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Determination of Phytochemicals, Proximate and Anti-nutritional Composition of the Aerial Parts of *Lepidagathis alopecuroides* (vahl).

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Abstract:*Lepidagathis alopecuroides (Vahl) is a perennial shrub belonging to the family Acanthaceae and is traditionally used in the treatment of stomach pain and diarrhoea. The plant also exhibits larvicidal, piscicidal and insecticidal properties due to its bioactive constituents. This study evaluated the phytochemical constituents, proximate composition, secondary metabolites and anti-nutritional contents of the aerial parts of Lepidagathis alopecuroides using standard analytical methods.Qualitative phytochemical screening revealed moderate amounts of saponins, flavonoids, terpenoids and alkaloids, while tannins, quinones, phenols, coumarins, phytosterols, phlobatanins and anthraquinones were detected in trace amounts. Proximate analysis showed the presence of ash (10.01 ± 0.035%), crude fibre (8.15 ± 0.098%), lipid (4.80 ± 0.042%), protein (24.33 ± 0.248%), carbohydrate (51.82 ± 0.219%) and calorific value (347.76 ± 0.269 Kcal/100 g). Quantitative analysis of secondary metabolites revealed flavonoids (32.16 ± 3.11 mg/100 g), alkaloids (4.17 ± 0.76 mg/100 g) and saponins (2.70 ± 0.03 mg/100 g). The anti-nutritional contents were phytate (16.38 ± 0.20 mg/100 g), oxalate (25.81 ± 0.51 mg/100 g), phenol (7.28 ± 0.20 mg/100 g), tannins (1.30 ± 0.02 mg/100 g) and hydrogen cyanide (0.40 ± 0.01 mg/100 g). The results indicate that Lepidagathis alopecuroides possesses appreciable nutritional and phytochemical constituents with relatively low levels of anti-nutritional factors. Appropriate processing methods such as boiling and cooking may further reduce the anti-nutrient contents, thereby enhancing its nutritional and medicinal value for possible dietary and pharmaceutical applications.*

Keywords: *Lepidagathis alopecuroides, proximate composition, phytochemicals, anti-nutrients*

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1.0 Introduction

Plants play important roles in sustaining life by providing essential nutrients required for the growth, development and maintenance of humans and animals. Some of these plants such as *Telfairia occidentalis*, water leaf, carrots, lettuce, cabbage, onions and broccoli are consumed as edible vegetables either in raw form as salads or after cooking. Other edible shrubs, such as Gnetum africanum, Heinsia crinata, *Lasianthera africana*, and *Lepidagathis alopecuroides,* are processed and used in soups or prepared as medicinal extracts

because of their rich phytochemical composition and abundance of secondary metabolites. All edible plants provide the body with some minerals, vitamins, protein, oils, carbohydrates and energy. These plants have medicinal values due to their useful secondary metabolites (Joseph and Raj. 2010). These natural products are beneficial to humans because their antioxidant components reduce oxidative stress (Agudo *et al*., 2007), combat diseases (Farnsworth, 1996; Ugbogu *et al*., 2016) and protect body cells against damage caused by unstable free radicals (Bamola *et al*., 2018). (Aguda *et al*., 2007), fight diseases (Farnworth, 1996; Ugbogu *et al*., 2016) and protect the cells from unstable free radicals (Bamola *et al*., 2018).

According to Inoue and Craker (2014) medicinal plants serve as sources of drugs in the treatment of various diseases due to their bioactive compounds. Bamola *et al*. (2018) reported that pharmaceutical industries utilize bioactive compounds isolated from plant extracts in the formulation and development of drugs for the treatment of various diseases.

Lepidagathis alopecuroides (Vahl) is a perennial shrub belonging to the family Acanthaceae and is commonly found in riverine areas of Southern Nigeria, particularly in Rivers and Cross River States (Obomanu et al., 2005).

(Obomanu *et al*., 2005). The plant contains secondary metabolites such as tannins, alkaloids, saponins, flavonoids and glycosides, which exhibit important medicinal properties. It is also valuable as it portrays some larvicidal, piscicidal and insecticidal characteristics owing to its bioactive compounds (Obomanu *et al*., 2006).

Although previous studies have reported some biological and medicinal activities of *Lepidagathis alopecuroides*, there is limited information on its detailed phytochemical, proximate and anti-nutritional composition. Therefore, this study was designed to evaluate the phytochemical constituents, proximate composition, and antinutritional contents of the aerial parts of *Lepidagathis alopecuroides* to assess its nutritional and medicinal potential. Consequently, there has been increasing scientific interest in evaluating underutilized medicinal plants for their nutritional and pharmacological benefits.

The findings from this study may provide useful scientific information on the nutritional quality, safety and therapeutic potential of the plant for possible dietary and pharmaceutical applications.

2.0 Material and methods

2.1 Collection and preparation of samples

Fresh aerial parts of *Lepidagathis alopecuroides* were harvested at a farm in Emogha Local Government Area of Rivers State, Nigeria. It was authenticated by Prof. B. O. Green in the Department of Plant Science and Biotechnology, Rivers State University, Port Harcourt, Nigeria.

The plant was processed at the Post Graduate Laboratory of Rivers State University, Port Harcourt. The processed coarse powder was stored in a closed, dried sample bottle for further use.

2.2 Chemical Analysis

Phytochemical, proximate and anti-nutrient composition of the plant sample were determined in duplicates.

2.2.1 Proximate Analysis (A.O.A.C., 2010)

Proximate analyses of *Lepidagathis alopecuroides* were determined using the following parameters: residual moisture content, ash, crude fat, crude fibre, protein and carbohydrate. The carbohydrate content was calculated by using equation 1

$$\%Carbohydrate = 100 - \%ash - \%moisture - \%protein - \%crude\ fibre \quad (1)$$

2.3 Analysis of Antinutrient Compounds

2.3.1 Determination of total phytate

The method of Ola & Oboh (2000) was adopted for the determination of phytate by titration

against Iron (111) chloride solution. The ground plant material was extracted with 2% HCl for 3hrs and filtered. The filtrate was processed and titrated with $FeCl_3$ after adding ammonium thiocyanate as an indicator.

2.3.2 Determination of oxalate (Ologbo et al., 2000)

The plant sample was treated with 0.75 M of H_2SO_4, filtered, and 25 mL of the filtrate was titrated against 0.05M $KMnO_4$ until a faint colour was obtained.

2.3.3 Determination of tannin (Jaffe, 2003) Spectrophotometric method

Procedure: Phenol content (as a proxy for tannins) was quantified by incubating the plant extract with Folin-Ciocalteu reagent, and absorbance was measured at 250 nm using tannic acid as a standard.

2.3.4 Determination of saponin (A.O.A.C., 2010)

The sample was weighed, extracted with polar and non-polar solvents, and the residue dried and weighed.

2.3.5 Determination of Cyanogenic glycosides (Onwuka, 2005)

Procedure: Extraction of cyanide was done and followed by titration to determine the amount of hydrogen cyanide (HCN) present.

3.0 Results and Discussion

The results obtained from the phytochemical screening, proximate composition, secondary metabolite quantification and anti-nutritional evaluation of the aerial parts of *Lepidagathis alopecuroides* (Vahl) are presented in Tables 1–5 and Fig. 1.

Table 1. Phytochemicals present in *Lepidagathis alopecuroides* (Vahl) (Shaikh and Patil 2020)

Phytochemicals	Results
Saponins	++
Flavonoids	++
Alkaloids	++
Tannins	+
Quinones	+
Terpenoids	++
Phenols	+
Coumarins	+
Anthraquinones	+
Phlobatanins	+

Key: Moderate ++; Trace +

Table 2: Proximate composition of *Lepidagathis alopecuroides* (vahl)

Sample id	% Residual moisture	% Ash	% Crude fibre	% Lipid	% protein	% Carbohydrate	Caloric value Kcal
1.	0.91 ± 0.014	10.01 ± 0.035	8.15 ± 0.098	4.80 ± 0.042	24.33 ± 0.248	51.84 ± 0.219	347.76 ± 0.269

Mean ± S.D of duplicate values

Table 3: Comparison of proximate composition of Lepidagathis alopecuroides with results of other shrubs (Okezie *et al*., 2017)

Sample	% Moisture	% Ash	% Fibre	%Fat	%Protein	% Carbohydrate	% Caloric value Kcal
Lepidagathis alopecuroides	0.91 ± 0.014	10.01 ± 0.035	8.15 ±0.098	4.80 ±0.042	24.33 ± 0.248	51.82 ± 0.219	347.76 ± 0.269
**Ocimum grattisimum*	8.36	6.85	9.60	8.17	28.27	38.55	340.81
**Gnetum africanum*	10.35	7.39	9.20	6.95	24.38	41.75	327.07
**Telferia occidentalis*	10.20	5.84	8.12	6.04	25.12	44.70	333.64

***Adapted from Okezie *et al*., 2017**

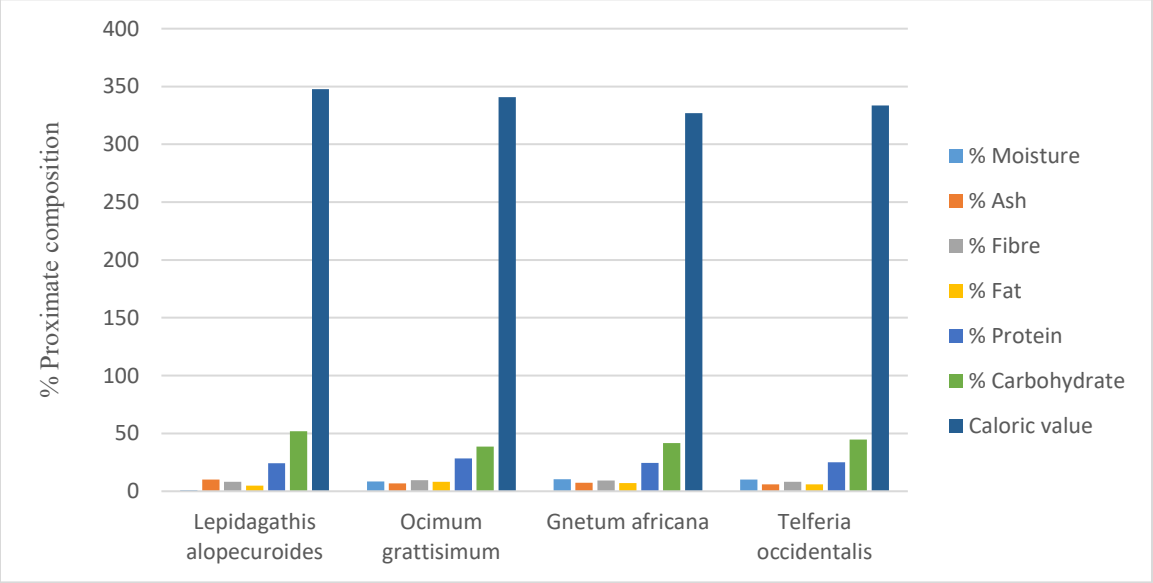

Fig. 1: Percentage Proximate comparison of Lepidagathis alopecuroides with other shrubs

Table 4: Concentration of some secondary metabolites in *Lepidagathis alopecuroides* (vahl)

Secondary metabolites	Concentration (mg/100g)
Saponins	2.7 ± 0.03
Flavonoids	32.16 ± 3.11
Alkaloids	4.17 ± 0.76

***Mean ± S.D of duplicate values**

Table 5: Anti-nutritional contents for *Lepidagathis alopecuroides* (Vahl)

Anti-nutrients	Concentration mg/100g
Phytates	16.38 ± 0.2
Tannins	1.30 ± 0.002
Phenol	7.28 ± 0.2
Hydrogen cyanide	0.40 ± 0.01
Oxalate	25.81 ± 0.51

***Mean ± S.D of duplicate values**

The analyses revealed that the plant contains nutritionally and pharmacologically important constituents, suggesting that the plant could serve as a useful dietary supplement and medicinal resource.

The phytochemical constituents detected in *Lepidagathis alopecuroides* are presented in Table 1. The results showed the presence of saponins, flavonoids, alkaloids and terpenoids in moderate amounts (++), while tannins, quinones, phenols, coumarins, anthraquinones and phlobatanins were present in trace amounts (+). These findings indicate that the plant possesses diverse bioactive compounds that may contribute to its medicinal properties. Flavonoids and phenolic compounds are known for their antioxidant activities and their ability to scavenge free radicals responsible for oxidative stress and cellular damage (Cook and Samman, 1996; Rice-Evans *et al*., 1995). Alkaloids are also reported to possess analgesic, antimicrobial and anti-inflammatory properties (Trease and Evans, 1989), while saponins have hypocholesterolemic and antimicrobial activities (Oakenfull and Sidhu, 1990; Sharan *et al*., 2012). The presence of these phytochemicals therefore supports the ethnomedicinal use of *Lepidagathis alopecuroides* in traditional medicine.

The proximate composition of *Lepidagathis alopecuroides* is presented in Table 2, while the comparison with other edible shrubs is shown in Table 3 and Figure 1. The plant contained residual moisture (0.91 ± 0.014%), ash (10.01 ± 0.035%), crude fibre (8.15 ± 0.098%), lipid (4.80 ± 0.042%), protein (24.33 ± 0.248%), carbohydrate (51.82 ± 0.219%) and caloric value (347.76 ± 0.269 Kcal). The low residual moisture content compared with *Ocimum gratissimum* (8.36%), *Gnetum africanum* (10.35%) and *Telferia occidentalis* (10.20%) reported by Okezie *et al*. (2017) suggests improved storage stability and reduced susceptibility to microbial spoilage. High moisture content is usually associated with rapid deterioration because water provides a suitable medium for microbial growth (Gbadamosi *et al*., 2011).

The ash content of *Lepidagathis alopecuroides* was higher than those of *Gnetum africanum* (7.39%), *Ocimum gratissimum* (6.85%) and *Telferia occidentalis* (5.84%) as shown in Table 3 and Figure 1. Ash content is an indicator of total mineral composition in plant materials; therefore, the high ash value suggests that the plant may be rich in essential minerals necessary for metabolic and physiological processes in the body.

The crude fibre content of the plant (8.15 ± 0.098%) was comparable to those of the other shrubs. Dietary fibre plays important roles in digestion by increasing faecal bulk, enhancing bowel movement and reducing the risk of constipation and colon cancer (Dawczynski *et al*., 2007). Ugbogu *et al*. (2016) also reported that fibre assists in the absorption of trace elements in the intestine. The appreciable fibre content therefore indicates that the plant may contribute positively to digestive health.

The lipid content of *Lepidagathis alopecuroides* (4.80 ± 0.042%) was lower than those of *Ocimum gratissimum* (8.17%), *Gnetum africanum* (6.95%) and *Telferia occidentalis* (6.04%) as presented in Table 3 and Figure 1. Although low fat content reduces caloric density and may help reduce risks of obesity and cardiovascular diseases, lipids remain essential for energy storage and transport of fat-soluble vitamins (Ononogbu, 2002; WebMD.com, 2024).

The protein content obtained for *Lepidagathis alopecuroides* was 24.33 ± 0.248%, which compares favourably with the values reported for *Gnetum africanum* and *Telferia occidentalis*. Protein is essential for tissue growth, repair and enzyme synthesis (Nelson and Michael, 2000). The relatively high protein content therefore indicates that the plant may contribute significantly to dietary protein intake. Variations in protein values among vegetables may arise from differences in soil

nitrogen content and environmental conditions (Saidu and Jideobi, 2009).

The carbohydrate content of *Lepidagathis alopecuroides* (51.82 ± 0.219%) was higher than those of the other shrubs compared in Table 3 and Figure 1. Carbohydrates are major sources of energy and are required for the production of ATP needed for metabolic activities (Udousoro and Ekanem, 2013). The high carbohydrate content contributed to the high caloric value of the plant (347.76 ± 0.269 Kcal), which was also higher than those reported for *Ocimum gratissimum*, *Gnetum africanum* and *Telferia occidentalis*. This indicates that the plant can serve as a valuable energy source for both humans and livestock.

The concentrations of selected secondary metabolites in *Lepidagathis alopecuroides* are presented in Table 4. Flavonoids recorded the highest concentration (32.16 ± 3.11 mg/100 g), followed by alkaloids (4.17 ± 0.76 mg/100 g) and saponins (2.7 ± 0.03 mg/100 g). The high flavonoid content suggests strong antioxidant potential since flavonoids are known to protect cells against oxidative stress and degenerative diseases (Rice-Evans *et al*., 1995; Krishnaiah *et al*., 2009). Spencer *et al*. (2009) and Kim *et al*. (2011) also reported the effectiveness of flavonoids in hepatoprotective and digestive health functions. Saponins are known to reduce cholesterol absorption by forming complexes with cholesterol in the intestine (Oakenfull and Sidhu, 1990), while alkaloids possess several pharmacological properties useful in drug therapy (Trease and Evans, 1989).

The anti-nutritional contents of *Lepidagathis alopecuroides* are shown in Table 5. The plant contained phytate (16.38 ± 0.2 mg/100 g), tannins (1.30 ± 0.002 mg/100 g), phenol (7.28 ± 0.2 mg/100 g), hydrogen cyanide (0.40 ± 0.01 mg/100 g) and oxalate (25.81 ± 0.51 mg/100 g). Although anti-nutrients may interfere with nutrient absorption, their concentrations in this study were relatively low and may be reduced further through processing methods such as boiling, cooking and fermentation.

The phytate content obtained was higher than the value reported for *Heinsia crinata* by Etuk *et al*. (2013). Phytates reduce the bioavailability of minerals such as calcium, iron and zinc by chelation (Ory, 1991). However, moderate consumption is generally considered safe when adequate mineral intake is maintained (Mangels, 2014). The tannin content of the plant was comparatively low, suggesting minimal interference with protein digestibility. Tannins are also known to possess antibacterial, antioxidant and anti-diabetic activities (Chung *et al*., 1998; Sharma *et al*., 2021).

The phenolic content obtained in this study was considerably lower than values reported for *Citrus maxima* extracts by Ani and Abel (2018), indicating that the plant is safe for consumption after proper processing. Phenolic compounds are known for their antioxidant and anti-inflammatory properties and their roles in reducing chronic diseases (Jimenez *et al*., 2024; Mark *et al*., 2019).

The hydrogen cyanide content of *Lepidagathis alopecuroides* was lower than that reported for *Heinsia crinata* by Etuk *et al*. (2013). Hydrogen cyanide inhibits cytochrome oxidase in the respiratory chain and interferes with ATP production (Vetter, 2000). However, cooking and boiling substantially reduce cyanide levels and minimize toxicity risks. The oxalate concentration obtained was relatively high compared with that of *Heinsia crinata*. Oxalates can reduce calcium absorption and contribute to kidney stone formation through precipitation as calcium oxalate crystals (Osagie and Offiong, 2011; Rimer *et al*., 2017; Wang and Wang, 2024). Nevertheless, proper processing methods can significantly reduce oxalate levels.

Overall, the results indicate that *Lepidagathis alopecuroides* contains appreciable quantities of nutrients and beneficial phytochemicals with relatively low concentrations of anti-nutritional factors. The plant therefore possesses significant nutritional and medicinal potential

and could contribute positively to human health when properly processed and consumed.

4.0 Conclusion

The work shows that *Lepidagathis alopecuroides* have lots of phytochemicals, high percentage of mineral composition and natural products which synergise to provide the needed nutrients for the body while combating some diseases due to these natural products.

The concentrations of anti-nutrients were extremely low. Some processing methods such as heating, boiling and cooking of the plant would further reduce the presence of these anti-nutrients making it useful to the body.

However, some, natural products like phenols and tannins are reported to fight against some diseases and infections. Therefore, the use of *Lepidagathis alopecuroides* would provide the needed nutrients for metabolic processes and mitigate against some diseases for improved health.

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Consent for publication

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Emmanuel Michael Umoh conceived and designed the study, supervised the experimental work, participated in data interpretation, and prepared the initial manuscript draft. Idongesit Ignatius Udoh contributed to sample collection, laboratory analyses, data acquisition, and literature review. James Okon Effiong assisted in the experimental procedures, statistical analysis, and validation of results. Idongesit George Etim contributed to data interpretation, critical revision of the manuscript, and overall coordination of the research activities. All authors read, reviewed, and approved the final version of the manuscript and agreed to be accountable for all aspects of the work.