

PHYTOCHEMICAL AND BIOCHEMICAL PROFILE OF RAW EXTRACTS AND OIL FROM *CURCUMA LONGA* RHIZOMES GROWN IN DIABO (COTE D'IVOIRE).

Abstract

Aims: The aim of this study was to determine the phytochemical and biochemical profile of *Curcuma longa* grown in Diabo, central Côte d'Ivoire.

Place and Duration of Study : Department of Biochemistry-Genetics, Unit for Training and Research in Biological Sciences, Peleforo University GON COULIBALY Korhogo, BP 1328 Korhogo; The first part of this study was carried out from April 1, 2023 to December 31, 2023; the second part was conducted from March 01, 2024 to July 31, 2024.

Methodology: Several phytochemical tests based on colorimetric reactions were used to perform qualitative and quantitative analyses of polyphenols, flavonoids and tannins. To characterize the oil and crude extracts, the indices of iodine and refractiveas well as pH and Brix were determined.

Result: The results show the presence of several secondary metabolites in the crude extracts, including polyphenols (53.1 ± 2.08 mg Eq AG/g extract), flavonoids (2.7 ± 0.22 mg Eq AG/g) and catechic tannins (9.1 ± 0.7 mg Eq C/g) in variable quantities. Biochemical analyses showed a pH close to neutral (pH = 6.68). The Brix value (23.20 ± 0.26 min), shows that *Curcuma longa* extracts, in particular EHCL, contain several soluble solids such as reducing sugars, acids, salts, pectins and nitrogenous matter. As far as the oil is concerned, the iodine value obtained was 84.1 ± 0.1 g I₂ /100 g fat; this high value is close to 100 g. The refractive index was 1.52 ± 0.61 .

Conclusion: Phytochemical and biochemical characteristics show that *Curcuma longa* rhizomes could have interesting pharmacological potential.

Key words: Characterization, *Curcuma longa*, Phytochemistry, oil. ;

1. Introduction

Medicinal plants are important for pharmacological research and drug development, not only when their constituents are used directly as therapeutic agents, but also as raw materials for drug synthesis or as models for pharmacologically active compounds [1]. Moreover, a significant proportion of the drugs used today are derived from plants. Some 50% to 60% of

pharmaceutical products are either of natural origin, or synthesized from natural compounds [2]. As pharmaceutical research on natural resources expands and new therapeutic agents are sought, the exploration of medicinal plants has become essential. In this context, *Curcuma longa* is of particular interest because of its potentially beneficial bioactive molecules for modern pharmacopoeia. The active ingredients of medicinal plants are often linked to their phytochemical and biochemical profile in crude extracts or oil; secondary metabolites or other biochemical characteristics of medicinal plants are widely used in therapeutics as preventive anti-inflammatory, antimicrobial, antiseptic, diuretic and antioxidant agents [3]. The aim of this study was to determine the phytochemical and biochemical profile of the raw extracts of *Curcuma longa* and its oil.

Material and methods

1.1. Material

1.1.1. Plant material

The plant material studied was *Curcuma longa* (Figure 1) in the form of fresh rhizomes harvested in Diabo, central Côte d'Ivoire, in December 2024. They were dried for 10 days at room temperature in the laboratory.

1.2. Methods

1.2.1. Collection of fresh *Curcuma longa* rhizomes

The rhizomes were harvested in Diabo, central Côte d'Ivoire, in December 2024. Before processing, they were cleaned and washed to remove impurities and dirt. They were then cut up into small pieces. The cut rhizomes were placed out of direct sunlight for drying. The drying process lasted 10 days.

1.2.2. Obtaining powdered *Curcuma longa*

Once dry, the rhizomes were ground to a fine powder using a grinder. The powder obtained was carefully preserved for future extractions (Figure 2).

1.2.3. Obtaining the raw extracts of *Curcuma longa* and its oil

2.2.3.1 Preparation of hydroethanol extracts of *Curcuma longa*

- Preparation of hydroethanol extract

In a hydroethanol solution of 300 mL ethanol and 700 mL distilled water, 100 grams of turmeric powder were added. Further extraction was carried out as described for aqueous extraction (Figure 3).

2.2.3.2. Oil extraction

Fat content was determined using the method defined by [4]. After extraction with Soxlet for 8 h, the hexane was evaporated using a rotary evaporator. The flask containing the fat was then placed in an oven at 45°C for 24 h to remove any residual hexane. The oil was left to cool in a desiccator for 5 min to avoid wetting the plant material. Finally, the flask containing the fat (P2) was weighed. This oil is shown in **figure 4**.



Figure 1: Fresh rhizomes of *Curcuma longa* (Kouamé, 2024)



Figure 2: Pulverized *Curcuma longa* (Kouamé, 2024)



Figure 3: Hydroethanol extract of *Curcuma longa* (Kouamé, 2024)



Figure 4: Oil from *Curcuma longa* (Kouamé, 2024)

1.2.4. Calculation of extract yields [5].

- Dry extract yield

The yield of dry extracts obtained after evaporation is calculated according to the following contribution :

$$= \frac{(\text{Mass of dry extract})}{(\text{Mass of plant matter})} \times 100$$

- Fat yield

The fat yield obtained is calculated using the following formula:

$$\text{Yield} = [(P_1 - P_2) \times 100 / P_3]$$

P₁: Weight of flask containing dry extract after evaporation (g)

P₂: Empty drum weight (g)

P₁-P₂= Fat weight (g)

P₃: Test plug (g)

1.2.5. Phytochemical study

1.2.5.1. Qualitative analysis of extracts

1.2.5.1.1. Polyphenol research

Total polyphenols were identified using the ferric chloride reaction [6]. The appearance of a more or less dark blue-black or green color indicates the presence of phenolic compounds.

1.2.5.1.2. Flavonoids research

Flavonoids were determined by the cyanidin reaction [6,7]. The use of 2-3 magnesium chips produced a heat release followed by the appearance of a pink-orange or purplish coloration. The addition of 3 drops of isoamyl alcohol intensified this coloration, indicating the presence of flavonoids.

1.2.5.1.3. Catechic tannins research

Stiasny's reagent (hydrochloric formalin solution) was used to detect the presence of catechic tannins [7,8]. Catechic tannins were characterized by the presence of large, flaky precipitates.

1.2.5.1.4. Polyterpenes and sterols in *Curcuma longa* extracts

Sterols and polyterpenes were identified using the Liebermann reaction [6,7]. The appearance of a purple or violet ring at the interface, which then changes color from blue to green, confirmed the presence of polyterpenes and sterols.

1.2.5.1.5. Leucoanthocyanins in *Curcuma longa* extracts

The formation of a red coloration confirmed the presence of leucoanthocyanins as indicated by [9].

1.2.5.1.6. Saponosides in *Curcuma longa* extracts

Ten (10) mL of plant extract was poured into a test tube. After stirring for a few minutes, the height of the foam was measured. A foam exceeding 1 cm indicates the presence of saponins. The persistence of the foam reflects the abundance of saponins.

1.2.5.1.7. Research on alkaloids in *Curcuma longa* extracts

Dragendorff's reagent and Bouchardat's reagent were used to identify alkaloids in the various extracts [10]. The formation of an orange coloration or precipitate in the first tube indicated the presence of alkaloids. The presence of a reddish-brown coloration in the second tube also indicated the presence of alkaloids.

1.2.5.2. Quantitative analysis of *Curcuma longa* crude extracts

1.2.5.2.1. Polyphenols

The method described by [11] was used to quantify polyphenols. Absorbance was measured using a UV (ultra-violet) spectrophotometer at a wavelength of 760 nm . This measurement is proportional to the concentration of polyphenols in the extract, with distilled water used as the blank.

1.2.5.2.2. Flavonoids

The method of [12] was used for flavonoid determination. The mixture was vigorously mentagitated before being assayed by UV-visible spectrophotometer at a wavelength of 510 nm.

2.2.5.2.3. Tannins

The determination of tannins in *Curcuma longa* extracts was carried out according to the method of [13]. Tannin concentrations were determined using calibration curves established with catechin (0 - 300 µg/mL).

2.2.6. Biochemical characteristics of *Curcuma longa* oil

❖ Refractive index

The refractive index was measured using a refractometer, as described by [14] .The oil was heated to 40°C for 30 min before being placed in the refractometer receptacle.

❖ Iodine index (i)

The method described by [15] was used. Released iodine was titrated with a 0.1 N sodium thiosulfate solution in the presence of a 1% (w/v) starch starch solution as a color indicator until complete decolorization. At equilibrium, the solution changes from black to colorless. The iodine value is calculated according to the following formula:

$$i = \frac{12.69 \times (V_0 - V)}{100}$$

V₀: volume in mL of sodium thiosulfate used in the control test,
V: volume in mL of sodium thiosulfate used for the test sample,
N: concentration of sodium thiosulfate solution used and m: mass of test sample' expressed in grams (g).

2.2.7 Biochemical characteristics of dry extracts

2.2.7.1. Determination of pH

The pH of the optimized *Curcuma longa* powder extract was determined by immersing the electrode of the HANNA pH meter in 50 mL of extract. Prior to this measurement, the pH meter was calibrated using two buffer solutions: pH = 4.0 and pH = 7.0. The pH value of the extract was read directly from the pH meter display.

2.2.7.2. Determination of Brix degree (°Brix)

The °Brix, which corresponds to the soluble matter content of *Curcuma longa* extract, was measured using a Leica AR 200 infrared refractometer ; three (3) drops of *Curcuma longa* extract were placed directly on the instrument's prism. The device was then turned towards a light source, and the reading was taken on the eyepiece scale, at the intersection of the graduated light and dark zones. Readings were performed in triplicate [16].

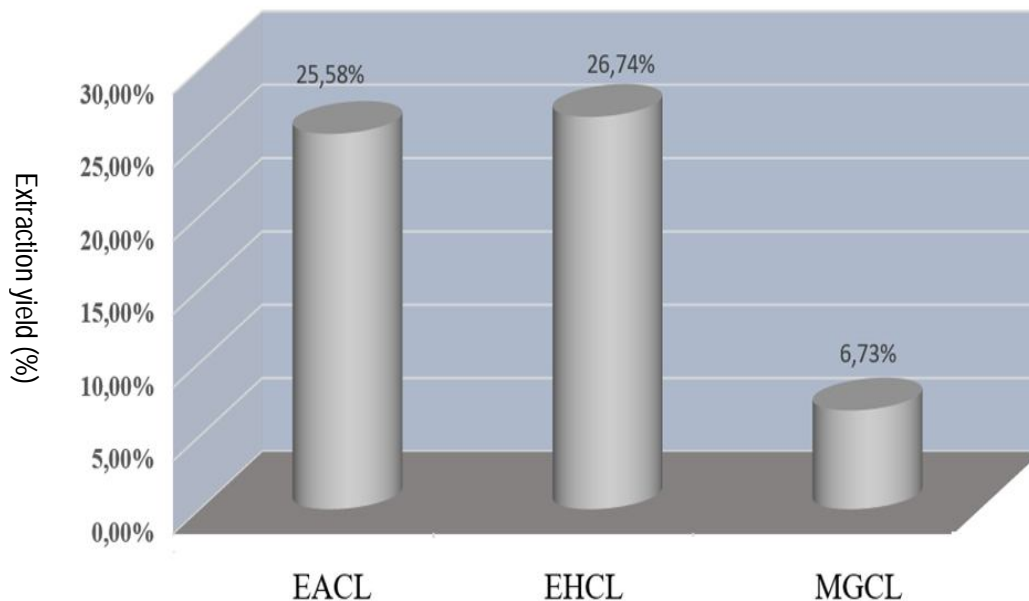
2.2.8. Statistical analysis

Statistical analysis of the results was performed using a one factor analysis of variance (ANOVA ONE WAY). Results are presented as mean and standard error of the mean (Mean ± SEM). Graphical representation of the data was performed using Graph Pad Prism 10.2.3 (Graphpad Software, Inc., USA), SPSS (IBM Corp., USA) and Excel (Microsoft Corp., USA).

2. RESULTS AND DISCUSSION

2.1. Yield

Extraction yields of the crude extracts are shown in **figure 5**. The yield of the hydroethanol extract (26.74%) is significantly higher than that obtained with the aqueous extract (25.58%); this could be explained by the solubility of turmeric's active ingredients in both ethanol and water. These yields are higher than those obtained by [17], who performed a decoction of turmeric rhizomes (10% for the aqueous extract and 7% for the alcoholic extract). The yield of *Curcuma longa* oil (6.73%) obtained in our work is lower than that obtained by [18], which is 8.44%.



EACL : *Curcuma longa* aqueous extract ; **EHCL** : *Curcuma longa* hydroethanol extract ; **MGCL** : *Curcuma longa* fat.

Figure 5: Yield of *Curcuma longa* fat and raw extracts

2.2. Phytochemical analysis

The results of the qualitative and quantitative analyses of phytochemical components are shown in **tables 1** and **2** respectively.

The results in **table 1** show that polyphenols, alkaloids, catechic tannins, leucoanthocyanins and flavonoids are present in both *Curcuma longa* extracts. This suggests that the two solvents used (water and ethanol) enabled better extraction of these secondary metabolites. Thus, *Curcuma longa* rhizomes may possess biological properties attributable to these different secondary metabolites. However, gallic tannins, terpenes and sterols, as well as saponosides, were absent in both extracts, which may indicate a lack of affinity of these secondary metabolites with the solvents used. The results shown in **table 2** confirm the presence of polyphenols, flavonoids and tannins in the raw *Curcuma longa* extracts. The polyphenol content of the hydroethanol extract is significantly ($p \leq 0.05$) higher than that of the aqueous extract. According to [19], the high phenolic content of alcoholic extracts can be explained by

the fact that ethanol is effective in extracting active ingredients; curcumin solubilizes more in ethanol than in water. With regard to the aqueous extract, our results show that the polyphenol content (19.4 mg Eq AG/g) is higher than that found by [20], which is 7.48 mg Eq AG/g. On the other hand, studies carried out by [21] recorded a polyphenol content of 18.12 mg Eq Q/g, which is close to our results. With regard to the hydroethanol extract, our results (53.1 mg Eq AG/g) are lower than those obtained by [20], who showed that the alcoholic extract (ethanol) is very rich in polyphenols, with a content of 105.92 mg Eq AG/g. In addition, studies by [22] and [23] have shown that *Curcuma longa* rhizomes are rich in polyphenols and tannins in aqueous and methanolic extracts; this is in agreement with our results obtained in the case of aqueous and hydroethanolic extracts. Similarly, studies by several authors have shown that phenolic compound content varies considerably from one species to another [24].

Table 1: Chemical composition of aqueous and hydroethanolic extracts of *Curcuma longa*

CHEMICAL GROUPS	EXTRACTS	
	Aqueous extracts	Hydroethanol extracts
Alkaloids	+	+
Total polyphenols	+	+
Leucoanthocyanes	+	+
Catechic tannins	+	+
Gallic tannins	-	-
Flavonoids	+	+
Saponosides	-	-

Legend: +: present; -: absent

Table 2: Polyphenol, flavonoid and tannin contents of aqueous and water extracts. hydroethanol of CL

Extracts	Polyphenols (mg Eq AG/g)	Flavonoids (mg Eq Q/g)	Tannins (mg Eq C/g)
EHCL	53.1 ± 2.08*	2.7 ± 0.22	9.1 ± 0.7*
AECL	19.4 ± 2.58*	1.6 ± 0.49	5.2 ± 0.73*

*: There is a significant difference ($p \leq 0.05$)

2.3. Biochemical parameters of CL dry extracts and oil.

Table 3 shows the values of the biochemical parameters of the dry extracts. The Brix value (23.20 ± 0.26 min), shows that *Curcuma longa* extracts, in particular EHCL, contain several soluble dry materials such as reducing sugars, acids, salts, pectins and nitrogenous materials. This wealth of bioactive compounds, such as curcuminoids, is associated with anti-inflammatory and antioxidant properties, enhancing therapeutic efficacy. In addition, the presence of reducing sugars and acids can improve the bioavailability of active ingredients while contributing to formulation stability, which is crucial for the preservation of pharmaceutical products [25]. The pH value of EHCL ($\text{pH} = 6.68 \pm 0.01$) obtained, indicates that *Curcuma longa* is closer to neutrality; this pH could influence the stability and efficacy of the bioactive compounds present in *Curcuma longa* extracts. Indeed, at this pH, certain phenolic compounds and flavonoids could be more stable, which would enhance the antioxidant properties of the extract [26].

The values for the biochemical parameters of *Curcuma longa* oil are also given in **table 3**. The iodine index measures the degree of fatty acid unsaturation in a given oil. The I_i value obtained in this study (84.1 ± 0.1 g I₂ /100g), is in the same range as values obtained with olive, peanut and castor oils, varying between 75 and 94 g I₂ /100 g oil [27]. These results are in line with those reported by [28] (75 to 106 g I₂ /100 g). Turmeric oil is thought to be rich in unsaturated fatty acids due to the presence of double bonds in its chemical composition. *Curcuma longa* is said to contain a significant proportion of lipids, including unsaturated fatty acids such as linoleic acid (omega-6) and oleic acid (omega-9). These unsaturated fatty acids give the oil beneficial health properties, such as anti-inflammatory effects and protection for the cardiovascular system [29]. The presence of unsaturated fatty acids in turmeric oil also confers neuroprotective properties, antibacterial and antifungal activities [30-32]. Consumption of *Curcuma longa* oil may slow tumour growth, by inducing apoptotic processes in tumour cells or inhibiting angiogenesis [33,34].

The refractive index is considered a criterion of oil purity. It varies with the wavelength of incident light, as well as with the temperature of analysis. This index is proportional to the molecular weight of the fatty acids, as well as to their degree of unsaturation. The refractive index value obtained in this study (1.52 ± 0.61) is higher than that indicated by the Codex Alimentarius standard, which varies between 1.46 and 1.47 for vegetable oils. This high value is linked to the presence of unsaturated fatty acids in the chain [35].

Table 3: Biochemical parameters of *Curcuma longa* dry extracts and oil

Biochemical parameters		Values
Dry extracts from CL	pH	6.68 ± 0.01
	°Brix (min)	23.20 ± 0.26
CL oil	Acid value	1.52 ± 0.61
	Iodine index (g I ₂ /100 g fat)	98.22 ± 0.84

3. Conclusion

This study revealed a diversity of bioactive substances consisting mainly of polyphenols, alkaloids, catechic tannins, leucoanthocyanins and flavonoids characteristic of *Curcuma longa*. Total polyphenols, flavonoids and tannins are more abundant in the hydroethanol extract than in the aqueous extract. This plant is also characterized by a number of biochemical parameters, including pH, which favors the stability of phenolic compounds and flavonoids, thereby enhancing the antioxidant properties of the extracts. As for turmeric oil, it is characterized by the presence of unsaturated fatty acids, which are thought to promote strong cardiovascular activity. *Curcuma longa* is proving to be a valuable phytotherapeutic resource, whose valorization could contribute to the development of modern medicine.

COMPETING INTERESTS

Author has declared that no competing interests exist.

Disclaimer (Artificial intelligence)

Option 1:

Authors hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

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