Phytochemical Properties and Effect of Temperature on Proximate and Mineral Composition of *Curcuma longa* Linn. Rhizomes Ethanolic Extract

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Authors' contributions

This work was carried out in collaboration between all authors. Author RMS supervised the work, performed the statistical analysis and contributed to the protocol (writing of the manuscript). Authors CFO, MPA, JO and BOO carried out laboratory work and contributed to the protocol. All authors read and approved the final manuscript.

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Short Communication

ABSTRACT

**Aims:** To investigate the phytochemical property of fresh *Curcuma longa* rhizomes and the effect of temperature on proximate and mineral composition of dried *C. longa* rhizomes.

**Study Design:** Activity directed antioxidant and phenolic content investigation of *C. longa* rhizomes using *in vitro* methods.

**Place and Duration of the Study:** Medicinal Plants Section, Bioresources Development Centre, Ogbomoso, Nigeria between August and October, 2016.

**Methodology:** Fresh *C. longa* rhizomes was washed and divided into five portions (A-E). Portion A-D was dried (at room temperature, 40°C, 50°C and 60°C respectively) and used for proximate and mineral composition investigation while portion E (fresh *C. longa* rhizomes) was used for screening the phytochemical composition of the rhizomes.

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Results: The result of this study showed the presence of alkaloids, tannin, saponin, phenol, cardiac glycosides, phlobatannin, flavonoids, anthraquinones, steroids, terpenes and cardenolides in C. longa rhizome. Also there was a significant $(P<0.05)$ increase in crude protein, crude fat and ash at $50^\circ C$ and a significant $(P<0.05)$ increase in crude fat and carbohydrate at $60^\circ C$ when compared with room temperature and $40^\circ C$. Also, at $40^\circ C$ there was a significant $(P<0.05)$ decrease in sodium, calcium and phosphorus when compared with room temperature, while at $50^\circ C$ there was a significant $(P<0.05)$ increase in sodium, potassium, magnesium, calcium, phosphorous and iron when compared with room temperature and $40^\circ C$.

Conclusion: In conclusion, the use of C. longa rhizomes in ethno-medicine for the treatment/management of a lot of diseases may be due to the presence of some phytochemicals, nutrients and minerals found in the plant and also, the concentration of these nutrients may be affected by temperature.

Keywords: Curcuma longa; rhizomes; mineral analysis; temperature; phytochemistry.

1. INTRODUCTION

Curcuma longa Linn. commonly known as Turmeric belongs to the Zingiberaceae family has a lot of report in literature about the use of the rhizomes in ethno-medicine for the treatment of diseases such as cough, bronchitis, diabetes, arthritis and cancer [1]. The plant is cultivated worldwide for its rhizomes including Asia, China, India, Malaysia, Nigeria and Thailand because of its medicinal values, its use as spice in food and for its flavor in tea [2,3,4]. The plant has a short stem with large oblong rhizomes, which are often branched and brownish yellow in colour [5]. C. longa rhizome is added to the composition of many traditional remedies for the treatment of diseases and it is also locally applied to wound as an antiseptic for skin abrasions and cuts [6]. Also, the fiber in C. longa cleanses the digestive tract by removing potential carcinogens from the body. It was also reported by [7], that the fiber in C. longa rhizome adds bulk to the food and prevents the intake of excess starchy food and may hence prevent metabolic conditions like hypercholesteremia and diabetes mellitus [1]. There are also reports from literature that C. longa could be a good source of protein, carbohydrate and minerals including calcium, sodium, potassium and phosphorus [8]. Storage of C. longa rhizomes at a lower moisture level is paramount to increasing its shelf life hence supporting post-harvest handling. Also, the temperatures at which moisture is reduced in C. longa could affect the proximate and mineral composition. It is for this reason therefore, that this study investigated the effect of temperature (room temperature, $40^\circ C$, $50^\circ C$ and $60^\circ C$) on proximate and mineral composition of C. longa rhizomes ethanolic extract. The phytochemical properties of the extract were also investigated.

2. MATERIALS AND METHODS

2.1 Plant Material

The rhizomes of Curcuma longa Linn. were obtained from Medicinal Plants Section, Bioresources Development Centre, Ogbomoso, Nigeria. The plants were identified and authenticated at the Department of Botany, Obafemi Awolowo University, Ile-Ife, Osun State, Nigeria. Curcuma longa Linn. specimen with voucher number: IFE-17577 was deposited at Ife Herbarium, Obafemi Awolowo University, Ile-Ife, Nigeria.

2.2 Preparation of Curcuma longa Linn. Rhizomes Ethanol Extract

Fresh Curcuma longa rhizomes was washed and divided into five portions (A-E). Portion A-D was weighed to a mass of 18 g each of C. longa rhizomes cut into tiny pieces and dried at room temperature, $40^\circ C$, $50^\circ C$ and $60^\circ C$ respectively. The dried rhizomes at different temperatures (portion A-D) were separately pulverized into powder, and 10 g each was macerated in 20 mL of absolute ethanol for 72 hours at room temperature on a flask shaker and filtered with Whatman No. 1 filter paper [9]. The filtrates obtained were used for screening the proximate and mineral composition of the extracts (portion A-D). The fresh C. longa rhizome (portion E) was also chopped into tiny pieces, ground and macerated in absolute ethanol for 72 hours. It was then filtered and the filtrate was used for investigating the phytochemical properties of C. longa fresh rhizomes.
2.3 Qualitative Phytochemical Screening

A mass of 10 g of fresh *Curcuma longa* rhizome ethanolic extract was screened for the presence of plant secondary metabolites using standard procedures described by [10,11,12].

2.3.1 Test for tannins

0.5 g of ground fresh *C. longa* was boiled in 20 mL of water in a test tube and filtered. Few drops of 0.1% ferric chloride were added. A green or blue black coluration confirms the presence of tannins.

2.3.2 Test for phlobatannins

Deposition of red precipitate when 0.5 g of ground fresh *C. longa* was boiled with 1% hydrochloric acid was taken as evidence for the presence of phlobatannins.

2.3.3 Test for saponins and flavonoids

2.0 g of ground fresh *C. longa* was boiled in 20 mL of distilled water in a water bath and filtered. Filtrate (10 mL) was mixed with 5 mL of distilled water and shaken vigorously. The formation of froth was taken as evidence for the presence of saponin. Aluminium chloride (3 mL of 1%) was added to 5 mL of the sample. A yellow colouration observed indicate the presence of flavonoids. Also, diluted ammonia solution (5 mL) was added to a portion of the ethanol extract followed by the addition of concentrated H$_2$SO$_4$. A pink colour in the ammonical (lower) phase indicates the presence of free hydroxyl anthraquinones.

2.3.4 Test for steroids

Acetic anhydride (2 mL) was added to 0.5 g of ground fresh *C. longa* with 2 mL of concentrated H$_2$SO$_4$. The colour change from blue to violet indicated the presence of steroids.

2.3.5 Test for cardiac glycoside

5 mL of *C. longa* ethanolic extract was treated with 2 mL of glacial acetic acid containing one drop of ferric chloride solution. This was underplayed with 1.0 mL of sulphuric acid. A brown ring at the interface indicates a deoxy sugar characteristic of cardiac glycoside.

2.3.6 Test for alkaloids

*C. longa* ethanolic extract was tested with the alkaloidal reagent Mayer’s as follows; 2 drops was added to 10 mL of the ethanolic extract. Presence of a creaming precipitate indicated the presence of alkaloids in the extract.

2.3.7 Test for phenolics

Two drops of 5% w/v of FeCl$_3$ was added to 1.0 mL of the plant extract. A greenish precipitate indicates the presence of phenolics.

2.3.8 Test for anthraquinones

3.0 cm$^3$ ground fresh *C. longa* ethanolic extract was shaken with 10.0 cm$^3$ of benzene, filtered and 5.0 cm$^3$ of aqueous ammonia solution (10% v/v) was added to the filtrate. A pink colour in the ammonical (lower) phase indicates the presence of free hydroxyl anthraquinones.

2.3.9 Test for cardenolides and dienolides

1.0 cm$^3$ of ground fresh *C. longa* ethanolic extract was added to 2.0 cm$^3$ of glacial acetic acid containing one drop of 5% w/v FeCl$_3$ solution. This was then underplayed with 1.0 cm$^3$ of the concentrated H$_2$SO$_4$. A brown ring at the interface indicated the presence of a deoxy sugar characteristic of cardenolides.

2.3.10 Test for triterpenes

1.0 cm$^3$ of ground fresh *C. longa* ethanolic extract was added to 5 drops of acetic acid anhydride followed by a drop of concentrated H$_2$SO$_4$. The mixture was steamed for 1 hour and neutralized with NaOH followed by the addition of chloroform. A bluish-green colour indicates the presence of triterpenes.

2.3.11 Test for chalcones

Ammonia solution (2 mL) was added to 5 mL of ground fresh *C. longa* ethanolic extract. Formation of a reddish colour confirmed the presence of chalcones.

2.4 Proximate Analysis

Chemical composition of the pulverized samples were determined according to [13] for crude protein (method 988.05), crude fat (method 2003.06), total ash (method 942.05), crude fibre (method 958.06), dry matter and moisture (method 967.08), while carbohydrate was obtained by difference.
2.5 Mineral Elements Compositions Determination

The percentage sodium, potassium and phosphorus composition were determined spectrophotometrically [13].

2.6 Statistical Analysis of Data

Data for each group were collected and summarized in a tabular form. Data were represented as the mean ± standard error of mean (SEM). One-way analysis of variance (ANOVA) was first used followed by Bonferroni t-test post hoc comparisons to determine the significant differences at 95% (P<0.05) using Primer (version 3.01).

3. RESULTS AND DISCUSSION

The result of this study showed the presence of alkaloids, tannin, saponin, phenol, cardiac glycosides, phlobatannin, flavonoids, anthraquinones, steroids, terpenes and cardenolides in *C. longa* rhizome (Table 1). Alkaloids present in *C. longa* rhizomes shows that they can be used in curing headache associated with hypertension, management of cold, chronic catarrh and migraine [14]. The presence of tannin in *C. longa* rhizomes shows that the plant could be used as antioxidant in the treatment of intestinal disorder like dysentery [15]. Also, saponin and flavonoid present in this plant shows that it has a strong antioxidant property [16]. Thus, the plant could be used to lower cholesterol, to improve sex hormone and also in the management of inflammations [17].

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The presence of saponins in *C. longa* rhizome could probably be responsible for the use of the rhizome in ethno-medicine as an expectorant in the treatment of cough. Flavonoids also exhibit anti-inflammatory, antiangiogenic, anti-allergic effects, analgesic and antioxidant properties [14,1].

There was a significant (P<0.05) decrease in crude protein; and an increase in crude fat and ash at 50°C. There was also a significant (P<0.05) increase in crude fat and carbohydrate at 60°C when compared with room temperature and 40°C (Table 2). Also, there was a significant (P<0.05) increase in ash content at 60°C when compared with 40°C (Table 2). A statistical significant increase in crude protein, crude fat, ash and carbohydrate values was observed across the different drying temperatures of *C. longa*. In this study, there was a significant increase in crude protein, crude fat, ash and carbohydrate with increase in temperature (Table 2). The values obtained in this study were found to be higher than values reported in previous studies [18]. The result of this study also showed a decrease in moisture content with increasing temperature. The results obtained were similar to other studies that observed a decrease in moisture with increased drying temperature which ultimately have an overall effect on the shelf life and stability of the plant [19,20]. At 40, 50 and 60°C, there was a significant decrease in (P<0.05) in moisture content when compared with room temperature (Table 2).

At 40°C there was a significant (P<0.05) decrease in sodium, calcium and phosphorus when compared with room temperature while at 50°C there was a significant (P<0.05) increase in sodium, potassium, magnesium, calcium, phosphorous and iron when compared with room temperature and 40°C (Table 3). At 60°C, there was a significant (P<0.05) increase in magnesium, calcium and iron when compared with room temperature and an increase in sodium and phosphorus when compared with 40°C (Table 3). Mineral analysis of *C. longa* rhizomes dried at different temperatures showed the presence of sodium, potassium, magnesium, calcium, phosphorous and iron. There was a significant (P<0.05) increase in sodium, potassium, magnesium, calcium, phosphorous and iron with increasing temperature (Table 3). These observed increases could probably be explained by the increase in total amount of soluble solids that results from decreased moisture content [21]. The iron content of samples dried at room temperature was much higher than values reported by [20] and [18]. However, the results were similar to values reported by [22] and [23] in similar studies.
Table 2. Proximate analysis on Curcuma longa rhizome ethanolic extract (%)

|                     | Room temperature | 40°C    | 50°C    | 60°C    |
|---------------------|------------------|---------|---------|---------|
| Crude protein       | 9.93 ± 0.04      | 9.64 ± 0.06 | 9.43 ± 0.04 | 9.33 ± 0.06 |
| Crude fat           | 3.13 ± 0.01      | 3.08 ± 0.01  | 3.26 ± 0.01 | 3.21 ± 0.01  |
| Crude fibre         | 8.29 ± 0.02      | 8.24 ± 0.02  | 8.39 ± 0.03 | 8.33 ± 0.02  |
| Ash                 | 6.71 ± 0.03      | 6.61 ± 0.03  | 7.01 ± 0.03 | 6.90 ± 0.03  |
| Moisture            | 10.91 ± 0.00     | 10.64 ± 0.03 | 9.73 ± 0.02 | 9.54 ± 0.04  |
| Carbohydrate        | 61.80 ± 0.02     | 61.84 ± 0.01 | 61.88 ± 0.01| 62.19 ± 0.03 |

Values are mean ± SEM; n = 2; a Significantly different from room temperature at P< 0.05; b Significantly different from 40°C at P< 0.05

Table 3. Mineral composition of Curcuma longa rhizome (%)

|        | Room temperature | 40°C    | 50°C    | 60°C    |
|--------|------------------|---------|---------|---------|
| Sodium | 0.06 ±0.00       | 0.05 ± 0.00 a | 0.07 ± 0.00 ab | 0.06 ± 0.00 b |
| Potassium | 1.70 ± 0.02  | 1.64 ± 0.01  | 1.81 ± 0.02 | 1.74 ± 0.01  |
| Magnesium | 0.19 ± 0.00   | 0.19 ± 0.00  | 0.22 ± 0.00 | 0.21 ± 0.00  |
| Calcium | 0.08 ±0.00       | 0.07 ± 0.00 a | 0.10 ± 0.00 ab | 0.09 ± 0.00 ab |
| Phosphorus | 0.12 ±0.00     | 0.11 ± 0.00 a | 0.13 ± 0.00 ab | 0.12 ± 0.00 b |
| Iron    | 0.22 ±0.00       | 0.21 ± 0.00  | 0.25 ± 0.00 b | 0.23 ± 0.00 a |

Values are mean ± SEM; n = 2; a Significantly different from room temperature at P< 0.05; b Significantly different from 40°C at P< 0.05

4. CONCLUSION

In conclusion, the use of C. longa rhizomes in ethno-medicine for the treatment/management of a lot of diseases may be due to the presence of some phytochemicals, nutrients and minerals found in the plant and also, the concentration of these nutrients may be affected by temperature.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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