

## Bioactive compounds and antioxidant potential of aqueous and hydroethanolic extracts of *Cola lateritia* fruit parts

### Abstract :

**Generalities:** *Cola lateritia* is an under valorized (neglected and unutilized) plant traditionally used in certain Cameroonian villages as medicine to manage certain diseases. The present work aimed to assess the phytochemical composition and the antioxidant potential of *C. lateritia* fruits.

**Methods:** The fresh fruits of *C. lateritia* were purchased at *Mfoundi-Market* (Yaoundé 5, Centre Region of Cameroon), washed, sliced, dried, and powdered before being extracted with distilled water (AE) and ethanol/distilled water (70:30, v/v) (HEE)]. The bioactive compounds (polyphenols, saponins, tannins, flavonoids, and alkaloids) of the different extract of fruit were quantified using standard methods. The Antioxidant potential of the different extracts was assessed using various methods [scavenging of DPPH, phosphomolybdate method (TAC), and iron-reducing power (FRAP)].

**Results:** The results indicated that the different extracts of *C. lateritia* fruit contain phytochemicals (TPC, flavonoids, tannins, saponins and alkaloids). The AE of the skin (AE-sCLf) presented the highest contents (590.33  $\mu\text{g CaE/g DM}$ , 191.73  $\mu\text{g QE/g DM}$  and 945.33  $\mu\text{g SaE/g DM}$ ) for polyphenols, flavonoids and saponins respectively while, the AE of pulp (AE-PCLf) showed the highest content in alkaloids (121.59  $\mu\text{g QiE/g DM}$ ). Lowest tannin content was also reported in AE of pulp (2.02  $\mu\text{g CaE/g DM}$ ). The extracts of *C. lateritia* exhibited a good radical scavenging activity of DPPH (with an inhibition percentage of 74.084 and 69.40% respectively for HE-E-SCLf and AE-sCLf at 4 mg/mL). It's reducing power towards molybdate and iron was concentration-dependent with values ranging from 0.116 to 2.393  $\mu\text{g AAE/g DM}$  for TAC and from 0.0015 to 0.0032  $\mu\text{g AAE/g DM}$  for FRAP respectively. The highest iron-reducing activity of 941.410  $\mu\text{g AAE/g DM}$  was observed with AE-sCLf at 4 mg/mL. The PCA analysis showed a strong positive correlation between Bioactive and antioxidant activities.

**Conclusion:** The *C. lateritia* fruit extracts (AE and HEE) contain various bioactive compounds, which exhibited good antioxidant activities through different mechanisms. It suggests *C. lateritia* as a potential nutritious food as well as functional food useful for the prevention of management of cardiometabolic diseases.

**Keywords:** *Cola lateritia*, antioxidant potential, bioactive compounds, aqueous extract, hydro-ethanolic extract.

### Introduction

Most studies showed the evidence that specific foods provide health benefits to human (treatment and/or diseases prevention) (Kouogueu *et al.*, 2021; Aluko, 2012). This could explain why, people have achieved a good life quality by eating fruits and vegetables, other foods derived from plants, and using nutraceuticals, dietary supplements or phytotherapy. During the recent decades, high attention has been laid on fruit consumption because of their nutritional and health benefits (Aluko, 2012). In fact, most studies reported that, daily intake of fruits keeps us healthy. Fruits contain secondary metabolites (bioactive compounds) which are not directly involved in organism development, but contributes to the defense system against biotic and abiotic stresses (Maury *et al.*, 2020). All the bioactive compounds

(phenolic compounds, vitamins, terpenes and organic acids) present in the fruits are universally promoted as healthy (very important to the nutritional status) and beneficial to human. Fruits have sufficient amount of nutrients especially vitamins and minerals. They are also sources of phytochemicals that function as antioxidants and anti-inflammatory agents through diverse protective mechanisms (Liu, 2013). Fruits include diverse groups of plant foods such as *cola lateritia*, which are neglected and underutilized. The changes in lifestyle and demographics, consumer incomes, and shifting preferences due to advanced knowledge has become essential for the human nutrition. *Cola lateritia* is used as an alternative for dietary intake, however its potential is still not fully exploited. The reason is that, underutilized and alternative fruits appear as a good opportunity for local growers to gain access to small and big markets where consumers appreciate the presence of nutrients and bioactive compounds able to prevent most metabolic diseases (Donno and Turrini, 2020). The research of new sources of phytochemicals is an important opportunity to promote the prevention of certain diseases. The phytochemicals obtained from this fruit could have a beneficial application in rising the health of our populations. Many strategies such as the metabolism of bioactive compounds (including their bioaccessibility and bioavailability), the sensory and nutritional traits of the food with biological active molecules, should be investigated. Finally, it's very important, to study the toxicity of this fruit in its natural state.

*Cola lateritia* is a tree of the Malvaceae family. It is present from Guinea in West Africa to the South-East of the Democratic Republic of Congo (Brink, 2008). It has also been introduced in other countries such as Cape Verde, where it has become naturalized. Also known as (small) *Ouara* in French, *Amoreira* in Portuguese or *Efok ahié* in Cameroon (Brink, 2008), it is used locally in carpentry, medicine and food. There are two varieties of *Cola lateritia*, distributed from southern Nigeria to Gabon and var. *maclaudii* present from Guinea to south-west Nigeria (Ekalu and Habila, 2020; Brink, 2008; Burkill, 1985).

Various parts of the *Cola lateritia* plant (inner bark, leaves, etc.) can be prepared in the form of ointments or decoctions, for their therapeutic virtues (treatment of tuberculosis, skin rashes or coughs). They are also administered to fight against lung diseases, to soothe intercostal or menstrual pain, or as an anti-abortion during pregnancy. In Ivory Coast bark decoctions are used as a vaginal douche against infertility. In Cameroon as in Ivory Coast, the pulp of *Cola lateritia* is edible and is consumed locally. The seed, on the other hand, is chewed in the same way as the nuts of *Cola acuminata* and *Cola nitida*. In Côte d'Ivoire, the buds and young leaves are eaten as vegetables and are particularly appreciated during festivals and initiation ceremonies (Brink, 2008; Burkill, 1985). The study of the effect of different solvent extraction on bioactive compounds and antioxidant potential of *C. lateritia* fruit parts aimed to provide interest in finding innovative sources for natural nutraceuticals, antioxidants and contribute to generate new needs in the food supply.

## 2. Materials and methods

### 2.1. Chemical reagents

The chemical reagents used in the study included: Follin-Ciocalteu [(HO)<sub>3</sub>C<sub>6</sub>H<sub>2</sub>CO<sub>2</sub>, H<sub>2</sub>O; Merck], Sodium Carbonate (Na<sub>2</sub>CO<sub>3</sub>; Merck), Gallic Acid (C<sub>7</sub>H<sub>6</sub>O<sub>5</sub>; Merck), Aluminum Chloride (AlCl<sub>3</sub>; Merck), Potassium Acetate (CH<sub>3</sub>COOK ; Merck), Quercetin (C<sub>15</sub>H<sub>10</sub>O<sub>7</sub>, 2H<sub>2</sub>O; Merck), Ethanol 99%, Vanillin (C<sub>8</sub>H<sub>8</sub>O<sub>3</sub>; Merck), Chloridric acid (HCl, 12 N), Iron chloride (FeCl<sub>3</sub>; Merck), Ascorbic acid (C<sub>6</sub>H<sub>8</sub>O<sub>6</sub>; Merck), Sulfuric acid (H<sub>2</sub>SO<sub>4</sub> ; 98%), Sodium dihydrogen phosphate (NaH<sub>2</sub>PO<sub>4</sub>; Merck), Dibasic hidrogen phosphate (Na<sub>2</sub>HPO<sub>4</sub>; Merck), Ammonium molybdate (H<sub>24</sub>Mo<sub>7</sub>N<sub>6</sub>O<sub>24</sub>; Merck), Potassium ferricyanide [K<sub>3</sub>Fe(CN)<sub>6</sub>; Merck], Trichloroacetate (CCl<sub>3</sub>CO<sub>2</sub>Na ; Merck), Nitroprusiate (C<sub>5</sub>FeN<sub>6</sub>Na<sub>2</sub>O, 2H<sub>2</sub>O; Merck),

Sulfanilic acid [4-(H<sub>2</sub>N)C<sub>6</sub>H<sub>4</sub>SO<sub>3</sub>H; *Merck*], Acetic acid (CH<sub>3</sub>COOH; *Merck*), N(1-naphthylene) Ethylenediamine Dihydrochloride (NED) [C<sub>10</sub>H<sub>7</sub>NH(CH<sub>2</sub>)<sub>2</sub>NH<sub>2</sub>, 2HCl; *Merck*].

## 2.2. Collection, Identification and Preparation of Plant Material

The fruits of *cola lateritia* were purchased at *Mfoundi-Market* (Yaoundé 5, Centre Region, Cameroon). The plant material was identified by comparison study by the taxonomists at the National Herbarium of Cameroon (voucher number 41643/HNC). Upon arrival to the laboratory, the fruit samples were washed and rinsed. The pulp, seed and pericarp were manually removed, sliced and dried at 50 °C in a ventilated oven (*Heraeus*, Germany) during a week until a constant weight was obtained, before being powdered (1 µm size) and stored at room temperature (25 ± 2 °C) in glass bottles protected from light and humidity.

## 2.3. Preparation of the aqueous extract of *cola lateritia* fruit

Fifty (50) g of powders of each part of *C. lateritia* fruit (pulp, seed and pericarp) were dissolved in 500 mL of distilled water at a ratio of 1:10 (m/v). The mixture was homogenized and left at room temperature (25 ± 2 °C) protected from light for 12h. The mixture was then filtered through *Wattman* paper (N° 2). The filtrate obtained was dehydrated in a ventilated oven (*Heraeus*, Germany) at a temperature of 50 °C for 72h and the different aqueous extracts (AE) obtained were kept at room temperature (25 ± 2 °C) in small dark bottles.

## 2.4. Preparation of the hydro-ethanolic extract of *cola lateritia* seed

Fifty (50) g of the powdered kernel of *C. lateritia* fruit was dissolved in 500 mL of a Water-Ethanol (80%) solvent system (1:1, v/v) for a ratio of 1:10 (m/v). The resulting mixture was homogenized and left at room temperature (25 ± 2 °C) protected from light for 12h. The mixture was then filtered through *Wattman* paper (N° 2). The filtrate obtained was dehydrated in a ventilated oven (*Heraeus*, Germany) at a temperature of 50 °C for 72h and the different hydro-ethanolic extracts (HEE) obtained were kept in small dark bottles to be later used for the different analysis.

## 2.5. Estimation of the contents in bioactive compounds of the different extracts of *C. lateritia* fruits

### 2.5.1. Estimation of total polyphenols content

Total polyphenols were estimated by the method described by Singleton and Rossi in 1965 using the Folin-ciocalteu reagent. Approximately, 0.1 mL of each extract (4 mg/mL) was mixed with 0.75 mL of Folin-ciocalteu reagent (10-fold dilution). The entire mixture was incubated at room temperature (25 ± 2 °C). Five (5) min later, 0.75 mL of a sodium carbonate solution (Na<sub>2</sub>CO<sub>3</sub>, 6%) was added. The mixture was homogenized and incubated for 90 min at room temperature (25 ± 2 °C; in the dark), and then read the absorbance at 725 nm against a reagent blank. Gallic acid (0-1000 µg/mL) was used as a reference and the total polyphenols content was expressed as microgram of catechin equivalent per gram of dry matter (µg CaE/g DM).

### 2.5.2. Estimation of total flavonoid content

The colorimetric method described by Aiyegoro and Okoh in 2010 using Aluminium chloride was used to estimate total flavonoids content. Approximately, 0.5 mL of each extract (4 mg/mL) was added to 1.5 mL of methanol. Subsequently 0.1 mL of aluminium chloride (AlCl<sub>3</sub>, 10%), 0.1 mL of potassium acetate (CH<sub>3</sub>COOK, 1M), and 2.8 mL of distilled water were added. The mixture was well homogenized and incubated for 30 min at room temperature (25 ± 2 °C) and the absorbance was read at 415 nm wavelength against the reagent blank. Quercetin (0-1000 µg/mL) was used as a reference and the total flavonoid

content was expressed as microgram quercetin equivalent per gram dry matter ( $\mu\text{g QE/g DM}$ ).

### **2.5.3. Estimation of total tannins content**

The method of Bainbridge *et al.* (1996) was used to determine the total tannins content of the different extracts. Approximately, 1 mL of each extract (4 mg/mL) was mixed with 5 mL of working solution [50 g vanillin + 4 mL HCl (1N) in 100 mL distilled water], and the mixture was incubated at 30 °C for 20 min. The absorbance was read at the wavelength of 500 nm against the blank (without extract). Tannic acid (0-1000  $\mu\text{g/mL}$ ) was used to establish the calibration range and tannin content. Results were expressed as microgram tannic acid equivalent per gram of dry matter ( $\mu\text{g TAE/g DM}$ ).

### **2.5.4. Estimation of total alkaloids content**

Quantification of total alkaloids in the different extracts of *C. lateritia* fruit was performed by the method described by Singh *et al.* (2004), with some modifications. Approximately, 100 mg of each extract was subjected to extraction in 10 mL of ethanol solution (80%). The whole was well homogenized and centrifuged at a speed of 5000 rpm for 10 min. After centrifugation, 1 mL of the supernatant of each extract was taken and introduced into a test tube, followed by the respective addition of 1 mL of acidified  $\text{FeCl}_3$  (0.025 M) solution (0.5 M HCl) and 1 mL of an ethanolic solution of 1,10-phenanthroline (0.05 M). The whole mixture was again incubated at 100 °C in a water bath for 30 min. The absorbance of the reddish complex formed was read at the wavelength of 510 nm against the blank. Quinine at the concentration of 25  $\mu\text{g/mL}$  was used as the primary standard and the alkaloid content was expressed as  $\mu\text{g}$  quinine equivalent per gram of dry matter ( $\mu\text{g QiE/g DM}$ ).

### **2.5.5. Estimation of total saponins content**

The determination of total saponins in the extracts was performed by the method described by Hiai *et al.* (1976). Two hundred (200)  $\mu\text{L}$  of each extract was introduced into a test tube and then, 200  $\mu\text{L}$  of alcoholic vanillin solution (prepared in 80% ethanol) and 2000  $\mu\text{L}$  of sulphuric acid solution (72%) were added to it. The mixture was homogenized and placed in a water bath at 60 °C for 10 minutes. The absorbance of this prepared solution was read after incubation at wavelength of 535 nm against the blank. A saponins standard was used at different concentrations (0-1000  $\mu\text{g/mL}$ ) to establish the calibration range. Results were expressed as micrograms of saponins equivalent per gram of sample dry matter ( $\mu\text{g SaE/g DM}$ ). Three replicates were performed for each extract.

## **2.6. Evaluation of the antioxidant potential of the different extracts of *C. lateritia* fruits**

### **2.6.1. Scavenging capacity of DPPH (2,2-diphenyl-1-picrylhydrazyl) radical**

The DPPH radical scavenging capacity of each extract was measured by the method described by Sanchez-Moreno *et al.* (1998), with some slight modifications. Fifty (50)  $\mu\text{L}$  of methanolic solution of each extract at different concentrations (1-4 mg/mL) was added to 1.95 mL of DPPH methanolic solution (25  $\mu\text{g/L}$ ). At the same time, a negative control was prepared (50  $\mu\text{L}$  methanol + 1.95 mL methanolic solution of DPPH). The absorbance was read against a reagent blank at 515 nm wavelength after 30 min of incubation in a dark at room temperature ( $25 \pm 2$  °C). Ascorbic acid was used as a reference and was treated as the samples. The assays were performed in triplicate. Results were expressed as percent inhibition (I%). The formula of Yen and Duh of 1994 was used to calculate the percentage of inhibition of the DPPH radical according to the following formula:

$$(\% I) = \left( \frac{[Abs\ control - Abs\ test]}{Abs\ control} \right) \times 100$$

The values of trapping capacity 50 (TC<sub>50</sub>) or Inhibitory concentration 50 (IC<sub>50</sub>) were determined graphically by linear regression (logarithmic curve).

### 2.6.2. Evaluation of total antioxidant capacity (TAC)

The phosphomolybdenum method described by Prieto *et al.* (1999) was used to evaluate the total antioxidant capacity (TAC) of the different extracts from the *C.lateritia* fruit. 0.2 mL of each extract of varying concentrations (1-4 mg/mL) was mixed with 2 mL of working reagent solution (0.6 M H<sub>2</sub>SO<sub>4</sub> 98%, 28 mM NaH<sub>2</sub>PO<sub>4</sub> and 4 mM ammonium molybdate). The tubes were screwed down, and incubated at 95 °C for 90 min. After cooling, the absorbance of the solutions was measured at the wavelength of 765 nm against the blank reagent (2 mL of the reagent solution + 0.2 mL of distilled water) treated under the same conditions as the samples. Ascorbic acid (100 - 100 µg/mL) was used as the standard. The TAC was expressed as µg ascorbic acid equivalent per gram dry matter (µg AAE/ g DM).

### 2.6.3. Assessment of Ferric Reducing Antioxidant Power (FRAP)

The iron (Fe<sup>3+</sup>) reducing power of the different extracts was determined according to the method described by Oyaizu in 1986. One (1) mL of each extract at different concentrations (1-4 mg/mL) was mixed with 2.5 mL of 0.2 M phosphate buffer solution (pH 6.6) and 2.5 mL of 1% potassium ferricyanide K<sub>3</sub>Fe (CN)<sub>6</sub> solution. The mixture was incubated in a water bath at 50 °C for 20 min, then 2.5 mL of 10% trichloro-acetate (TCA) solution was added to the mixture to stop the reaction and the tubes were centrifuged at 3000 rpm for 10 min. An aliquot (2.5 mL) of supernatant was combined with 2.5 mL of distilled water and 0.5 mL of 0.1% aqueous iron chloride (FeCl<sub>3</sub>) solution. The absorbance of the reaction medium was read at 700 nm wavelength against a similarly prepared blank, replacing the extract with distilled water which allows calibration of the apparatus (UV-VIS spectrophotometer). Ascorbic acid (100 - 100 µg/mL) was used as a reference and treated under the same conditions as the samples. An increase in absorbance corresponds to an increase in the reducing power of the extract of interest tested. The reducing capacity of iron (Fe<sup>3+</sup>) was expressed as µg ascorbic acid equivalents per gram of dry matter (µg AAE/g DM).

## 2.7. Statistical analyses

*In vitro* experiments were performed in triplicate. The data were analyzed using a Statistical Package for Social Science (SPSS) software version 20.0 for Windows (IBM Corporation, USA). Results were expressed as mean ± standard deviation (SD). The data for multiple variable comparisons were performed by an Analysis of Variance (ANOVA, one factor) test, followed by a Least significant difference (LSD) test of Turkey, to compare the averages between the different samples. Pearson correlation tests and Principal Component Analysis (PCA) for continuous quantitative variables were performed in triplicate to assess the relationship between bioactive compound levels and observed activities. The results were considered as statistical significant for a P < 0.05. The Excel spreadsheet was used for data processing and plotting of graphs.

## 2.8. Results

### 2.8.1. Estimation of bioactive compounds

The results of this study revealed that AE-SCLf, HEE-SCLf, AE-PCLf and AE-sCLf contained phenolic compounds, flavonoids, tannins, saponins and alkaloids (Table 2). Their concentrations varied from one extract to another. The seed AE-SCLf showed the lowest content in polyphenols, flavonoids, alkaloid and saponins which were ( $190.33 \pm 2.89 \mu\text{g CaE/g DM}$ ,  $44.23 \pm 9.16 \mu\text{g QE/g DM}$ ,  $68.92 \pm 0.90 \mu\text{g QiE/g DM}$  and  $110 \pm 13.74 \mu\text{g SaE/g DM}$  respectively). In contrast, AE-sCLf of the skin presented the highest contents ( $590.33 \pm 12.70 \mu\text{g CaE/g DM}$ ,  $191.73 \pm 5.81 \mu\text{g QE}$  and  $945.33 \pm 47.08 \mu\text{g SaE/g DM}$ ) for polyphenols, flavonoids and saponins respectively. The pulp AE-PCLf showed the highest content in alkaloids ( $121.59 \pm 3.01 \mu\text{g QiE/g DM}$ ). For tannins, lowest content was also observed with the pulp AE ( $2.02 \pm 0.74 \mu\text{g CaE/g DM}$ ). In general, concerning the entire bioactive compound (polyphenols, flavonoids, alkaloids, tannins and saponins) assessed, AE-sCLf and HEE-SCLf exhibited approximately the same amount of these compounds with a slightly highest content in AE of the skin (AE-sCLf). Except for the AE-SCLf, the levels of alkaloids and saponins was approximately similar in the rest of extracts. All the extracts exhibited the same amount of tannins content. However, the AE-sCLf showed the best number of bioactive compounds followed by HEE-SCLf, AE-PCLf and AE-SCLf respectively. The pulp AE-PCLf showed the highest content in alkaloids.

**Table 1:** Contents of Bioactive compounds in the different extracts of *C. lateritia*

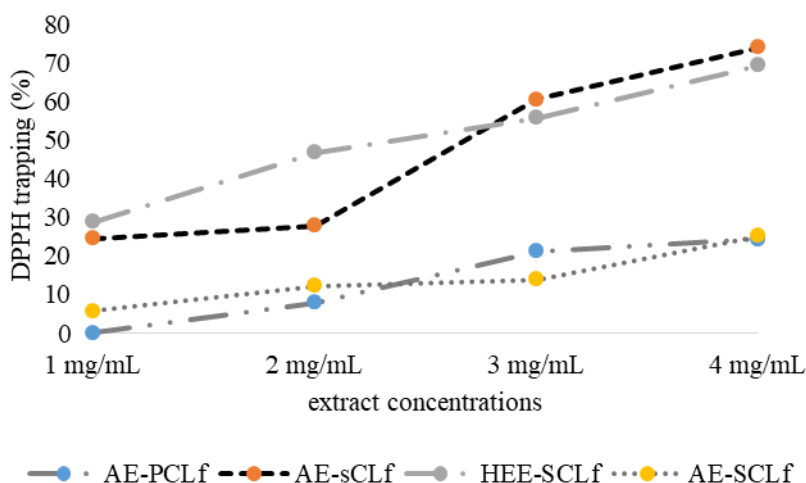
Bioactifs compounds	Polyphenols ( $\mu\text{g CaE/g DM}$ )	Flavonoids ( $\mu\text{g QE/g DM}$ )	Alkaloids ( $\mu\text{g QiE/g DM}$ )	Tannins ( $\mu\text{g TAE/g DM}$ )	Saponins ( $\mu\text{g SaE/g DM}$ )
AE-PCLf	$294.62 \pm 10.75^a$	$83.71 \pm 9.35^a$	$121.59 \pm 3.01^a$	$2.02 \pm 0.74^a$	$756.17 \pm 11.93^a$
AE-sCLf	$590.33 \pm 12.70^b$	$191.73 \pm 5.81^b$	$113.51 \pm 5.93^a$	$3.32 \pm 1.26^b$	$945.33 \pm 47.08^b$
HEE-SCLf	$427.95 \pm 12.96^c$	$135.79 \pm 3.34^c$	$110.55 \pm 0.93^a$	$4.32 \pm 0.88^b$	$830.5 \pm 10.26^c$
AE-SCLf	$190.33 \pm 2.89^d$	$44.3 \pm 9.16^d$	$68.92 \pm 0.90^b$	$3.03 \pm 0.76^b$	$110 \pm 13.74^d$

Values expressed as mean  $\pm$  standard deviation of triplicate trails; **CaE:** Catechin Equivalent; **TAE:** Tannic Acid Equivalent g; **DM:** Dry Matter; **QE:** Quercetin Equivalent; **QiE:** Quinine Equivalent; **EA:** Aqueous Extract; **HEE:** Hydro-Ethanollic Extract; **PCLf:** Pulp of cola lateritia fruit; **sCLf:** Skin of cola lateritia fruit; **SCLf:** Seed of cola lateritia fruit

## 2.8.2 Antioxidant potential of *cola lateritia* extracts

### 2.8.2.1. Scavenging capacity of the DPPH radical (2, 2-diphenylpicrylhydrazyl)

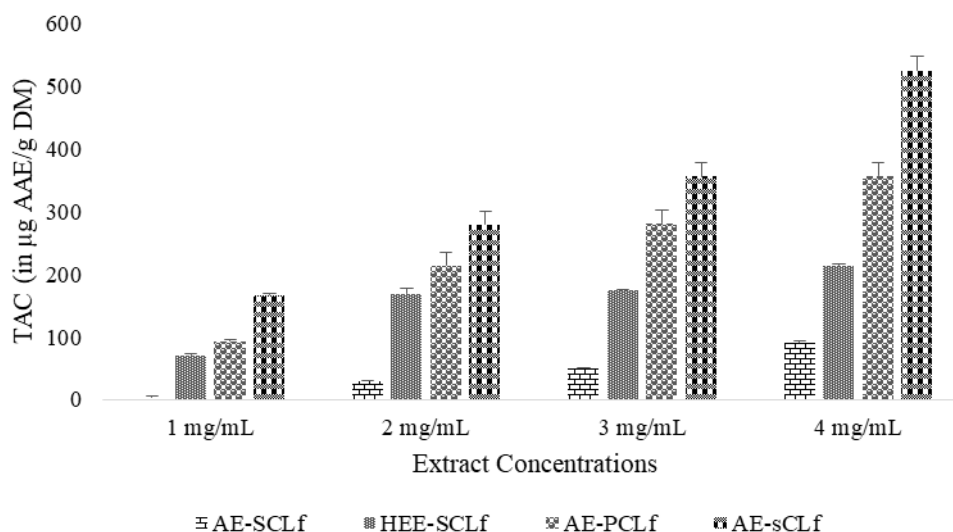
Figure 1 below represents the data of the DPPH radical scavenging capacity by the different extracts of *C. lateritia* fruit at different concentrations. For all the extracts, the activity was proportional to the tested concentrations. The maximum inhibition percentage of DPPH radical (74.084%, 69.40%) were observed with the HEE-SCLf and AE-sCLf respectively, at 4 mg/mL concentration. The AE-PCLf and AE-SCLf showed the lowest activity with just 25.23% inhibition at that concentration of 4 mg/mL.



**Figure 1:** DPPH radical scavenging capacity of different extracts of *C. lateritia* fruit

### 2.8.2.2. Total Antioxidant Capacity

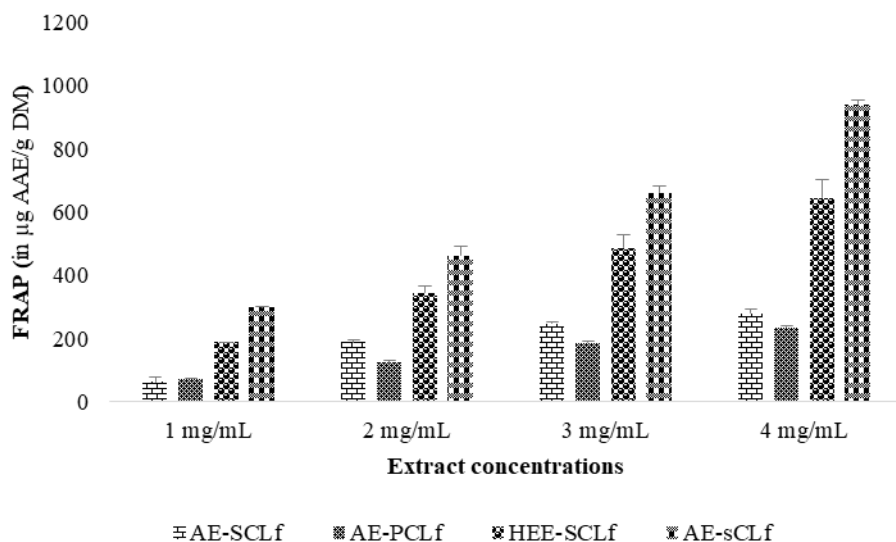
The results shown in Figure 2 are those of the total antioxidant capacity (TAC) of the different extracts of *C. lateritia* at different concentrations. The different extracts tended to reduce Molybdate (VI) to Molybdate (V) in a concentration dependent manner. At 4 mg/mL, the AE-sCLf extract presented the highest total antioxidant capacity (524.488  $\mu\text{g AAE/g DM}$ ), followed by the AE-PCLf (357.6  $\mu\text{g AAE/g DM}$ ), the HEE-SCLf (214.1  $\mu\text{g AAE/g DM}$ ) and least was the AE-SCLf (90.6  $\mu\text{g AAE/g DM}$ ).



**Figure 2:** Total antioxidant capacity (TAC, molybdate reduction) of different extracts of *C. lateritia* fruit

### 2.8.2.3. Iron reduction capacity of the different extracts from *C. lateritia* fruit

The data recorded in Figure 3 depict the ability of the different extracts of *C. lateritia* at different concentrations, to reduce ferric iron. The AE-SCLf and AE-PCLf presented the weakest activity of 280.512  $\mu\text{g AAE/g DM}$  and 234.230  $\mu\text{g AAE/g DM}$  respectively which increased with the increase of the tested concentration. This increase in activity dependent concentration was observed for all the extracts. The highest iron-reducing activity of 941.410  $\mu\text{g AAE/g DM}$  was observed with AE-sCLf at the extract concentration of 4 mg/mL.



**Figure 3:** Ferric iron reducing capacity of different extracts of *C. lateritia* fruit

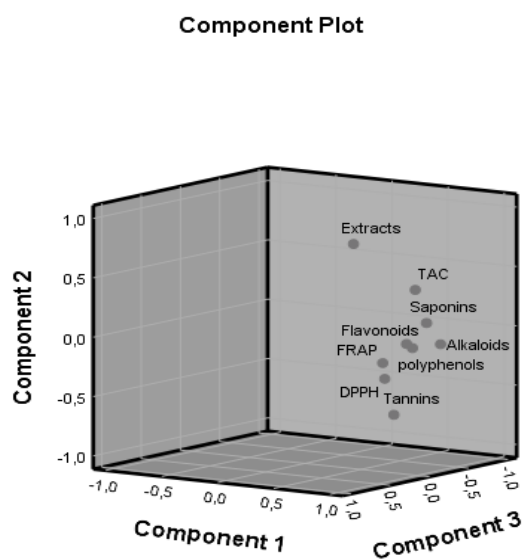
### 2.9. Correlations between bioactive compounds and the antioxidant activities

The antioxidant activities of all tested extracts of *C. lateritia*, measured by different mechanisms of action (DPPH, FRAP and TAC), were found to be correlated with bioactive compounds (polyphenols, flavonoids, tannins and alkaloids).

**Table 2:** correlation matrix of bioactive compounds and antioxidant potential in the different extracts of *C. lateritia*

Correlation Matrix									
Correlation	Extract	polyphenols	Flavonoids	Alkaloids	Tannins	Saponins	TAC	FRAP	DPPH
polyphenols	0,508	1,000	0,995	0,829	0,766	0,911	0,861	0,950	0,896
Flavonoids	0,551	0,995	1,000	0,784	0,727	0,912	0,860	0,959	0,908
Alkaloids	0,314	0,829	0,784	1,000	0,739	0,888	0,789	0,640	0,657
Tannins	-0,094	0,766	0,727	0,739	1,000	0,619	0,398	0,763	0,817
Saponins	0,588	0,911	0,912	0,888	0,619	1,000	0,894	0,770	0,791
TAC	0,771	0,861	0,860	0,789	0,398	0,894	1,000	0,725	0,626
FRAP	0,440	0,950	0,959	0,640	0,763	0,770	0,725	1,000	0,927
DPPH	0,302	0,896	0,908	0,657	0,817	0,791	0,626	0,927	1,000

In order to assess and visualize the existing relationships between the various antioxidant activities and the bioactive compounds contents of different extract concentrations of the *C. lateritia* fruit, a *Pearson* correlation and principal component analysis (PCA) tests were performed and carried out (Figures 4). Globally, with the respective correlation coefficient of 0.86, 0.79 and 0.89 bioactive compounds such as TPC, flavonoids, alkaloids and saponins exhibited a strong positive correlation for total antioxidant capacity (TAC); TPC, flavonoids, tannins and saponins were also strongly linked to a FRAP activity of the extracts. With a correlation coefficient  $\geq 0.8$ , TPC, flavonoids and tannins were the compounds which were exhibited the best DPPH scavenging activity. The strong relationship between these bioactive compounds and antioxidant result is a clear indication that the bioactive compounds of the fruit extracts contributed to the antioxidant potential. These results suggest that all these phytochemical constituents are involved in antioxidant mechanisms.



**Figure 4:** Principal components analyses of the different extracts

### Discussion

It is now well accepted that the consumption of plant-based foods is beneficial to human health. *Cola lateritia* is one of the fruits commonly consumed by many Cameroonian populations (South-west, North-west, South, East and Centre Regions) which has essential nutrients required for human well-being. This work was aimed to investigate the effect of solvent extraction on bioactive compounds and antioxidant properties of different parts of *C. lateritia* fruit. The bioactive compounds of the different extracts of fruit parts were quantified and the results are presented in Table 1. The results of this study revealed that AE-SCLf, HEE-SCLf, AE-PCLf and AE-sCLf contain phenolic compounds, flavonoids, tannins, saponins and alkaloids. Their concentrations vary from one fruit extract part to another, and in terms of solvent extraction, except for the alkaloids content, the values of the rest of bioactive contents were approximately the same in all the different extracts. However, the AE-sCLf showed the best number of bioactive compounds followed by HEE-SCLf, AE-PCLf and AE-SCLf respectively. The AE-PCLf showed the highest content in alkaloids. Herb phytochemicals have been reported to be efficiently extracted in solvents with higher polarity, which makes water a superior solvent for the extraction of bioactive compounds. Therefore, the selectivity of suitable solvents and extraction methods is important due to the various composition of phytochemicals. Most studies have demonstrated that methanol and ethanol solvents can extract more bioactive compounds in plants (Edoun *et al.*, 2020; Mbong *et al.*, 2020).

The differences between extraction methods (extraction conditions and extraction concentrations) not only influence the bioactive compound contents of extracts, but also affect their antioxidant potential. The current study showed that all the extracts of *C. lateritia* exhibited good antioxidant properties through the targeting of free radical (DPPH, fig 1) and metal reduction [FRAP (fig 3) and TAC (fig 2)]. In our study, we have noticed that, for the scavenging of DPPH radical and FRAP, AE-sCLf and HEE-SCLf exhibited the best activities. Contrary, AE-sCLf and AE-PCLf possess the highest antioxidant activities for TAC. In general, AE-sCLf showed strong inhibition of DPPH radical and the best activity on metal reduction (FRAP and TAC) compared to all the rest of extracts. We also found a good correlation on the levels of bioactive contents (polyphenols, flavonoids, alkaloids and

tannins), the scavenging and reducing power antioxidant assays at  $R^2 = 0.95$ . These results corroborated the study of Edoun *et al.*, 2020 and Konczak *et al.*, in 2002, who found respective correlation coefficients of 0.94 and 0.8315 for native Cameroonian and Australian herbs and spices. Fruits are very good sources of antioxidants and have many different antioxidant components. In fact, each fruits contains various kind of antioxidant compounds and this can vary among species to species as well the climate. Phytochemicals present in this extract may have a significant effect on antioxidant activities. Many studies showed that, the antioxidant activity or potential could be directly related to the total amount of phenols and flavonoids found in extracts (Mariya *et al.*, 2016 and Parente *et al.*, 2013). The presence of phenols, flavonoids, alkaloids and tannins in different extracts of *C. lateritia*, is an indication that these secondary plants metabolic have a synergistic effect on the various therapeutic properties (Onyesom *et al.*, 2015). All bioactive components (polyphenols, flavonoids and alkaloids) are major antioxidants in natural products and their antioxidant potential have been demonstrated in previous studies. In addition, the antioxidant activity of polyphenols and flavonoids depends on the number of hydroxyl groups in the molecule and structure. A strong activity in the presence of low polyphenols and flavonoids contents may be explained by the fact that the other phytochemical compounds such as alkaloids and tannins, exhibited an antioxidant property (Edoun *et al.*, 2020; Mbong *et al.*, 2020).

### Conclusion

These results suggested that, the extract of the different part of *C. lateritia* fruit possess a good number of phytochemicals (polyphenols, flavonoids and alkaloids) which led to a high antioxidant property through the trapping of DPPH radical and metal reduction (FRAP and TAC). The correlation between antioxidant activities and bioactive compounds of the different extracts was high ( $R^2 = 0.95$ ). Further, studies need to be conducted to identify compounds that are clearly correlated with the antioxidant activities of extracts as well as their synergistic interactions. These additional results will provide new insight into the mechanisms that lead to the special bioactivities of *Cola lateritia* fruits.

### Abbreviations

**TCA:** TriChloroAcetate; **NED:** N- (1-naphthylene) -ethylenediamine dihydrochloride; **CaE:** Catechin Equivalent; **QE:** Quercetin Equivalent; **QiE:** Quinin Equivalent; **AE:** Aqueous extract; **HEE:** Hydro-ethanolic extract; **m/v:** mass per volume; **CRASAN:** Centre for Food, Food Security and Nutrition Research, **IMP:** Institute of Medical Research and Medicinal Plants Studies; **TAC:** Total Antioxidant Capacity; **FRAP:** Ferric Reducing Antioxidant Potential; **PCLf:** Pulp of *Cola lateritia* fruit ; **SCLf:** Seed of *Cola lateritia* fruit; **sCLf:** Skin of *Cola lateritia* fruit; **AOAC:** Association of Official Analytical Chemists.

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