time using Whatman No. 1 filter paper. The resulting extract was concentrated under a rotary evaporator at 50 °C to yield a dark brown mass and then stored at 4 °C for further study.

### Qualitative phytochemical screening.

Using standard methods as reported by<sup>21,22</sup>, qualitative phytochemical investigation was done on the extract of *Uvaria chamae* to check for the occurrence of saponins, phlobatanins, cardiac glycosides, flavonoids, tannins, phenols, steroids, terpenes, thiols and alkaloids.

### Quantitative phytochemical screening

Quantitative phytochemical determination for total phenol, total tannins, total flavonoids, and total alkaloids was done on the extract of *Uvaria chamae* leaves using the method of.<sup>23,24, 25</sup> respectively as previously reported.<sup>26</sup>

### In-Vitro antioxidants activities

*In-vitro* antioxidant activities; Nitric oxide (NO), free radical scavenging activity, and total antioxidant capacity were all determined using the methods described by<sup>27,28,29</sup> Marcocci *et al.* (1994), Oyaizu, (1986) and Prieto *et al.* (1999) respectively.

### In-vitro anti-inflammatory activities

The *in-vitro* anti-inflammatory activity of *Uveria chamae* methanolic leaf extract was investigated by the assessment of albumin denaturation inhibition, membrane stabilization and anti-proteinase activity as reported by.<sup>30,31</sup>

Data Analysis: Statistical analysis was performed on all the data. When comparing treatment groups, one-way ANOVA was used to test for differences in the values, which were presented as Mean ± Standard deviation. At the 95% confidence level (p<0.05), or p-values less than 0.05, the results were deemed significant

## Results and Discussion

African is blessed with wide diverse species of both plants and animals. Most plant in Africa are largely underutilized to their full potentials and as such scientific investigation towards plant derived foods supplementation and medicinal bioactive compounds for livestock and

human always keep evolving since time in memorial. The various bioactive ingredient found in plant are varies slight from one organ to another as deciphered from past studies. Herbal medicine is a very common practice in developing and underdeveloped nations. One measure in furnishing health status is crucial to have a thorough understanding of the antioxidant and inflammatory potentials of herbs. A study of the phytochemistry, *in-vitro* antioxidant and anti-inflammatory of the methanol leaf extract of *U. chamae* constitutes one of the scientific procedures to ascertain the medicinal attributes of this endangered plant. These biological activities strongly infer positive indications to their therapeutic potentials.

The qualitative and quantitative phytochemistry investigations carried out on the methanol leaf extract of *U. chamae* depict the presence of phenols, alkaloids, tannins, flavonoids, and terpenes. Among ten bioactive compounds tested for, flavonoids showed abundant presence with the higher concentrate of 1.21±0.00mg/g CAE while phenol, tannins, and alkaloids showed concentrations of 0.65±0.00mg/g GAE, 0.66±0.02mg/g TAE, and 0.41±0.00mg/g ATE respectively as shown in **Tables 1 and 2**. An appreciable quantity of these compounds (alkaloids, tannins, flavonoids, and alkaloids) were observed from the methanol extracts of the leaves. The presence suggests the ability of this plant to play a major role as an antidiarrhoeal, antihemorrhagic and anti-inflammatory.<sup>32</sup> Studies have depicted that the alkaloids compounds mediate analgesic properties and response to inflammatory actions of foreign invading pathogens<sup>37</sup>. The structural orientation of these bioactive molecules highly influenced their medicinal activities. Functional groups such as OH groups, phenolic ring, thiol groups, and specific amino acid residues of these compounds are the biological tools employed interfering with mechanisms and mode of action of pathogenic microbes<sup>38</sup>.

In this perspective, the availability of free hydroxyl group of phenol and flavonoids in conjunction with other vital functional groups may be accountable for their antiradical annealing effect and enzyme inhibiting potentials of bioactive compounds observed in this present study.

**Table 1:** Qualitative phytochemistry of *U. chamae* methanol leaf extract

Phytochemicals	Presence
Saponins	-
Phlobatannin	-
Cardiac glycoside	-
Flavonoids	+
Tannins	+
Phenol	+
Terpenes	+
Steroids	-
Alkaloids	-
Thiols	-

Key: + = Presence, - = Absent

**Table 2:** Quantitative phytochemical analysis of methanol leaf extract of *U. chamae*

Phytochemical	Concentration (mg/g)
Phenol (GAE)	0.65 ± 0.00
Flavonoid (CAE)	1.21 ± 0.00
Tannin (TAE)	0.66 ± 0.02
Alkaloid (ATE)	0.41 ± 0.00

Values are Mean ± Standard deviation of triplicate determinations. GAE = Gallic acid equivalent, CAE = Catechin equivalent, TAE = Tannic acid equivalent, ATE =Atropin equivalent.

However, the results present study further confirm the previous study of <sup>19</sup> characterized by the presence of alkaloids and flavonoids as well as tannins and methanol, the most suitable solvent for the extraction of polar and non-polar compounds from plant materials such as leaves.

The present investigation of methanol leaf extract of *U. chamae* shows significant ( $p < 0.05$ ) increase in the in-vitro anti-oxidants activities of DPPH (1,1-diphenyl-2-picryl hydrazyl), NO (Nitric Oxide) RP (Reducing power) TAOC (Total antioxidant capacity) in a dose-dependent manner as presented in Table 3 below. Bioactive molecules with anti-radical properties are capable of annealing the deleterious activities *in-vitro* via various mechanism such as electron deprotonation, radical quenching and scavenging, which also could be done via combination of two or more of the above mechanism mention<sup>39</sup>. The affirmation of this properties are connoted by the colour reduction in the DPPH, NO, reducing power and total antioxidant capacity, of purple to yellow, dark pink to light pink, yellow to prussian blue, lemon green to dark green. Flavonoids are phytochemicals present in *Uvaria chamae* and possess antioxidant capacity recently reported by.<sup>33</sup> Flavonoids terminate free radicals such as superoxide, hydroxyl radicals and lipid peroxy that can occur due to metabolism and oxidative stress.<sup>34</sup> It helps increase the response to disease-causing mediators such as viruses and carcinogens. IC<sub>50</sub> of the free radical scavenging activities of the methanol leaf extract of *U. Chamae* extract are presented in Table 4.

The ant-inflammatory activity of herbal plants are linked to their phytoactive ingredient content such as saponins, alkaloids, tannins, cardiac glycosides and flavonoids.<sup>26</sup> The methanol extract of *U. chamae* show a significant ( $p < 0.05$ ) increased in-vitro anti-inflammatory activities (Albumin denaturation, Antiproteinase and Membrane stabilization) in a dose-dependent manner (table 5). A glance at the extract efficacy relative to aspirin depicts low inhibitory activities (IC<sub>50</sub>) as connoted in

table 6. The observed anti-inflammatory properties of the extract can be attributed to its bioactive compounds. These bioactive compounds include flavonoids, tannins, saponins and alkaloids. These phytochemicals are known to have anti-inflammatory potentials.<sup>26</sup> Most flavonoids exert their anti-inflammatory effects by inhibiting enzymes that produce eicosanoids such as lipooxygenase, phospholipase A2, and COX; Other means include inhibition of histamine release, phosphodiesterase, protein kinase and transcriptional enzyme activation.<sup>35</sup> Tannins is known to reduce inflammation by scavenging cellular oxidants and blocking inflammatory mediators like cytokines, COX-2, and more.<sup>36</sup>

### Conclusion

Phenols are high have antioxidant activities, while flavonoids are effective in anti-inflammatory actions. The existence of both in the methanol extract of *U. chamae* leaves strongly reveals the extract's potential to ameliorate oxidative stress and inflammatory-induced diseases. These biological attributes buttress its usefulness in folklore medicine.

### Conflict of Interest

The authors declare no conflict of interest.

### Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

**Table 3:** *In-vitro* Antioxidant Activity of Methanol Leaf Extract *U. Chamae*

Conc. (mg/ml)	% Inhibition		700nm	695nm
	DPPH	NO	RP	TAC
0.20	20.1 ± 0.90 <sup>a</sup>	33.5 ± 0.07 <sup>a</sup>	0.43 ± 0.02 <sup>a</sup>	0.36 ± 0.03 <sup>a</sup>
0.40	33.6 ± 1.29 <sup>b</sup>	45.3 ± 2.10 <sup>b</sup>	0.62 ± 0.01 <sup>b</sup>	0.55 ± 0.01 <sup>b</sup>
0.60	39.5 ± 0.42 <sup>c</sup>	54.9 ± 0.70 <sup>c</sup>	0.75 ± 0.03 <sup>c</sup>	0.76 ± 0.02 <sup>c</sup>
0.80	44.9 ± 0.99 <sup>d</sup>	57.4 ± 0.37 <sup>c</sup>	0.89 ± 0.24 <sup>d</sup>	1.06 ± 0.03 <sup>d</sup>
1.00	52.4 ± 1.52 <sup>e</sup>	64.5 ± 0.41 <sup>d</sup>	1.08 ± 0.01 <sup>e</sup>	1.36 ± 0.02 <sup>e</sup>

Values are means ± standard deviations of quadruplet determinations. Values not sharing common superscript on the same column differ significantly ( $p < 0.05$ ). DPPH = 1,1-diphenyl-2-picryl hydrazyl, NO = Nitric oxide, RP = Reducing power, TAC = Total antioxidant capacity

**Table 4:** IC<sub>50</sub> of the free radical scavenging activities of the methanol leaf extract of *U. Chamae*

Parameters	Methanol (mg/ml)	Standard (mg/ml)
TAC	0.35 ± 0.02 <sup>a</sup>	0.03 ± 0.00 <sup>b</sup>
Reducing power	0.28 ± 0.06 <sup>a</sup>	0.04 ± 0.00 <sup>b</sup>
Nitric oxide	0.57 ± 0.03 <sup>a</sup>	0.085 ± 0.00 <sup>b</sup>
DPPH	0.91 ± 0.01 <sup>a</sup>	0.06 ± 0.00 <sup>b</sup>

Values are means ± standard deviations of triplicate determinations. Values not sharing common superscript on the same row differ significantly ( $p < 0.05$ ). Standards compounds are as follows; DPPH & reducing power = ascorbic, nitric oxide = catechin, TAC = gallic acid

**Table 5:** *In-vitro* Anti-inflammatory Activity of Methanol leaf Extract of *U. chamae*

Conc. (mg/ml)	% Inhibition		
	Albumin denaturation	Antiproteinase	Membrane stabilization
0.02	37.5 ± 0.31 <sup>a</sup>	14.2 ± 0.71 <sup>a</sup>	23.4 ± 0.94 <sup>a</sup>
0.04	39.0 ± 0.68 <sup>b</sup>	17.6 ± 0.52 <sup>b</sup>	33.6 ± 0.47 <sup>b</sup>
0.06	40.9 ± 0.53 <sup>c</sup>	21.5 ± 0.02 <sup>c</sup>	38.2 ± 0.47 <sup>c</sup>
0.08	42.5 ± 0.31 <sup>d</sup>	23.7 ± 0.78 <sup>d</sup>	46.3 ± 4.41 <sup>d</sup>
0.10	45.3 ± 0.23 <sup>e</sup>	29.3 ± 0.68 <sup>e</sup>	54.8 ± 0.67 <sup>e</sup>

Values are means ± standard deviations of quadruplet determinations. Values not sharing common superscript on the same column differ significantly ( $p < 0.05$ ).

**Table 6:** IC<sub>50</sub> of the anti-inflammatory activities of the methanol leaf extract of *U. chamea*

Parameters	Methanol (mg/ml)	Aspirin (mg/ml)
Antiproteinase	0.16 ± 0.02 <sup>a</sup>	0.98 ± 0.01 <sup>b</sup>
Membrane Stabilization	0.21 ± 0.02 <sup>a</sup>	0.86 ± 0.00 <sup>b</sup>
Albumin denaturation	0.16 ± 0.01 <sup>a</sup>	0.90 ± 0.00 <sup>b</sup>

Values are means ± standard deviations of triplicate determinations. Values not sharing common superscript on the same row differ significantly (p<0.05).

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