Scholars Academic Journal of Pharmacy
Abbreviated Key Title: Sch Acad J Pharm
ISSN 2347-9531 (Print) | ISSN 2320-4206 (Online)
Journal homepage: http://saspublishers.com

OPEN ACCESS

Pharmacology

Phytochemical Study Assay of Phenolic Compounds and Determination of the Antioxidant Activity of Aqueous Extracts from the Leaves of three Medicinal Plants Used in the Haut-Sassandra Region, Daloa (Côte d'Ivoire)

DIBY Yao Bernard[1*], KAHOU BI Gohi Parfait[1], KOUAME Djè Kouamé Wenceslas[2], N'DIA Kouadio Frédéric[3], YAPO Angoué Paul[3]

[1]Jean Lorougnon GUEDE University, Training and Research Unit of Agroforestory, Laboratory of Agrovalorisation, PO Box 150 Daloa, Côte d'Ivoire
[2]Polytechnic University of Man, Training and Research Unit-Engineering, Agronomic, Forestry and Environmental (UFR-IAFE), BP 20 Man, Ivory Coast
[3]Nangui ABROGOUA University, Nature Sciences Training and Research Unit, Laboratory of Physiology, Pharmacology and Pharmacopoeia, PO Box 801 Abidjan 02, Côte d'Ivoire

DOI: https://doi.org/10.36347/sajp.2025.v14i02.004 | **Received:** 13.01.2025 | **Accepted:** 17.02.2025 | **Published:** 26.02.2025

***Corresponding author:** DIBY Yao Bernard
Jean Lorougnon GUEDE University, Training and Research Unit of Agroforestory, Laboratory of Agrovalorisation, PO Box 150 Daloa, Côte d'Ivoire

Abstract **Original Research Article**

Cassia occidentalis, *Jatropha curcas* and *Chromoleana odorata* are plants used in the treatment of pathologies in Côte d'Ivoire. With the aim of treating pathologies caused by oxidative stress using antioxidants, a study was carried out into the antioxidant activity of aqueous extracts of these plants and to identify the molecules responsible for this effect. After aqueous extraction, the extracts obtained were used to carry out phytochemical tests, phenolic compound assays and antioxidant activity tests using the DPPH• and ABTS+• methods. Phytochemical screening revealed the presence of sterols, polyphenols, flavonoids, alkaloids and saponins. Respective contents of total polyphenols are 3.40 ± 0.04, 3.15 ± 0.03 and 2.80 ± 0.02 mgEAG/g DM for *Cassia occidentalis*, *Jatropha curcas* and *Chromoleana odorata* and those of flavonoids are 54.15 ± 1.2, 50.40 ± 1.2 and 48.50 ± 1.1 mg EqQ/g DM for *Cassia occidentalis*, *Chromoleana odorata* and *Jatropha curcas*. Using the DPPH test, the mean IC50 values were 2.74 ± 0.1 µg/mL for *Cassia occidentalis* and *Chromolaena odorata* and 2.73 ± 0.1 µg/mL for *Jatropha curcas*. ABTS test revealed mean IC50 values of 10.35 ± 0.1 µg/mL for *Cassia occidentalis* and *Chromoaena odorata* and 7.01 ± 0.1 µg/mL. *Jatropha curcas*. These antioxidant powers are thought to be due to the effects of certain phenolic compounds present in these plants, confirming their natural antioxidant activity.

Keywords: *Cassia occidentalis*, *Jatropha curcas*, *Chromoleana odorata*, antioxidant activity, DPPH, ABTS.

INTRODUCTION

Medicinal plants have always been an important source of numerous active compounds for the treatment of many diseases. They are also used to effectively combat oxidative stress by inhibiting the production of free radicals (Adjanohoun & Aké-Assi, 1979). Oxidative stress refers to the imbalance between the production of free radicals and the quantity of antioxidants available and usable by the body. This imbalance is sometimes at the root of many human diseases, such as cancer, cardiovascular disease, diabetes and Alzheimer's disease (Gönenç *et al*., 2013; Inbathamizh *et al*., 2013). Diseases caused by oxidative stress are generally manifested by ageing, which leads to a reduction in antioxidant defences and encourages the proliferation of free radicals in cells (Moon & Shibamoto, 2009). In order to prevent these diseases caused by oxidative stress, new anti-oxidant molecules are needed to combat oxidative stress and its many consequences. Traditional medicine is therefore used because it is accessible, widely available, less expensive and socially and culturally acceptable. The use of synthetic antioxidant molecules is currently

being called into question because of the potential toxicological risks. In addition, those derived from medicinal or food plants are becoming increasingly important, in particular because of their beneficial effects on health (Xu *et al.*, 2017). This is the case with polyphenols, which are natural compounds widely found in plants, attracting a great deal of interest for their beneficial action against free radicals (Marc *et al*., 2005). With this in mind, the use of plants as natural antioxidants offers an alternative to oxidative stress and, consequently, the elimination of free radicals. Côte d'Ivoire has a rich diversity of flora and several medicinal plants that deserve to be explored in the search for antioxidant or therapeutic molecules. It has proved necessary to study the antioxidant activity of three plants from the Ivorian flora. Aim of this study is to evaluate the antioxidant activity of the total aqueous extract of the leaves of these plants and to establish a comparative study of the effect of each aqueous compound through phytochemical screening.

MATERIALS AND METHODS

Plant Material

Plant material consists of the leaves of three plants: *Jatropha curcas*, *Chromoleana odorata* and *Cassia occidentalis*. The selected plants were collected in their natural habitat in the Haut-Sassandra region (Côte d'Ivoire) during January 2024. After harvesting, they were dried in the shade (at room temperature), in order to avoid as far as possible the degradation of the molecules through the mechanisms of fermentation and degradation of the organic characteristics of our raw materials.

Extraction

Leaves of the three plants (*Cassia occidentalis* L., *Jatropha curcas* L. and *Chromolaena odorata* L.) were harvested, washed and then dried in the shade at room temperature (25-30°C) for three weeks at the Biochemistry Laboratory of Jean Lorougnon GUEDE University, Daloa (Côte d'Ivoire) They were then ground into powder using an electric grinder (Retschsk 100). Various powders obtained were stored in jars and then deposited in a dry place at an ambient temperature of 25°C in the said laboratory for later use. Extraction method used was that described by (Zirihi *et al*., 2003). One hundred grams (100g) of *Cassia occidentalis* (or *Jatropha curcas* or *Chromolaena odorata*) fine powder is macerated in one litre of distilled water under magnetic stirring for twenty-four hours at room temperature. Macerate obtained is filtered first on cotton wool and then on Wattman N°1 paper. Filtrate is then concentrated under reduced pressure at 60°C using an R-III-Buchi BUC-1103011V0 rotary evaporator (France). After drying in an oven at 45°C for 24 hours, the total aqueous extract of *Cassia occidentalis* (or *Jatropha curcas* or *Chromolaena odorata*) leaves is obtained and then stored in the freezer at -5°C. All the extracts obtained were weighed to assess the extraction yield according to the following formula:

$$R = Mf/Mi \times 100$$

R: extraction yield (%); **Mf**: final mass of total aqueous extract obtained (g); **Mi**: initial mass of fine leaf powder from each plant (g)

Phytochemical screening

Main groups of chemical compounds were sought in the aqueous extracts of the leaves of each plant using conventional methods of characterisation by colour reaction (Békro *et al*., 2007). Solutions showing positive reactions indicate the presence of the highlighted chemical groups in the extract. Summaries of the reactions are given in Table 1.

Table 1: Chemical group identification reaction

Chemical compounds	Reagents	Expected positive results
Alkaloids	Valser and Mayer (KI_3)	Creamy white colouring
	Dragendorff K[Bi]	Orange colouring
Polyphenols	$FeCl_3$	Blackish-blue colouring
Saponosides	Foam test	Persistent foam
Terpenes and sterols	Liebermann (EtOH + $(CH3CO)_2$ +H_2SO_4)	Purple to green colouring
Flanonoids	Cyanidine	Orange-pink colouring
Catechin	Stiasny (30% CH_2O + HCl concentré 2/1)	Formation of large flakes
Tannins Gallic	$FeCl_3$	Intense blue-black colouring

Total polyphenols content

Phenolic compounds are compounds with several hydroxyl groups. They are assayed using the Folin-Ciocalteu reagent according to the method described by (Singeton *et al*., 1999). A volume of 200 µL of each extract, suitably diluted in methanol, is placed in a test tube and 1 mL of Folin-Ciocalteu reagent is added. After 5 minutes incubation at room temperature, 800 µL sodium carbonate (7.5%) was added to the mixture. Resulting mixture is kept in a water bath in the dark for 30 minutes. Absorbance is measured at 765 nm against a blank using a spectrophotometer. Three repetitions were carried out. A calibration curve was run in parallel under the same operating conditions, using gallic acid as the standard at different concentrations. Concentration of total polyphenols is expressed in milligrams of gallic acid equivalent per gram of dry plant matter (mg GAE/g DVM) (Ben Moussa *et al*., 2022).

Flavonoids content

Flavonoids are yellow pigments, generally polyphenolic, and the method used to measure them is the cyanidin reaction. Total flavonoids are measured using the method (Zhishen *et al*., 1999). A 250 µL volume of each extract is diluted in methanol in a vial. To this mixture, 1 mL of distilled water is added, followed by 75 µL of NaNO2 (5%). 5 minutes later, 75 mL of a 10% $AlCl_3$ solution was added, followed after 6 minutes by the addition of 500 µL NaOH (1N) and 600 µL distilled water. Mixture is stirred immediately. Absorbance is measured at 415 nm against a blank using a spectrophotometer. Three repetitions were carried out. A calibration curve was run in parallel under the same operating conditions, using quercetin as the standard at different concentrations. Concentration of total flanonoids is expressed in milligrams of quercetin equivalent per gram of dry plant matter (mg QE/g DVM) (Ben Moussa *et al*., 2022).

Antioxidant activity

Scavenging test for the free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH)

Free radical scavenging activity was measured by the DPPH (2,2- diphenyl-1-picrylhydrazyl) method according to the work of (Roukia *et al*., 2015). Stock solution is prepared by dissolving 24 mg of DPPH in 100 mL of methanol. Solution obtained has an absorbance of approximately 0.98 ± 0.021 at 517 nm using the spectrophotometer. 1.68 mL of this solution was mixed with 1600 µL of the sample at various concentrations (3, 125 and 100 µg/mL). Reaction mixture was shaken well and incubated in the dark for 30 min at room temperature. Absorbance was then measured at 517 nm. Control was prepared as above, without any sample. Each test is repeated three times, and the results are presented as the average of the three tests. Percentage of DPPH radical trapping is calculated according to the following equation (Torres De Pinedo & Morales, 2007).

I % = [(control OD - sample OD) / control OD)] X 100

Control OD: Optical density of the control (methanoic solution of DPPH)

Sample OD: Optical density of the sample under test

IC50 or 50% inhibitory concentration is the concentration of the test sample required to reduce 50% of the DPPH● radical. IC50 were determined graphically by linear regression of the plotted graphs; percentages of inhibition as a function of the different concentrations of the fractions tested. Lower the IC50, the greater the antioxidant activity.

ABTS free radical scavenging test

Method used was that described by (Leong & Shui, 2002). A quantity of 38.40 mg of ABTS was dissolved in 10 mL of water before the addition of 6.75 mg of potassium persulphate. Resulting mixture was kept in the dark at room temperature for 12 h before use. It was then diluted with ethanol to give an absorbance of around 0.7 at 734 nm. Antioxidant activity was measured by adding 2 mL of an aqueous solution of the extract tested to 2 mL of the ABTS+• solution. Extracts were tested at the following concentrations: 2.5, 10, 100 and 200 µg/mL. Gallic acid, used as a reference antioxidant, was tested at the same concentration. Absorbance was read after 2 minutes on a spectrophotometer at 734 nm using ethanol as the blank. Three absorbance measurements were taken for each concentration tested (n=3). Expression of inhibition percentages and the calculation of 50% inhibitory concentrations (IC50) were carried out as described for DPPH

Statistical Analysis

Results were statistically analysed using the Tukey test combined with a one-factor ANOVA. This test was carried out using GraphPad Prism software version 5.0 (Microsoft, San Diego, California, USA). Values are expressed as averages followed by the standard error of the mean (m±esm). Difference between the means is considered statistically significant at the 5% level ($p<0.05$).

RESULTS AND DISCUSSION

Phytochemical screening

Phytochemical screening of the various aqueous plant extracts using specific reagents revealed the presence of the chemical compounds listed in Table 2.

Table 2: Chemical compounds present in the total aqueous extract of different plants

Metabolites	*Jatropha curcas*	*Chromolaena odorata*	*Cassia occidentalis*
polyphenols	++	++	++
Alkaloids	++	-	-
Gallic tannins	++	++	++
flavonoids	++	++	++
Saponosides	+++	+++	+++
terpenes and sterols	++	++	++

(+++): Abundant, (++): Less abundant, (-): Absent

Study of a plant's biological effects is generally assessed through its bioactive molecules. These molecules are obtained by extraction methods using appropriate solvents. In terms of phytochemical screening, this study revealed the presence of polyphenols, gallic tannins, flavonoids, saponosides, terpenes and sterols in all the aqueous extracts of *Cassia occidentalis*, *Jatropha curcas* and *Chromoleana odorata* leaves. In addition to these compounds, alkaloids are also present in *Jatropha curcas* extract, but absent in *Cassia occidentalis* and *Chromoleana odorata* extracts. Results of the triphytochemistry of aqueous extracts of

Chromoleana odorata and *Cassia occidentalis* leaves differ from those obtained in the work of (N'guessan *et al*., 2009 ; Kamsi *et al*., 2020) on the same extracts, as they noted the absence of gallic tannins and flavonoids but the presence of alkaloids. Moreover, the same is true of the chemical composition of *Jatropha curcas*, which differs from that of (Saraka *et al*., 2018), whose extract they observed to lack flavonoids and alkaloids. These results show that there is a difference in the chemical composition of these plants between these authors and our own.

Extraction yield

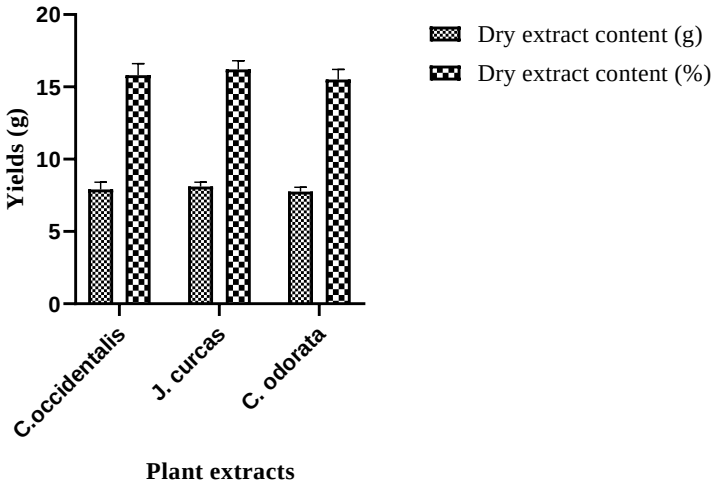

Figure 1: Dry extract yield of the various plants studied

Yield of extractions containing chemical compounds varies from one plant extract to another. Respective mean dry weights of *Cassia occidentalis*, *Jatropha curcas* and *Chromoleana odorata* extracts are roughly equal, at 7.90 ± 0.5 g, 8.10 ± 0.3 g and 7.75 ± 0.3 g. These values are then expressed as a percentage of 50 g of fine leaf powder from each plant to determine yield. Respective mean yield values for *Cassia occidentalis*, *Jatropha curcas* and *Chromoleana odorata* are 15.8 ± 0.8%, 16.2 ± 0.6% and 15.5 ± 0.7% (Figure 1). Each dry extract is characterized by its color and appearance. Dry extracts of *Cassia occidentalis* and *Chromoleana odorata* are black in color with a powdery appearance. *Jatropha curcas* dry extract, on the other hand, is brown and powdery. Yields obtained in this study are very similar, assuming that the water solvent extracts a good quantity of the molecules from the different plants. These results are in line with those obtained by (Ballo, 2013).

Total polyphenol content

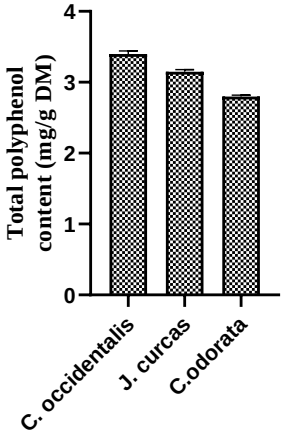

Figure 2: Total polyphenol content of aqueous plant extracts

Figure 2 shows the analysis of total polyphenol content in the leaves of *Cassia occidentalis*, *Jatropha curcas* and *Chromolaena odorata*. These levels were determined in an aqueous solvent (distilled water) using the Folin-Ciocalteu reagent. Estimates of total polyphenol content in each plant leaf extract showed the same trend, expressed as milligram equivalent of gallic acid per gram of dry matter (mg EAG/g DM). Average

total polyphenol values for the leaves of each plant are low, at 3.40 ± 0.04 mg EAG/g DM for *Cassia occidentalis*, 3.15 ± 0.03 mg EAG/g DM for *Jatropha curcas* and 2.80 ± 0.02 mg EAG/g DM for *Chromoleana odorata*. According to these results, the low content of total polyphenols in the aqueous extracts of said plants would be due to certain factors such as extreme climatic conditions such as high temperature, solar exposure, drought and also the presence of these compounds can vary during plant development from one locality to another (El Hazzat *et al*., 2015).

Flavonoid content

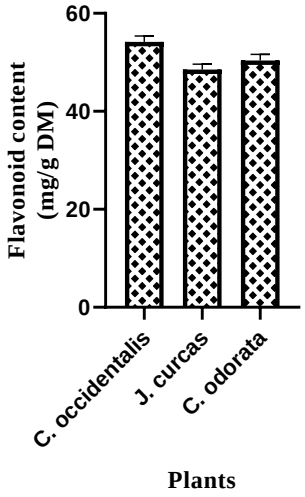

Figure 3: Flavonoid content of aqueous plant extracts

Results of flavonoid determination in aqueous extracts from the leaves of *Cassia occidentalis*, *Jatropha curcas* and *Chromolaena odorata* are shown in Figure 3. Average flavonoid contents vary from plant to plant, and are expressed in milligrams of quercetin equivalent per gram of dry matter (mg EqQ/g DM). Highest content is found in *Cassia occidentalis* with a value of 54.15 ±1.2 mg EqQ/g DM followed by *Chromolaena odorata*. with a value of 50.40 ±1.2 mg EqQ/g DM and *Jatropha curcas* with a value of 48.50 ±1.1 mg EqQ/g DM. According to the results, flavonoid content in these plants may depend either on flooding and salinity, which stimulate the biosynthesis of certain molecules such as flavonoids, or on intrinsic factors, including genetic factors and extrinsic factors such as climatic conditions, cultivation practices, maturity and harvesting time, and storage conditions (Falleh *et al*., 2008).

Evaluation of antioxidant activity by free radical scavenging 2,2-diphenyl-1-picrylhdrazyl (DPPH)

Antioxidant activity of aqueous extracts of *Cassia occidentalis*, *Jatropha curcas*, *Chromolaena odorata* and ascorbic acid was qualified spectrophotometrically and shown in Figure 4.

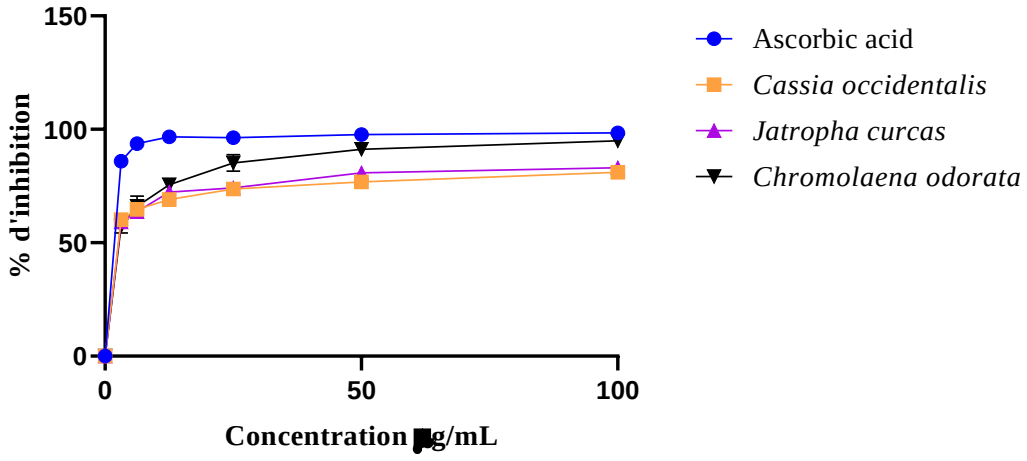

Figure 4: Percentage DPPH inhibition curves as a function of concentrations of different extracts of *Cassia occidentalis*, *Jatropha curcas*, *Chromolaena odorata* and ascorbic acid

Average inhibitory concentration (IC50) values are shown in Table 3.

Table 3: IC50 values for plant extracts and ascorbic acid obtained by DPPH test

Extracts/Standard	IC50 (µg/mL)
Cassia occidentalis	2.74 ± 0.08***
Jatropha curcas	2.73 ± 0.08***
Chromolaena odorata	2.74 ± 0.08***
Ascorbic acid	1.76 ± 0.08

*Each value represents the mean ± SD of three replicates. The comparison was made using a one-way analysis of variance (ANOVA 1) followed by the Turkey test; ***p<0.001 indicates a highly significant difference from the standard (control).*

Anti-free radical activity of aqueous plant extracts on the DPPH radical was estimated by spectrophotometer at 517 nm after reduction of this radical. Results show that the aqueous extracts of *Cassia occidentalis*, *Jatrpha curcas*, *Chromoleana odorata* and ascorbic acid (the reference molecule) have a fairly high antioxidant capacity. This power is confirmed by the high inhibition percentages (figure 4) and low inhibitory concentration 50% (IC50) values (table 3). Mean IC50 values for plant extracts were 2.74 ± 0.08 µg/mL for *Cassia occidentalis* and *Chromoleana odorata*, and 2.73 ± 0.08 µg/mL for *Jatrpha curcas*. These values are statistically significantly different (p<0.001) from those of ascorbic acid. Lowest IC50 value for ascorbic acid is 1.76±0.08 µg/mL. DPPH method, which is based on an electron or hydrogen atom transfer reaction that completes the antioxidant description of the various macerates of our studied plants, was used in this study. It is therefore advisable to use more than one method to measure the antioxidant capacity of a substance, in order to take into account the different mechanisms contributing to antioxidant action. Results of the different methods are dose-dependent. Compared with the reference antioxidant (ascorbic acid) with an inhibitory concentration 50% (IC50) of 1.76 ± 0.08 µg/mL, those of aqueous extracts of *Cassia occidentalis* and *Chromolaena odorata* (2.74±0.08 µg/mL) and *Jatropha curcas* (2.73 ± 0.08 µg/mL). And taking into account the fact that antioxidant power is inversely proportional to IC50 value, ascorbic acid has greater antioxidant activity than the aqueous extracts of the plants tested. Nevertheless, it is essential to remember that the aqueous extracts of the three plants have good antioxidant activities, in view of the results obtained in the various DPPH tests. As a result, our extracts, with approximately the same values, admit more or less the same antioxidant activities. DPPH antioxidant activity of the aqueous extract of *Chromolaena odorata* leaves corroborates that of (Konan *et al*., 2022), who showed that *Chromolaena odorata* essential oil possesses antioxidant activity with an IC50 value of 17.4 µg/mL. This is higher than our own value of 2.74 ± 0.08 µg/mL. In addition, the same authors also showed a significant difference (p<0.001) between the antioxidant activity of the essential oil of the aforementioned plant and ascorbic acid (vitamin C), with an IC50 value of 0.31 µg/mL lower than ours, which is 1.76 ± 0.1µg/mL. In a study by (Huang *et al*, 2020) on *Jatropha curcas* seeds and hulls, where DPPH free radical scavenging techniques were used to determine their antioxidant activities. Aqueous extract of *Jatropha curcas* seeds showed good scavenging activity for this radical, with an IC50 value of 13.63 ± 0.15 µg/mL. This value is also higher than that of the aqueous extract of *Jatropha curcas* leaves, which has good antioxidant activity, justified by an IC50 of 2.73 ± 0.08 µg/mL. Same authors showed a significant difference (p<0.001) between the antioxidant activity of the plant's hulls and that of ascorbic acid, with an IC50 of 8.35 ± 1.21 µg/mL, higher than the 1.76 ± 0.08 µg/mL of ascorbic acid used in our work. Similarly, several studies have shown that *Cassia occidentalis* has good antioxidant activity. This is the case of (Evenamede & Kafui, 2017) whose study focused on the antioxidant activity of an ethanolic extract from the roots of this plant. Results obtained are in line with those of (Zongo *et al*., 2023), who showed the existence of antioxidant activity in their work on *Cassia siberien*. These authors were able to explore the antioxidant potential using several tests, including the DPPH free radical scavenging test, with an IC50 value of 21.98 ± 0.19 µg/mL for the extract, confirming the antioxidant activity of the aqueous extract of *Cassia siberien* leaves, which nevertheless remains lower than ours, with an IC50 of 2.74 ± 0.1 µg/mL. These authors also showed a significant difference (p<0.001) between the antioxidant activity of the ethanolic extract of the leaves of the plant in question and that of ascorbic acid, with an IC50 of 13.45 ± 0.1 µg/mL higher than that obtained with a value of 1.76 ± 0.1 µg/mL.

Evaluation of antioxidant activity by ABTS+• free radical scavenging

Antioxidant activity of aqueous extracts of *Cassia occidentalis*, *Jatropha curcas*, *Chromolaena odorata* and gallic acid was qualified spectrophotometrically and shown in Figure 5.

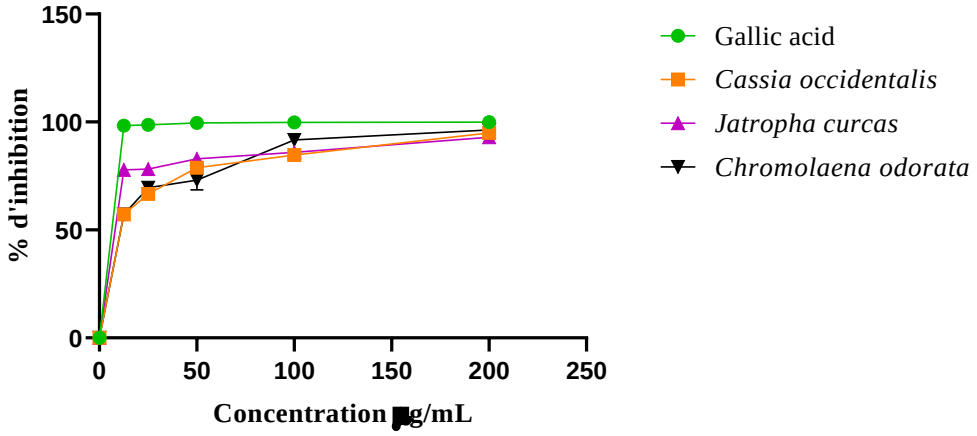

Figure 5: Percentage inhibition curves for ABTS as a function of concentrations of different extracts of *Cassia occidentalis*, *Jatropha curcas*, *Chromolaena odorata* and gallic acid.

Average inhibitory concentration (IC50) values are shown in Table 4.

Table 4: IC50 values for plant extracts and gallic acid obtained by ABTS test

Extracts/Standard	IC50 (µg/mL)
Cassia occidentalis	10.40 ± 0.08***
Jatropha curcas	7.01 ± 0.08***
Chromolaena odorata	10.40 ± 0.08***
Gallic acid	5.6 ± 0.08

*Each value represents the mean±standard deviation of three replicates. Comparison was made using one-way ANOVA followed by the Turkey test; ***p<0.001 indicates a highly significant difference from the standard (control).*

Reacting with gallic acid, ABTS forms the blue ABTS+• radical, which becomes colorless in the presence of the antioxidant. Addition of the antioxidant will reduce this radical and cause the mixture to discolor, as measured spectrophotometrically at 734 nm. Disappearance of the blue coloration is proportional to the ability to inhibit the ABTS+• radical. After measuring absorbance at 734 nm, the results showed that the aqueous extracts of *Cassia occidentalis*, *Jatropha curcas* and *Chromoleana odorata* leaves are active in terms of free radical scavenging. Comparing the activity of the reference molecule (gallic acid) and aqueous plant extracts, the results showed that gallic acid is more active. Depending on their anti-free radical activity, these extracts can be classified into two groups. A first group with an inhibitory concentration 50% (IC50) value of less than 10 µg/mL. This group concerns the aqueous extract of Jatropha curcas leaves with an IC50 of 7.01 ± 0.08 µg/mL. Second group is characterized by IC50 values around 10 µg/mL. This group contains the other two plants (*Cassia occidentalis* and *Chromoleana odorata*) with an IC50 of 10.40 ± 0.08 µg/mL (Table 4). These different IC50 values are statistically significant ($p<0.001$) compared with the reference molecule. Inhibition percentages are high for gallic acid and the various plant extracts (figure 5). Results obtained with the ABTS+• test, based on the proton scavenging capacity of the ABTS+• cationic radical, corroborate those already obtained with the DPPH test on the antioxidant capacity of extracts from the three plants. Analysis of the IC50 values shows that gallic acid (IC50 = 5.6 ± 0.08 µg/mL), *Cassia occidentalis* and *Chromolaena odorata* aqueous extracts (IC50 = 10.35 ± 0.08 µg/mL) and *Jatropha curcas* (IC50=7.01 ± 0.08 µg/mL) all inhibit the ABTS+• cationic radical. In fact, the results obtained concur with those of (Yuri *et al.*, 2024) whose work showed that the *Chromolaena odorata* plant has a high antioxidant activity. Authors studied the antioxidant activity of ethanolic extract of *Chromoaena odorata* leaves using the ABTS+• assay, a model widely used to assess the antioxidant capacity of various compounds. Compared to the ethanolic extract of *Chromoaena odorata* leaves with an IC50 = 29.064 µg/mL, our aqueous extract (with a lower IC50 of 10.35 ± 0.08 µg/mL) shows better free radical scavenging activity. Furthermore, the authors showed a significant difference($p<0.001$) between quercetin (standard) and the ethanolic extract of the leaves of the said plant with an IC50 of 2.29 µg/mL. Then, in their study on *Jatropha curcas* hulls, (Huang *et al*., 2020) showed good proton scavenging ability by the cationic radical ABTS+• with an IC50 of 21.47 ± 0.91 µg/mL higher than that obtained in our work on the aqueous extract of the leaves of the said plant (IC50=10.35 ± 0.08 µg/mL). Similarly, there was a significant $p<0.001$ difference between the activity of aqueous extract of *Jatropha curcas* hull and gallic acid. They also expressed the results of gallic acid activity in IC50 where they obtained a value of 4.08 ± 0.25 µg/mL, which remains higher than the value obtained in our work, i.e. 5.6 ± 0.1 µg/mL. In a study

carried out by (Zongo *et al*., 2023) on Siberian Cassia leaf extract, the IC50 value = 5.96 ± 1.2 µg/mL clearly shows an ABTS-+ radical scavenging activity that is also better than that of our plant with an IC50 of 10.35 ± 0.08 µg/mL. A significant difference $p<0.001$ was observed between the activity of the aqueous extract of *Cassia siberien* leaves and gallic acid. Authors also noted a significant IC50 for gallic acid, with a value of 1.83 ± 0.34 µg/mL, better than that obtained in our work (10.35 ± 0.08 µg/mL).

These results suggest that the antioxidant activity of plant extracts is due to certain bioactive compounds. In their work (N'guessan *et al*., 2007 ; Kagnou *et al*., 2020) showed a linear correlation between total polyphenol content and free radical scavenging activity. According to (Chen & Ho, 1995), the functional groups present in phenolic compounds in general can easily give up an electron or proton to neutralize free radicals. High antioxidant activity of the three plants is therefore linked to their high total phenol content. This assertion is confirmed here by phytochemical analysis followed by DPPH and ABTS tests. In addition to polyphenols, other compounds such as tannins and flavonoids are said to have antioxidant properties, as they are known for their ability to scavenge free radicals (Kanfon *et al*., 2018) and therefore may prevent the development of many chronic diseases (Vadivel & Biesalski, 2011). They are essential for reducing oxidative stress after a stroke, then enabling brain tissue regeneration, resorbing the lesion and thus motor deficits (Quintard, 2014). Several studies in animal models and human subjects have indeed confirmed the bioavailability of phenols to exert a protective role against oxidative stress and free radical damage (Szajdek & Borowska, 2008). Better antioxidant activity observed for the various substances used as standards compared with our extracts is justified by the fact that these substances used are either finished products that have undergone sufficient processing and are recognized as having excellent antioxidant potential. Antioxidant potential of drugs is, among other things, an important factor in research into anti-oxidant molecules.

CONCLUSION

This study was based on phytochemical screening, phenolic compound assay and antioxidant activity assessment in aqueous extracts of *Cassia occidentalis*, *Jatropha curcas* and *Chromoleana odorata* leaves. Phytochemical study revealed the presence of bioactive compounds in each extract, including polyphenols, gallic tannins, flavonoids, saponosides, terpenes and sterols. However, there was a complete absence of alkaloids in *Cassia occidentalis* and *Chromoleana odorata* extracts. Quantitative assaying of phenolic compounds revealed a slightly higher flavonoid content than total polyphenols in each of the plants studied. Antioxidant activity of these extracts was then assessed using two stable radical scavenging methods, DPPH and ABTS. Antioxidant potential of these extracts is marked by high inhibition percentage values and low inhibitory concentration 50% (IC50) values. This interesting antioxidant power of these plant extracts is attributed to the phenolic compounds, which are antioxidant molecules present in the said extracts and therefore capable of neutralising free radicals. Aqueous extracts of the leaves of *Cassia occidentalis*, *Jatropha curcas* and *Chromoleana odorata* could therefore be a potential alternative source of natural antioxidants.

REFERENCES

- Adjanohoun, E. J. & Aké-Assi L. (1979) Contribution au recensement des plantes médicinales de Côte d'Ivoire. Centre National de Floristique de l'Université Nationale de Côte d'Ivoire, Tome 1, 23-30.
- Gönenç, A., Hacisevki, A., Tavil, Y., Çengel, A., Torum, M. (2013). Oxidative stress in patients with essential hypertension : a comparison of dippers and nondippers. *European Journal of Internal Medicine*, 24, 139-144.
- Inbathamizh, L., Ponnu, T.M., Mary, E.J. (2013). In vitro evaluation of antioxidant and anticancer potential of *Morinda pubescens* synthesized silver nanoparticles. *Journal of Pharmaceutical Research*, 6, 32-35.
- Moon, J.K. & Shibamoto, T. (2009). Antioxidant assays for plant and food components. *Journal of Agricultural and Food Chemistry*, 57, 1655-1666.
- Xu, D. P., Li Y., Meng, X., Zhou, Y., Zheng, J., Zhang, J. I., Li, H. B. (2017). Natural antioxidants in food and medicinal plants : Extraction, assessment and ressources. *International Journal of Molecular Sciences*, 18(1), 1-32.
- Marc, F., Davin, A., Degléne, L., Ferrand, C. (2005). Méthodes d'évaluation du potentiel antioxydant dans les aliments. *Erudit M/S*. 2005 ; 20(4), 458-463.
- Zirihi, G.N., Kra, A.K.M., Bahi, C., Guédé-Guina, F. (2003). Plantes médicinales immunostimulantes : critères de sélection, techniques rapides d'extraction des principes actifs et méthodes d'évaluation de l'activité imminogène. *Revue de Médecine et de Pharmacie*, 17, 131-137.
- Békro, Y-A., MamyrBékova-Békro, J.A.., Boua, B.B., Tra Bi, F.H. & Ehilé, E.E. (2007). Étude ethnobotanique et screening phytochimique *de Caesalpinia benthamiana* (Baill.) Herend. et Zarucchi (Caesalpiniaceae). *Sciences & Nature*, 4 (2), 217-225.
- Singeton, V.L., Orthofer, R. & Lamuela-Raventos, M. (1999). Analysis of total phenols and other oxidation substratres and antioxidants by means of Folin-Ciocalteu reagent. *Methods in Enzymology*, 41, 152-177.
- Ben Moussa, M.T., Cherif, R.A., Lekhal, S., Bounab, A. & Hadef, Y. (2022). Dosage des composés phénoliques et détermination de l'activité antioxydante des extraits méthanoliques de

Brocchia cinerea VIS de l'Algérie (Sud-Est). *Algerian Journal of Pharmacy*, 4(1), 49-59.

- Zhishen, J., Mengcheng, T. & Jianming, W. (1999). The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chemistry*, 64(4), 555-559.
- Roukia, H., Karima, D., Mahfoud, H.M. & Mohammed, D.O. (2015). Composition chimique et activité anti-oxydante des huiles essentielles de *Deverra scoparia* Coss & Dur. (Appiaceae). *Lebanese Science Journal*, 16(2), 27-36.
- Torres De Pinedo, N.P. & Morales, C.S. (2007). Synthesis and evaluation of new phenolic-based antioxidants : structure-activity relationship. *Food Chemistry*, 103, 55-61.
- Leong, L.P. & Shui, G. (2002). An investigation of antioxidant capacity of fruits in Singapore markets. *Food Chemistry*, 76(1), 69-75.
- N'guessan, K., Kadja, B., Zirihi, G.N., Traoré, D. & Aké-Assi, L. (2009). Screening phytochimique de quelques plantes médicinales ivoiriennes utilisées en pays Krobou (Agboville, Côte d'Ivoire). *Sciences & Nature*, 6(1), 1-15.
- Kamsi, L.N., Mengome, L.E., Aboughe-Angone, S. & Engonga, P.E. (2020). Etude phytochimique (L.) Link et Cissus quadrangularis (Linn) deux plantes médicinales Gabonaises utilisées contre la filaire *Loa Loa*. *European Scientific Journal*, (16)21, 101-117.
- Saraka, A.I., Abo, K., Coulibaly, K. & Zirihi, G.N. (1018). Etude phytochimique et activité antifongique d'extraits de quelques Euphorbiaceae médicinales utilisées chez les Baoulé du District de Yamoussoukro (Côte d'Ivoire). *European Scientific Journal*,14(30), 256-274.
- Ballo, M.K. Etude phytochimique et évaluation de l'activité sur M. tuberculosis in vitro de 22 plantes utilisées dans le traitement traditionnel de la tuberculose au Mali. Thèse de pharmacie, université des sciences, des techniques et des technologies de Bamako (Mali), 140 p.
- El Hazzat, N., Iraqi, R. & Bouseta, A. (2015). Identification par GC-MS et GCFID-O des composés volatils des olives vertes de la variété « Picholine marocaine » : effet de l'origine géographique. *International Journal of Biological and Chemical Sciences*, 9(4), 2219-2233.
- Falleh, H., Ksouri, R., Chaieb, K., Karray-Bouraoui, N., Trabelsi, N., Boulaaba, M., Abdelly, C. (2008). Phenolic composition of *Cymara cardunculus* L. organs, and their biological activities. *C. R. Bio*.331, 372-379.
- Konan, N.S., Yéo, S.O., Traoré, L., Kouamé, B.A, MamyrBékova-Békro, J.A., Békro, Y-A. (2022). Chemical Composition and Antioxidant Activity of the Essential Oil of *Chromolaena Odorata* Harvested in the Region of the Mountain District of Côte D'ivoire. *Journal of Applied Pharmaceutical Sciences and Research*, 5(1), 21-25.
- Huang, S.L., Wei, H.W.X.Y.Z., Chih, T.L., Wen, S.L., Min, Y.C. & Yung, S.L. (2020). Antioxydant properties of *Jatropha curcas* L. seed shell and keenel extract. *Applied Sciences*, 10(1) : 3279.http://doi:10.3390/app10093279
- Evenamede, K.S., Kafui Kpegba, K., Oudjaniyobi Simalou, O., Boyode, P., Amegnona Agbonon, A. & Gbeassor, M. (2017). Etude comparative des activités antioxydantes d'extraits éthanoliques de feuilles, d'écorces et de racines de *Cassia sieberiana*. *International Journal of Biological and Chemical Sciences*, 11(6), 2924-2935.
- Zongo, E., Busuioc, A., Nâg-Tiero, R.M., Botezatu, A.V., Mihaila, M.D., Mocanu, A.-M., Avramescu, M., Koama, B.K., Kam, S.E., Belem, H., Somda, F.L.S., Ouedraogo, C., Rodica, G.A.O. & Dinica, M. (2023). Exploration of the Antioxidant and Anti-inflammatory Potential of *Cassia sieberiana* DC and *Piliostigma thonningii* (Schumach.) Milne-Redh, Traditionally Used in the Treatment of Hepatitis in the Hauts-Bassins region of Burkina Faso. *Pharmaceuticals*, 16, 133. https://doi.org/10.3390/ph16010133.
- Yuri, P., Rahmah, M.I., Nurzadrina, W. & Suhaera, B.Y. (2024). Optimistion de l'activité anti-oxydante à 90% d'éthanol de *Chromolaena odorata* L. ABTS radical prophylactic. *Journal of katalisator*, 9(1), 62-76.
- N'guessan, J.D., Zirihi, G.N., Kra, A.K.M., Kouakou, K., Djaman, A.J. & Guédé-Guina, F. (2007). Free radical scavenging activity, flavonoid and phenolic contents of selected Ivorian plants. *International Journal of Natural and Applied Sciences*, 4, 425-429.
- Kagnou, H., Sinalou, O., Tchani, G.W., Sanvee, S., Agbodan, K.A., Toundou, O. & Kpegba, K. (2020). Etude phytochimique et activité anti-oxidante comparatives des trois variétés de *Catharanthus roseus* (L) G. Don. *International Journal of Biological and Chemical Sciences*, 14(6), 2352-2361.
- Chen, C.W. & Ho, C.T. (1995). Antioxidant properties of polyphenols extracted from green tea and black tea. *Radical-chemistry*, 2, 35-46.
- Kanfon, R.E., Gnawe, M., Dossa, A.C.P., Yedomonhan, H. Wot, To D.V. & Sohounhloue, C.K.D. (2018). Caractérisation physico-chimique et évaluation de l'activité antiradicalaire des extraits de sept morphotypes de gombo (Abelmoschus spp.) cultivés au Bénin. *International Journal of Biological and Chemical Sciences*,12(3), 1447-1458.
- Vadivel, V. & Biesalski, H.K. (2011). Contribution of phenolic compounds to the antioxidant potential and type II diabetes related enzyme inhibition properties of *Pongamia pinnata* L. Pierre seeds. *Process Biochemistry*, 46(10), 1973-1980.
- Quintard, H. (2014). Etude des propriétés neuroprotectrices et neurorégénératives du

MLC901, issu de la Médecine Traditionnelle Chinoise face à l'ischémie globale et au traumatisme crânien chez le rongeur. Sciences agricoles. Université Nice Sophia Antipolis. 2014; 206 p.

- Szajdek, A. & Borowska, E.J. (2008). Bioactive compounds and health-promoting properties of berry fruits: A review. *Plant Foods for Human Nutrition*, 63, 147-156.