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Quantitative Phytochemical Screening and In-vitro Antioxidant Activities of *Jatropha gossypifolia* L. Leaf Extracts Aiming Antimicrobial and Therapeutical Applications

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ABSTRACT

Total phenolic (polyphenols, flavonoids, and condensed tannins) contents and antioxidant property (DPPH, ABTS, FRAP) were evaluated in hexane (HE), acetone (AC), ethanol (ET), 70% (v/v) ethanol/water (EW), and powder extracts of *Jatropha gossypifolia* leaves using spectrophotometric methods. The lowest and highest contents of polyphenols (10.27 ± 0.36 and 21.23 ± 0.95 mg GAE/g), flavonoids (4.83 ± 0.42 and 12.53 ± 0.58 mg QE/g), and condensed tannins (3.03 ± 0.18 and 7.13 ± 0.12 mg CE/g) were recorded in powder and EW, respectively. EW exhibited the highest scavenging activities displayed by the lowest IC50 values (0.23 ± 0.01 mg/mL DPPH and 0.2 ± 0.03 mg/mL ABTS). Moreover, it recorded the highest reducing power (92.85 ± 1.25 mg AAE/g), followed by HE (72.73 ± 1.99 mg AAE/g) and AC (41.84 ± 0.47 mg AAE/g). Relevant correlations were found between the polyphenols, flavonoids, condensed tannins and reducing antioxidant power ($r = 0.986$, $p < 0.01$; $r = 0.958$, $p < 0.01$; $r = 0.950$, $p < 0.05$; respectively). Based on their phytochemical and antioxidant properties, PCA classified the five extracts into three distinct clusters. The results of this study indicate that EW contains powerful bioactive molecules with noteworthy antiradical and electron donating properties. Therefore, it may be a promising candidate in the search for natural compounds with therapeutic and food/crop preservative applications.

1. Introduction

Plants are known to have beneficial therapeutic effects, and much research have been done on their ethnomedicinal uses. Plant-derived drugs remain an important resource in developing countries, and approximately 62–80% of the world's population still relies on traditional medicines for the treatment of common illnesses [1]. Plants produce several secondary metabolites that have many important biological properties, and antioxidant activity just to highlight a few. Antioxidants are important substances that have the ability to protect the organism as well as food products from the damage caused by oxidative stress. Due to this ability, there is a special interest in the presence of natural antioxidants in medicinal plants that may help the organism to maintain the normal balance of the free radicals naturally produced [2]. Antioxidants are classified into five types based on their mode of action: radical scavenging antioxidants, which are responsible for breaking the chain of radical propagation; chelators, which form complexes with metals and prevent them from initiating the formation of radicals; extinguishers, which deactivate high-energy oxidant species; oxygen scavengers, which remove oxygen from the systems, preventing their destabilization; and regenerators of antioxidants, which reconstitute antioxidants present in the food when they become radicals [3]. The sources of natural antioxidants can be extremely varied, because practically all plants contain antioxidants that allow them to protect themselves from solar radiation and pests, as well as to regulate the production of chemical energy.

Jatropha gossypifolia belongs to the family of Euphorbiaceae, and the order of Geraniales [4]. This species is worldwide known as "bellyachebush", "purge nut" and "red fig-nut flower" in English, and "herbe à mal de ventre" and "medicinier sauvage" in French. There are also the following vernacular names for *J. gossypifolia*: "baga" in Malinke and Dioula; "sataman" in Bambara, "lapalapa" in Yoruba; "athalai," in Hindi;

"pinhao-roxo" in Brazilian [5]. In Cameroon, this plant is known by various popular names, and the most common are "maagami balmol" and "sambaali", which derived from Haoussa and Ffulde, two local dialects spoken in the Sudano Sahelian zone of the country. This species is widely distributed in tropical and subtropical regions of Africa and the Americas [6]. *Jatropha gossypifolia* is an erect, monoecious, deciduous, soft wooded, perennial shrub that grows to a height of 2.5 m on average, but can reach 4 m in some areas. It has sticky leaves that are covered in extra-floral nectaries, deeply 3-lobed, and bronze when young but green when mature. Flowers are deep red with yellow centres [7]. Fruits are three-celled, explosively dehiscent capsules with one seed per locule. Capsules are green when mature, turning pale green before dehiscence, and are sparsely hairy. The seeds are spherical, carunculate, and greyish-brown, with a thick layer of starchy endosperm [8]. It has been reported that the name *Jatropha* is derived from the Greek words "jatos" and "trophe", which mean doctor and food, respectively, and refer to the medicinal and culinary properties of this plant [5].

The preparations based on this plant are commonly used for several human and veterinary diseases. Roots are traditionally used to treat leprosy, the decoction of leaves is used for boils, eczema, venereal diseases, and both as purgative and febrifuge, while seeds are used as emetic, purgative, and for body pain [9]. In Cameroon, the sap of *J. gossypifolia* is used by traditional healers as a disinfectant for wounds and the indigenous treatment of hepatitis. Among other applications, it has been reported to have anticoagulant [2]; hypoglycemic and antidiabetic [4, 7]; antiulcer [7]; antihypertensive [10]; anti-inflammatory [11]; analgesic, antipyretic [12]; antimicrobial [1, 13]; antitumor [14]; antianemic [15]; antifertility [16]; antiarrhythmic [17]; hepatoprotective [18]; and neuroprotective [19] properties. These pharmacological properties of *J. gossypifolia* may be related to the phytochemistry of this plant. *Jatropha* species are rich in phytochemicals. Previous studies reported alkaloids, coumarins, flavonoids, lignoids, phenols, saponins, steroids, tannins, and terpenoids in different extracts from different parts of *J. gossypifolia* [20] and some of them have been revealed to be antioxidant [2, 18, 21].

Given the dismayed rise of health effects associated with synthetic antioxidants and the potential applications of this plant, the current study aims to provide an up-to-date overview of phytochemistry and current

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data on antioxidant activity of different extracts from leaves of *J. gossypifolia*, which may be significant in providing insights for validation of its popular use and exploration of new source of herbal drugs and/or natural bioactive products.

2. Experimental Methods

2.1 Plant Material

The mature fresh leaves of *J. gossypifolia* (Fig. 1) were harvested in June 2019 on the edge of the Kaliao Mayo river, located in Maroua I Subdivision, Diamare Division, Far North Region. The leaves were aseptically transported to the laboratory, where they were identified by botanical experts of the Maroua regional unit of the National Institute of Agricultural Research (IRAD).



Fig. 1 *Jatropha gossypifolia* L. leaves

2.2 Preparation of Extracts

The collected leaves were washed with tap water and sun-dried until a constant weight was reached. The dried leaves were pulverized with the help of a mechanical blender, sieved, and transferred into a sterile glass container. A part of this powder was used at state without any extraction technique. Fifty grams of the powdered plant were extracted at room temperature by Soxhlet-assisted maceration using 100% (v/v) acetone, ethanol, and hexane as solvents and by maceration using 70% (v/v) ethanol/distilled water. Both extraction techniques were used according to the methods described by Jain et al. [18] and Saleem et al. [19], respectively. Five hundred millilitres of solvent were used, and occasional shaking and stirring were done for 24 hours during maceration. However, 6 hours were required for the Soxhlet extraction. The mixture was then filtered through Whatman filter paper (no 1) and the filtrate obtained was concentrated at low temperature (40-50 °C) with a rotary evaporator (IKA, Germany). The four dried extracts and the powder extract were stored at 4 °C for further use.

2.3 Quantitative Phytochemical Screening

2.3.1 Extract Preparation before Quantification

The extracts were preliminary prepared before the evaluation of the polyphenols, flavonoids, and condensed tannin contents, as well as antioxidant activity. This was carried out according to the procedure described by Kim et al. [22]. A gram of dried extract was dispersed into 25 mL of solvent specific to each extract. The set was kept under stirring at room temperature for 24 hours. The mixture was filtered (Whatman filter paper no. 1) and the recovered filtrate was kept for the next analysis.

2.3.2 Determination of the Total Polyphenol Content

The Folin-Ciocalteu method was used to quantify total polyphenols as described by Khalifa et al. [23]. Twenty microlitre of the filtered extract was introduced into test tubes containing 2980 µL of distilled water. Further, 500 µL of the Folin-Ciocalteu reagent (10-fold diluted) and 400 µL of Na₂CO₃ solution (20%, v/v) were added. Both reaction and control (only reagents) tubes were vortexed. After 20 min of incubation at room temperature in the dark, the optical density (OD) values were measured at 760 nm wavelength. Calibration was performed using gallic acid (0.2 g/mL) as standard solution, and the results obtained using a standard curve ($y = 0.031x + 0.0096$; $R^2 = 0.9837$) were expressed as mg gallic acid equivalent/g of extract (mg GAE/g).

2.3.3 Determination of the Total Flavonoid Content

The colorimetric aluminium chloride method was used to evaluate total flavonoids as described by Ezeonu [24]. One hundred microlitre of the filtered extract was mixed with 2.4 mL of distilled water and 0.15 mL of Na₂NO₂ (5%, v/v). After a stand of 6 min, 0.3 mL of freshly prepared AlCl₃, <https://doi.org/10.30799/jnpr.106.22080102>

6H₂O (10%, v/v) was added to the reaction tube. After 5 min of incubation at room temperature, 1 mL of 1 M NaOH was dropped into the tube and after shaking the OD values were read at 510 nm using a UV-visible spectrophotometer. The flavonoid content in each extract was estimated using the calibration curve ($y = 0.0017x - 0.0014$; $R^2 = 0.9958$) set from different concentrations of a standard solution of quercetin (0-40 µg/mL) and the results were expressed as quercetin mg equivalent/g of extract (mg QE/g).

2.3.4 Determination of the Content of Total Condensed Tannins

The contents of condensed tannins in crude extracts and powder were determined using the vanillin sulfuric acid method as described by Sun et al. [25]. Extract filtrate (50 µL) was mixed with 3 mL of vanillin (4%), and 1.5 mL of concentrated sulfuric acid. After homogenization, the preparation was incubated at room temperature for 30 min, and OD values were read at 500 nm against a blank containing pure ethanol. The contents of condensed tannins were extrapolated from a standard curve ($y = 0.0418x + 0.1151$; $R^2 = 0.9737$) of catechin (0-40 µg/mL) used as reference. The content of condensed tannins was expressed in mg of catechin equivalent per gram of extract (mg CE/g).

2.4 Antioxidant Activity Assay

2.4.1 DPPH Free Radical Scavenging Activity

The DPPH (1,1-diphenyl-2-picrylhydrazyl) activity measures the free radical scavenging capacity of the extracts. This activity was measured according to the protocol described by Rofida [26]. A volume of 0.5 mL of each methanolic solution of the extracts at different concentrations (from 0 to 10 mg/mL) was added to 2 mL of the DPPH solution (0.025 g/L). At the same time, a negative control is prepared by mixing 0.5 mL of methanol with 2 mL of the methanolic solution of DPPH. The OD was read against a blank at 517 nm wavelength after 60 min incubation in the dark at room temperature. L-ascorbic acid was used as positive control, and the absorbance was measured under the same conditions as both extracts and blank. The results were expressed as inhibition percentage of free radicals by each extract. It was calculated using the following formula:

$$\text{Inhibition percentage (\%)} = \frac{\text{OD (DPPH)} - \text{OD (Sample)}}{\text{OD (DPPH)}} \times 100$$

where OD (DPPH) is the OD value of the control test tube (test tubes containing DPPH solution only), OD (sample) is the OD value of the reaction test tube (tube containing DPPH solution and extracts). The inhibitory concentration (IC₅₀) was deduced from the curve of inhibition percentage as a function of extract concentration. IC₅₀ was used to quantify the concentration of antioxidants in each extract to remove 50% of free radicals from the living system.

2.4.2 ABTS Free Radical Scavenging Activity

ABTS (2,2'-azion-bis(3-ethylbenzothiazoline -6-sulfonic) acid) scavenging activity was evaluated according to the method described by Re et al. [27]. To prepare ABTS, 0.038 g of ABTS and 0.04 mg of potassium persulfate were mixed in 10 mL of distilled water. The solution was homogenised for 5 min and incubated for 16 h in dark conditions at room temperature to allow formation of the stable free radical. One hundred and fifty microlitre (150 µL) of the extracts (at concentrations ranging from 0 to 10 mg/mL) and 2 mL of ethanol diluted ABTS⁺ were mixed (assay tubes). Similarly, 2 mL of ethanol diluted ABTS⁺ was mixed with the extraction solvent to serve as blank tube. The mixtures were incubated for 10 min at room temperature, and the OD values were measured at 734 nm using a UV-visible spectrophotometer. L-ascorbic acid was considered as the positive control. Based on OD values, the percentages of free radical inhibition were calculated using aforementioned formula as above. The inhibition concentration (IC₅₀) of each extract was deduced from the ABTS inhibition percentage as a function of extract concentration curve.

2.4.3 Ferric Reducing Antioxidant Power (FRAP) Assay

The ferric reducing antioxidant power evaluates the ability of the extracts to reduce iron (III) to iron (II). This was performed according to the method described by Vijayalakshmi and Ruckmani [28]. In the reaction test tube, 1 mL of extract was mixed to 2.5 mL of phosphate buffer saline (0.2 M, pH 6.6) and 2.5 mL of potassium hexacyanoferrate solution [K₃Fe(CN)₆] at 1%. The mixture was homogenised and incubated for 30 minutes at 50 °C in a water bath, then 2.5 mL of 10% trichloroacetic acid were added and the mixture was centrifuged at 3000 rpm for 10 minutes. After centrifugation, 2.5 mL of the collected supernatant were added to 2.5 mL of distilled water and 0.5 mL of ferric chloride (FeCl₃) solution at 0.1%. L-ascorbic acid was used as standard. After reading the OD values at 700 nm, the results were expressed in mg ascorbic acid equivalent by gram of

extract (mg ACE/g) from a calibration curve obtained with different concentrations of ascorbic acid (0–10 µg/mL).

2.5 Statistical Analysis

The measurements were done in triplicate and the results were displayed as mean ± standard deviation. One-way ANOVA was used to determine the mean differences among the plant extracts and the HSD Tukey's multiple range test was applied to discriminate the pair of means significantly different at $p < 0.05$. Pearson's coefficient (r) was used to determine the correlations between the phytochemical and antioxidant variables at a significance level of $p < 0.05$. Principal Component Analysis (PCA) was used to divide the variables under consideration into two main components, and find out the relationship between the different extracts. PCA was performed with XLSTAT, while ANOVA and Tukey tests were run with Statgraphics software.

3. Results and Discussion

3.1 Phenolic Contents of Extracts and Powder of *Jatropha gossypifolia* Leaves

Table 1 indicates the polyphenol, flavonoid, and tannin contents of the extracts of *Jatropha gossypifolia*. Total polyphenols ranged significantly ($p < 0.05$) from 10.27 ± 0.36 mg GAE/g to 21.23 ± 0.95 mg GAE/g in the powder and hydroethanolic (70%, v/v) extract of *J. gossypifolia* leaves, respectively. The similar result is observed with the flavonoids content of *J. gossypifolia* which varied from 4.83 ± 0.42 mg QE/g for the powder to 12.53 ± 0.58 mg QE/g for the aqueous ethanolic extract. The same trend was observed with condensed tannins, for which the values varied between 3.03 ± 0.18 mg CE/g and 7.13 ± 0.12 mg CE/g.

Globally, phytochemicals were found lower concentrated in powder than in extracts of *J. gossypifolia*. This might be explained by the key role of solvent during the extraction procedure. When, the plant part is used without any extraction, the bioactive compounds remain trapped inside the cell and are therefore less available. While the use of solvent makes easier removal of these bioactives during the extraction and their concentration in the final dried extract. The same difference was observed between the four extracts. It is believed that the different solvents and extraction techniques influences the contents of bioactive compounds [29]. Generally, it is admitted that the mixture of polar and non-polar solvents is better than using just polar or non-polar ones. With regards to the results of table 1, water-ethanol (70%, v/v) was the best co-solvent system [30]. This has been confirmed by the highest extraction yield recorded by ethanol water solvent (29.72%). While hexane, acetone, and ethanol registered extraction yield of 22.77%, 26.93%, and 27.26%, respectively. According to Dahmoune et al. [31], adding a small amount of water to an organic solvent such as ethanol or methanol increases the mass transfer process as well as the solubility of the phenolic compounds from the plant matrix to the solvent.

The total polyphenol, and flavonoid contents of our extracts were higher than those reported by the Brazilian research team of Carvalho et al. [21] in the leaf extracts of *J. multifida*. These authors found 0.13 mg GAE/g and 2.322 mg QE/g of polyphenols and flavonoids, respectively. The same tendency was observed with flavonoids (0.540 mgQE/g) and tannins (0.870 mgCE/g) in the leaf extract of *J. curcas* [32]. In the same way, Ajayi et al. [33] recorded 1.5 mg QE/g, 2.5 mg GAE/g and 4 mg CE/g of flavonoids, polyphenols, and tannins, respectively, in the leaf extract of *J. gossypifolia*. All these differences may be related to the origin and geographical location of the *Jatropha* species [34]. The Far North of Cameroon is a very sunny region in which plants are forced to produce phenolic compounds to protect themselves from UV radiation of the sun. Furthermore, the aridity of the soil in this region promotes the accumulation of these phytoconstituents in sun-exposed parts of the plant, such as leaves, fruits, and stems [35].

Table 1 Polyphenols, flavonoids, and tannins contents of leaf extracts and powder of *Jatropha gossypifolia*

Extracts	Yield (%)	Total polyphenols (mg GAE/g)	Total flavonoids (mg QE/g)	Condensed Tannins (mg CE/g)
AC	26.93	14.76 ± 0.46^c	7.51 ± 0.35^b	5.51 ± 0.11^c
ET	27.26	12.92 ± 0.27^b	7.72 ± 0.11^b	4.72 ± 0.14^b
HE	22.77	17.04 ± 1.12^d	8.96 ± 0.25^c	5.96 ± 0.07^d
EW	29.72	21.23 ± 0.95^e	12.53 ± 0.58^d	7.13 ± 0.12^e
Powder		10.27 ± 0.36^a	4.83 ± 0.42^a	3.03 ± 0.18^a

Values in the same column with different superscript (a, b, c, d, e) are significantly different ($p < 0.05$). AC: Acetone extract; ET: Ethanol extract; EW: Ethanol/water (70%, v/v) extract; HE: Hexane extract

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The high content of phytochemicals in *J. gossypifolia* leaves collected in the northern Cameroon opens the way to several applications, both as a therapeutic and natural preservative. Indeed, phenolic compounds have been identified as bioactive and useful in the treatment of several stress-related diseases, the improvement of certain vital functions of living organisms, the fight against microbial adulteration of foodstuffs etc. [11, 13].

3.2 Antioxidant Activity of Leaf Extracts and Powder of *Jatropha gossypifolia*

The radical scavenging activities and reducing antioxidant power of extracts and powder from *Jatropha gossypifolia* leaves are presented in Table 2. Free radical scavenging activity was performed using DPPH and ABTS assays, while total antioxidant content was determined by the FRAP method. The scavenging activities were expressed in inhibition concentration 50 (IC50). This parameter evaluates the concentration of a compound able to inhibit 50% of the free radicals present in a living system. The IC50 determines the scavenging activity strength of the compound, so, the lower the IC50 values, the greater the strength in the scavenging activity of free radicals of the compounds. Scavenging activities based on either DPPH or ABTS free radicals varied significantly ($p < 0.05$) from one extract to another. In the ethanol water extract and the powder of *J. gossypifolia* leaves, IC50 values changes from 0.23 ± 0.01 mg/mL to 2.25 ± 0.03 mg/mL, respectively. The same trend was observed with ABTS IC50 values, which ranged from 0.2 ± 0.03 mg/mL to 1.15 ± 0.04 mg/mL. Therefore, it has been noted that the ethanol water extract had the highest scavenging activity against DPPH and ABTS radicals while the powder displayed the lowest scavenging activity against the same free radicals. Moreover, the scavenging activity of all the extracts and powder was lower than that of ascorbic acid (with IC50 values of 0.014 ± 0.001 mg/mL DPPH and 0.018 ± 0.001 mg/mL ABTS) use as standard. Based on IC50 values, the leaf extracts of *J. gossypifolia* collected in Maroua town (Cameroon) were found more active than the methanol extract of *J. curcas* as reported by Sharma and Singh [36] in India.

However, IC50 values of hexane (349.53 ± 2.27 µg/mL), ethyl acetate (192.11 ± 2.04 µg/mL), and methanol (48.59 ± 0.61 µg/mL) extracts reported by Dias et al [37] with the leaves of *J. mollissima* showed that they were more active against DPPH radical than our extracts from the leaves of *J. gossypifolia*. Scavenging activity against DPPH (0.55 ± 0.01 mg/mL, 2.25 ± 0.03 mg/mL and 0.014 ± 0.001 mg/mL respectively) and ABTS (0.69 ± 0.01 mg/mL, 1.15 ± 0.04 mg/mL and 0.018 ± 0.001 mg/mL respectively) radicals were significantly different ($p < 0.05$) for acetone extract, powder, and standard compound ascorbic acid. This showed that scavenging antioxidant activity depends on the assay used [38]. It is assumed that the ABTS test is a better method for determining scavenging antioxidant activity than the DPPH test. The ABTS test has a higher sensitivity as well as repeatability [39]. The reducing power (FRAP test) determines the ability of an extract to donate electrons. The outcome of this test was expressed in terms of ascorbic acid-equivalent reducing activity. It was observed that ferric reducing antioxidant power changed significantly ($p < 0.05$) between the different samples of *J. gossypifolia* leaves. The powder (15.16 ± 1.14 mg AAE/g) and ethanol water (92.85 ± 1.25 mg AAE/g) extracts recorded the lowest and the highest values of the reducing antioxidant power. The values obtained by all extracts were higher than those obtained by Prakash et al [40] with some Indian medicinal plants. The bark extracts of *A. indica*, *Casuarina equisetifolia*, and *Cinnamomum zeylanicum*, flowers of *Indigofera tinctoria*, and fruits of *Lawsonia inermis* yielded reducing power values ranging from 0.6 to 2.8 mg AAE/g.

Table 2 DPPH and ABTS radical scavenging activities and ferric reducing antioxidant power of powder and extracts of *Jatropha gossypifolia* leaves

Extracts	IC ₅₀ DPPH (mg/mL)	IC ₅₀ ABTS (mg/mL)	FRAP (mg AAE/g)
AC	0.55 ± 0.01^{dA}	0.69 ± 0.01^{dB}	41.84 ± 0.47^c
ET	0.75 ± 0.01^{eA}	0.81 ± 0.04^{eA}	35.63 ± 1.95^b
HE	0.37 ± 0.01^{cA}	0.35 ± 0.04^{cA}	72.73 ± 1.99^d
EW	0.23 ± 0.01^{bA}	0.2 ± 0.03^{bA}	92.85 ± 1.25^e
Powder	2.25 ± 0.03^{fA}	1.15 ± 0.04^{fB}	15.16 ± 1.14^a
AA	0.014 ± 0.001^{aA}	0.018 ± 0.001^{aB}	-

Values in the same column with different lowercase superscript (a, b, c, d, e) are significantly different ($p < 0.05$). Values in the same line (only for scavenging activity) with different uppercase superscript (A, B) are significantly different ($p < 0.05$). AA: Ascorbic Acid; AC: Acetone extract; ET: Ethanol extract; EW: Ethanol/Water (70% v/v) extract; HE: Hexane extract. ABTS: 2,2'azion-bis (3-ethylbenzothiazoline-6-sulfonic) acid; DPPH: 1,1-diphenyl-2-picrylhydrazyl; FRAP: Ferric Reducing Antioxidant Power; IC50: Inhibition Concentration 50.

3.3 Correlation and Multivariate Analysis

Table 3 presents the correlations between the parameters evaluated in the extracts of *J. gossypifolia* leaves. Significant positive correlations were

found between polyphenols and both flavonoids ($r = 0.975$; $p < 0.01$) and tannins ($r = 0.970$; $p < 0.01$). The same trend was found between reducing power and polyphenols ($r = 0.986$; $p < 0.01$), flavonoids ($r = 0.958$; $p < 0.01$), and tannins ($r = 0.950$; $p < 0.05$). This could indicate that the majority of the bioactive phytochemicals found in *J. gossypifolia* react with reactive oxygen species as electron donors. This pathway also implies reduction of an oxidized antioxidant molecule to regenerate the “active” reduced antioxidant. The correlation between DPPH and ABTS was significant ($r = 0.887$; $p < 0.05$). This could be explained by the specificity of both parameters in revealing the scavenging radical property of samples [41]. ABTS was significantly connected to polyphenols ($r = -0.972$; $p < 0.01$), flavonoids ($r = -0.938$; $p < 0.01$), and tannins ($r = -0.974$; $p < 0.01$) contents. The negative correlation noted between scavenging activity and phenolic contents of the plant extracts is justified by the fact that scavenging activity has been expressed in terms of inhibition concentration 50 (IC50). So, the lower IC50 lead higher the phenolic content as well as the scavenging.

A similar correlation was reported between scavenging activity and flavonoid content ($r = -0.82$; $p < 0.01$) with kounou, a local sorghum beverage from the Far North of Cameroon [42]. Moreover, Bayoï et al. [43] revealed that the DPPH radical scavenging activity appears to depend on phenols and flavonoid contents of some Cameroonian plant extracts.

Table 3 Pearson correlation matrix between phytochemical contents and antioxidant activity of extracts of *J. gossypifolia* leaves

Polyphenols	Flavonoids	Tannins	DPPH	ABTS	FRAP
1.000					
0.975**	1.000				
0.970**	0.950*	1.000			
-0.819	-0.820	-0.929*	1.000		
-0.972**	-0.938**	-0.974**	0.887*	1.000	
0.986**	0.958**	0.950*	-0.813	-0.987**	1.000

Correlation coefficients are statistically significant at $p < 0.05$ (*) and $p < 0.01$ (**). DPPH : scavenging activity against 1,1-diphenyl-2-picrylhydrazyl ; ABTS : scavenging activity against 2,2'-azion-bis (3-ethylbenzothiazoline -6-sulfonic) acid ; FRAP : Ferric reducing antioxidant power

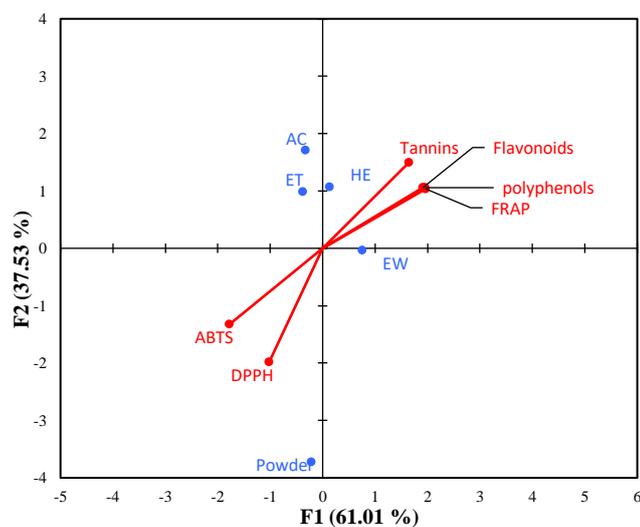


Fig. 2 PCA of parameters and samples from *J. gossypifolia* leaves. AC: Acetone extract; ET: Ethanol extract; EW: Ethanol Water (70%) extract; HE: Hexane extract. ABTS: 2,2'-azion-bis(3-ethylbenzothiazoline-6-sulfonic) acid; DPPH: 1,1-diphenyl-2-picrylhydrazyl ; FRAP: Ferric Reducing Antioxidant Power. DPPH and ABTS scavenging activities are expressed as IC50.

Fig. 2 shows a biplot in which the six quantitative variables are reduced to two main factors, F1 and F2, which account for 61.01 percent and 37.53 percent of the total variance, respectively. This resulted from varimax rotation. The polyphenol and flavonoid contents, as well as the reducing antioxidant power, were all positively loaded by the first factor F1. The same factor was found to be negatively related to ABTS scavenging activity, while the DPPH scavenging activity was negatively loaded by the factor F2. Ethanol water extract was loaded to F1, indicating that it contained the most polyphenols and flavonoids as well as the highest ferric reducing antioxidant power. Given its location, this extract can be classified as a separate cluster. Acetone, ethanol, and hexane extracts were all linked to the positive part of factor F2. Because the three extracts are so close together, they may be grouped into another cluster. Powder extract was loaded on the minus side of factor F2 and was very far away from the other extracts. This means it forms a distinct cluster. As a result, <https://doi.org/10.30799/jnpr.106.22080102>

the five samples obtained from *J. gossypifolia* leaves formed three distinct clusters. This confirms the effect of the type of solvents used on the bioactive properties of the plant extracts [44]. The clustering was in accordance with the techniques used during the extraction process. The leaves of *J. gossypifolia* (Euphorbiaceae) were used after being powdered on one side and maceration and soxhlet-assisted extraction on the other. As a result, the location of the samples in the PCA biplot was determined by the extraction method, which is known to influence their physicochemical and biological properties. This was consistent with the findings of Shang et al. [45], who demonstrated the effect of extraction methods on the physicochemical properties and activities of *Astragalus cicer* (Fabaceae).

4. Conclusion

In this study, various leaf extracts of *J. gossypifolia* were quantitatively assayed for its phytochemical contents and antioxidant activities. The EW presented the highest quantity of phytochemicals and the best antioxidant activity when compared to AC, HE, and ET likewise the least. This has been emphasized by the lowest IC50 values of the EW. These promising results from antioxidant assays demonstrated that the phenolic compounds of *J. gossypifolia* leaves might replace synthetic toxic antioxidant use in the treatment of stress-related diseases (hepatitis, gastroenteritis, etc...), crop protection, as well as food preservation. Regarding the basic empirical knowledge on the health benefits potential of this plant in northern Cameroon as well as the scientific proof of its biological activity provided by the current study, further research is needed to characterize its phenolic compounds in depth.

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