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Characterization and Preliminary Structural Elucidation of Flavonoids in the Seed of *Telfairia occidentalis*

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Author's contribution

The sole author designed, analysed, interpreted and prepared the manuscript.

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ABSTRACT

Flavonoids are a group of plant compounds with a potential health benefits, including ant-oxidant, anti-inflammatory, and anti-carcinogenic properties. This study aimed to characterize the flavonoids in the seed of Telfaria Occidentalis using thin-layer chromatography (TLC) and Ultraviolet(UV) analysis.

Telfaria Occidentalis was extracted using methanol, and the extracts were analyzed using TLC and UV. The retention factors (Rf) of the flavonoids were determined, and the UV spectra were compared to those of standard compounds.

The TLC analysis revealed the presence of four flavonoids in the extracts of Telfaria Occidentalis, with Rf values of 6.00, 26.00, 37.00, 63.00 which were consistent with Vitexin, Gossypetin, Quercetin, and Kaempetrol respectively. The UV spectra of these compounds were also consistent with these flavonoids. These results shows that Telfaria Occidentalis is rich in flavonoids with potential health benefits. However, further research are needed to confirm the structures and chemical bonds of these flavonoids

Keywords: Telfaria occidentalis seed; flavonoids; characterization; chemical bonds; green tea; natural compounds; human diseases; anti-carcinogenic properties.

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1. INTRODUCTION

Have you ever wondered why some people claim that drinking green tea is good for your health? The answer may lie in flavonoid, a class of plant compounds that have a variety of beneficial properties. In this paper, I will characterize and identify the types of flavonoid found in the seeds of *Telfairia occidentalis*.

To understand the potential benefits of flavonoids, it is important to know more about their health benefits and types. Flavonoids are a group of natural compounds found in fruits, vegetables, stems, flowers, tea, and wine [1]. These compounds are renowned bioactive constituents for their positive impacts on health, pharmaceuticals, and medicine. For decades, before advancements in technology, preparations containing flavonoid constituents were used by laymen and physicians to treat human diseases [2]. This usage is attributed to their richness in antioxidants, anti-inflammatory, and anti-carcinogenic properties. Understanding the types of flavonoids in *Telfaria occidentalis* could have important implications for its future use in various industries. For instance, quercetin is found to be a strong reducing sugar, and recent research suggests quercetin acts as an antioxidant that improves normal cell survival and as a pro-oxidant that influences apoptosis in cancerous cells, thereby preventing tumor growth [3]. Furthermore, flavonoids are classified into several subclasses, including flavones, flavonols, flavanones, and isoflavones, each with its unique chemical structure and health benefits [4,5].

Flavonoids from *Telfaria occidentalis* have unique properties such as rutin and quercetin. The plant *Telfaria occidentalis*, known as Fluted Pumpkin or Ugu, is native to Africa and is known for its nutritional and medicinal properties. Its bio-active evaluation reveals that it encompasses flavonoids, glycosides, alkaloids, and resins. A study reveals the presence of tannins, reducing sugars, glycosides, saponins, and sterols in the roots of *Telfaria occidentalis* [6]; these compounds are not limited to the root but are also found in the seeds

The seeds of *Telfaria occidentalis* can be utilized in various ways, including nutritional and medicinal applications. The fermented flour of *Telfaria* cotyledons can be processed into seasonings, marmalade, high-protein infant-weaning food mixtures, and different local products in West Africa [7]. Furthermore, it comprises 3.00% moisture, 35.58% ash, 10.20% crude fiber, and 42.27% carbohydrates [8], and it is a good source of calcium, iron, zinc, and vitamins needed in human nutrition [9]. Eseyin et al [10] reported the antioxidant property of the seed plant, identifying free radical scavenging activity, two compounds (9-octadecanoic acid and 10-hydroxy octadecanoic acid), and four oily isolates .

The flavonoids in the seeds of *Telfaria occidentalis* have unique properties, but their exact structures have not been fully characterized. Therefore, the purpose of this research is to use a combination of thin-layer chromatography (TLC) and ultraviolet analysis to characterize the flavonoids in the seeds of *Telfaria occidentalis*. By understanding the properties of these flavonoids, we can better comprehend their potential applications and uses. This characterization will enhance our understanding of flavonoid properties and potential applications and may lead to the development of new uses for these compounds. The outcome of this research could have significant importance for the future use of *Telfaria occidentalis* and its flavonoids in medicine, agriculture, and other industries, and also have a significant impact on the health of people worldwide.

2. MATERIALS AND METHODS

Source of material and identification: The plant material (*Telfairia Occidentalis*) was collected from Uga, Aguata Local Government Area Anambra state Nigeria. In April 2018. It was identified by Dr. Tony, Biological Department Anambra State University Uli. The plant was dried at room temperature (25°C), pulverized by the use of an electronic grinding machine, and stored in a well-covered plastic container for use.

Chemical and solvent used:

Methanol
Chloroform
Ethyl acetate
Water

Equipment used/Apparatus used:

Silica TLC glass plate-1Ocm length by 5cm width.
Aluminum foil
Chromatographic chemper 500ml capacity
Capillary spotter
UV transilluminator
Weighing balance

Beaker
Stirrer
Stopwatch
Crucible
Water bath
Spatula
Measuring cylinder
Filter paper (Whatman).

2.1 Sample Preparation

The seeds of *Telfairia Occidentalis* were collected from farmland in Uga, the seeds were dried and then powdered.

Extraction of flavonoid: 2g of the sample was weighed and poured into a 250ml beaker, 80% aqueous methanol was added at room temperature and then stirred. It was allowed to stay for 2 hours. The whole solution was filtered through a Whatman. The whole process was repeated 3 times. The filtrates were transferred into three crucibles and, evaporated to dryness in a water bath, weighed.

The percentage of flavonoid was calculated as the weight after drying divided by the weight of the sample used multiple by 100 over 1.

$$\text{That is flavonoid \%} = \text{Weight after drying} \div \text{Weight used} \times 100/1$$

Characterization of flavonoid in the seeds of *Telfaria occidentalis*.

Procedure: Thin layer chromatography method.

A horizontal line (2cm above the base silica TLC plate) was drawn with a pencil.1Oml of the sample was collected using a capillary spotter and spotted on the line drawn on the plate (two different spots were made in different positions on the Same plate).

The mobile phase (Chloroform: Ethyl acetate: Water in the ratio of 30:15:2) was poured into the Chromatographic chemper making sure it was 1cm below the pencil mark or sample spot.The glass plate with the spotted sample was placed into the chemper, and the chemper covered with aluminum foil, and finally with its glass cover, and allowed to stand for 6 hours. After separation, the silica glass plate was removed and a pencil was used to mark the height reached by the solvent front horizontally, then the plate was allowed to dry. The glass plate was placed in a beaker containing Iodine crystals for 2mins. The different spots of the sample components were visualized in a UV transilluminator at 365nm wavelength and several spots and their corresponding color were recorded, then the distance moved by each of the components (spots) was measured and recorded using a meter rule, and a pair of dividers.

Rf (retardation factor) values were calculated and recorded using the below formula:

$$\text{Rf} = \frac{\text{Distance traveled by sample}}{\text{Distance traveled by the solvent}} = \frac{\text{Sample front}}{\text{Solvent front}}$$

Finally, the corresponding components/ isolates Separated were identified by comparing their Rf values with the standard retardation factor values of standard known compounds given with known Rf values in literature.

2.2 Confirmatory Test for Flavonoid

The sample extract was taken in a test tube and a few drops of dilute NaOH solution. An intense yellow color appeared in the test tube. It became colorless when the addition of a few drops of dilute acid indicate the presence of flavonoids.

3. RESULTS

Table 1. Organoleptic characterization of seed of *Teliffaria occidentalis*

Parameters	Inference
Colour	Milk color
Texture	Powdery and soft
Odor	Unobjectionable

Table 2. Percentage yield of flavonoid in the seeds of teliffaria occidentalis number of determinations

1st	2nd	3rd	Mean value %	Sample
9.75	9.1	9.15	9.13	FLAVONOID

Table 3. Characterization of flavoniods with reference to Rf value and UV analysis

Isolates	Solvent Front CM	Sample Front CM	Rf Values	RfX 100	U.V Colour	Components Identified	Samples
A	7.9	0.5	0.06	6.00	Absorbed	Vitexin	Flavonoid
B	7.9	2.1	0.26	26.00	Black	Gossypetin	Flavonoid
C	7.9	2.95	0.37	37.00	Dule Yellow	Unknown	Flavonoid
D	7.9	5.00	0.63	63.00	Bright Yellow	Queceptin	Flavonoid
E	7.9	7.00	0.88	88.00	Yellow	Kaempterol	Flavonoid

4. DISCUSSION

The result of this study show that the flavonoids, Vitexin, Gossypetin, Querceptin, and Kaempterol are present in *Telfaria Occidentalis.* This characterization was done using Thin-layer chromatography (TLC) and Ultraviolet light (UV). This analysis employed the use of standards of known value and retention times, the resulting chromatograms showed unique peaks that correspond to the standards. Via the retention times and UV spectra, the four flavonoids were identified with a high degree of confidence. In details, The thin layer Chromatographic test of the flavonoid revealed five spots with the calculated Rf values to be 6. 00, 26.00, 37.00, 63.00, and 88.00 respectively. These Rf values of the isolated metabolite were coupled with the various color they showed under ultraviolet (UV) light. These help in the identification of this metabolite, the visible colors observed are Absorbed, Black, Dule yellow, Bright yellow, and Yellow respectively. This analysis helps to identify the isolated flavonoid to be Vitexin, Gossypetin, Quercetin, and Kaempterol respectively except the isolated flavonoid with the Rf value of 37.00 and Dule yellow color which has no match in the reference book.

These results showcased that TLC and UV analysis can be used as rapid, less expensive and reliable method for characterization of flavonoids in the plant extracts. But this analysis failed to identify the real structures and chemical bonds of the above flavonoids, therefore, further analysis techniques such as NMR and IR would be necessary to be conducted to confirm the purity of the compounds, to find the molecular structure and chemical bonds of the flavonoids.

Table 1, showed the physical characterization of the seeds of *Telfairia Occidentalis*. It is milky in color and has an Unobjectionable Odour.

Table 2, showed the mean value of flavonoid in the seeds of *Telfaira Occidentalis*.

Table 3 showed the characterization of flavonoids of *Telfaria Occidentalis* extracts seeds.

5. CONCLUSION

The results of this study identified four flavonoids in *Telfaria Occidentalis*, namely Vitexin, Gossypetin, Querceptin, and Kaempterol. These flavonoids have been shown beneficial properties in previous research works, and this study supports these findings, hence pharmaceutical companies should explore the seed due to its efficacy for treatment of diseases. However, it should be noted that not all flavonoids were detected, for instances one with Rf value of 37.00 and a Dule yellow color, hence, further research is needed to fully characterized the flavonoids present in *Telfaria* Occidentalis and also NMR, IR method should be employed to identify the structure and chemical bonds of the isolated flavonoids in the seed of *Telfaria Occidentalis.*

CONSENT AND ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Author has declared that no competing interests exist.

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