

Original Article

IN VITRO ANTIOXIDANT AND ANTI-INFLAMMATORY ACTIVITIES OF *ALCHORNEA CORDIFOLIA* (SCHUMACH AND THONN.) MÜLL. ARG. AND *ANTROCARYON KLAINEANUM* PIERRE EXTRACTS

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Received: 12 May 2020, Revised and Accepted: 25 Jun 2020

ABSTRACT

Objective: This study aims to evaluate the *in vitro* antioxidant, anti-inflammatory activities of the aqueous and hydroethanolic extracts recipe of *Alchornea cordifolia* and *Antrocaryon klaineinum*. A preliminary phytochemical screening was carried out.

Methods: The total phenols content was determined by the Folin Ciocalteu reagent method, while the antioxidant activity of both extracts was characterized by the 2,2-diphenyl-1-picrylhydrazil (DPPH) and β -carotene assays. The anti-inflammatory activity of the extracts was evaluated as the inhibition of Bovine Serum Albumin (BSA) denaturation and antiproteinase activity.

Results: The aqueous extracts of *Alchornea cordifolia* and *Antrocaryon klaineinum* contained more polyphenols [270 mg Ascorbic acid equivalent (AAE)/g dry weight (dw)] than the hydroethanolic recipe extract (262.41 mg AAE/g dw) at the same concentration level. On the other hand, the aqueous and hydroethanolic recipe extract had the same radical scavenging activity with the antiradical power of 0.851 and 0.830, respectively. Similarly, the recipe extract had the same reducing activity with reducing the power of 94.2 ± 2.03 mg EAA/g dw and 97.4 ± 4.16 mg EAA/g dw for the aqueous and hydroethanolic recipe extract of *Alchornea cordifolia* and *Antrocaryon klaineinum* respectively. For the anti-inflammatory activity it was observed that both extracts possess the same activity as Diclofenac® with an IC₅₀ of 50.21 μ g/ml. The phytochemical screening of the extracts revealed the presence of alkaloids, flavonoids, carbohydrates, phenols and tannins, which may account for their activities.

Conclusion: The plant recipe extract studied possess antioxidant and anti-inflammatory potentials, which may be beneficiary to its consumers.

Keywords: Antioxidant, Anti-inflammatory, *Alchornea cordifolia*, *Antrocaryon klaineinum*, Phytochemical study, Recipe

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DOI: <http://dx.doi.org/10.22159/ijpps.2020v12i8.38272>. Journal homepage: <https://innovareacademics.in/journals/index.php/ijpps>.

INTRODUCTION

Free radicals are known as Reactive Oxygen Species (ROS) are produced by the human body in a normal metabolic process and its environ. There is a permanent system of antioxidant defense of the body against ROS but, an imbalance between antioxidants and pro-oxidants in favor of pro-oxidants leads to oxidative stress [1]. Oxidative stress and inflammation are strongly linked [2, 3]. Inflammation, when it reacts disproportionately, it may be inappropriate, pathological and altering homeostasis thus, contributing to the onset of disease [4]. The synthetic anti-inflammatories and antioxidants currently used are associated with serious side effects like weight gain, gastric irritation [5]. Therefore, the development of anti-inflammatory drugs with little or no side effects is needed from medicinal plants. *Alchornea cordifolia* (Schumach. and Thonn.) Müll. Arg (Euphorbiaceae), also called "aboe" in ewondo, "dibobondji" in Douala, is one of such plants which is widely distributed in tropical and subtropical regions of Africa. In the central and coastal regions of Cameroon, Dibong *et al.* [6] reported that the leaves of this plant are used for the treatment of anemia. This plant has many order traditional applications, including treatment of pain and inflammatory disorders, hormonal-related gynecological disorders, infertility, urinary, respiratory and intestinal problems as well as malaria-like fevers and anti-HIV1 integrase activity [7-16]. Earlier studies on the chemical constituents of *A. cordifolia* identified compounds such as tannins, phenolic acids, flavonoids and alkaloids [11]. The varied popular uses of different parts of *A. cordifolia* have led to many modern pharmacological investigations like antibacterial [16], antifungal [17], spasmolytic [18] and reproductive activities [13].

Antrocaryon klaineinum Pierre (Anacardiaceae) called "angongui" in ewondo, "ngonga" in bassa is an evergreen tree, deciduous, medium-sized up to 35 to 45 m tall [19]. The barks of *Antrocaryon klaineinum* are used as a decoction for the treatment of painful periods, sales ailments and even sexually transmitted infections in Cameroon [6]. The crude extract and the isolated compounds of this plant were evaluated for their ability to inhibit the 3D7 strain of *Plasmodium falciparum* and were evaluated to treat liver complaints and to facilitate the production of breast milk [20, 21]. Phytochemical studies of *A. klaineinum* extracts have shown the presence of secondary metabolites with antimalarial properties [22]. As of now, very few authors reported the evaluation of the antioxidant and anti-inflammatory activities of the recipe from these plant. Therefore, the aim of the present study was to evaluate the antioxidant and anti-inflammatory activities of the aqueous and hydroethanolic extracts recipe of *Alchornea cordifolia* (Schumach and Thonn.) Müll. Arg. and *Antrocaryon klaineinum* Pierre.

MATERIALS AND METHODS

Plant extracts

The leaves of *Alchornea cordifolia* were collected in the locality of Mfou Center Region of Cameroon in July 2017. The plant identification was done at the Cameroon National Herbarium by comparison with specimen number 9657/SRF/Cam of *Alchornea cordifolia* (Schumach and Thonn.) Müll. Arg. (Euphorbiaceae).

The barks of *Antrocaryon klaineinum* were collected in July 2017, in Ebolowa (Southern Region of Cameroon). The plant identification

was done at the Cameroon National Herbarium by comparison with specimen number 1742/SRF/Cam of *Antrocaryon klaineum* Pierre (Anacardiaceae).

The fresh vegetal material collected was dried in the shade at room temperature (25 °C) for two weeks to constant weight. The aqueous extracts were prepared by boiling 100 mg of plant material in 1000 ml of water. The hydro-ethanol extract was obtained from a maceration of 100 g of plant material in 1000 ml of water-ethanol (70:30).

The boiled extracts obtained was allowed to cool and then filtered using a 0.23 mm diameter filter. The filtrate is then left on the bench for 24 h and the supernatant dried in a lyophilizer.

The macerate obtained is filtered on hydrophilic cotton and then on Wattman No. 1 paper, then the filtrate is dried in an oven at 70 °C for 48 h. The dried extracts obtained were weighed and stored in the refrigerator at 4 °C.

As indicated in an ethnobotanical study, *Alchornea cordifolia* and *Antrocaryon klaineum* are the most plants used in the treatment of infectious diseases in four cities of Cameroon as reported by Ndjib et al. [23]. For the preparation of the recipe solutions from the obtained plant extracts, the same amount of mass of each extract is weighed and solubilized in sterile distilled water to obtain a final concentration of 100 mg/ml [24].

Determination of the total phenols content by the Folin-Ciocalteu method

The total polyphenols were evaluated according to the spectrophotometric method using the Folin-Ciocalteu reagent as described by Chew et al. [25]. In this method, 1817 µl of distilled water was introduced into the test tube, 115 µl of Folin Ciocalteu diluted 1:10 and 345 µl of 15% sodium carbonate (Na₂CO₃) were added. Tubes were vortexed and incubated for 2 h, and the absorbance measured at 765 nm. Calibration was performed using a freshly prepared aqueous solution of Ascorbic acid.

DPPH radical scavenging test

The molecule of 1,1-diphenyl-2-picrylhydrazyl (DPPH) is characterized as a stable free radical by virtue of the delocalization of the spare electron over the molecule as a whole, so that the molecule does not dimerize, as would be the case with most other free radicals. To prepare a standard solution of 1,1-diphenyl-2-picrylhydrazyl (DPPH[•]), 10 mg of DPPH was dissolved in 25 ml of methanol [26]. From this solution, 5 ml was taken and mixed with 45 ml of methanol. After preparing the different solutions, 1950 µl of the DPPH solution was pipetted into test tubes and 50 µl of each extract at different concentrations was then added to each test tube to a final volume of 2 ml per tube. All tests were carried out in triplicate in a dark room. The optical density was measured at a wavelength of 515 nm using a spectrophotometer of the brand Jenway 6305, Germany, after 120 min of incubation.

Kinetic study, development of study conditions

At the beginning of this evaluation, a kinetic study of the reactivity of DPPH with the different extracts recipe was carried out. It consisted in monitoring the reactivity of the DPPH over time by measuring the absorbance every 15 min for 120 min [27].

β-carotene bleaching coupled the auto-oxidation of linoleic acid assay

The reductive capacity of extracts was estimated by the method [27] as described previously by Moure et al. [28] and Djova et al. [29].

Anti-inflammatory assays

Bovine serum albumin (BSA) denaturation assay

Protein denaturation was performed as described by Djova et al. [30].

Proteinase inhibitory action

The anti-proteinase activity of ASE was determined using the Oyedepo and Famurewa method [31] with slight modifications. The reaction mixture (2 ml) was containing 0.06 mg trypsin, 1 ml of 20 mmol Tris HCl buffer (pH 7.4) and 1 ml test sample of different concentrations. The reaction mixture was incubated at 37 °C for 5 min and then 1 ml of 5% (W/V) casein was added. The mixture was incubated for an additional 20 min, 2 ml of 70% perchloric acid was added to terminate the reaction. Cloudy suspension was centrifuged and the absorbance of the supernatant was read at 210 nm against buffer as blank. The experiment was performed in triplicate.

Chemical screening

The secondary metabolites present in the plant extracts studied were investigated through several methods that differ in their reagents and solvents as well as in the coloration observed and their presence was noted (+) and their absence (-) [32].

Data analysis

Graphs were obtained using the Microsoft Excel 2013 Spreadsheet. Statistical analyzes were performed in GraphPad/Prism 7 for Windows (San Diego, California, USA) and ANOVA Windows SPSS 23 software. The analysis of the variance (ANOVA) was used to compare the averages between more than two groups. The materiality threshold was set at $p < 0.05$.

RESULTS

Total phenolic content

The evaluation of the total phenolic content in the extracts made it possible to quantify them in two categories of antioxidants as shown in table 1.

Table 1: Total polyphenol content of a mixture of aqueous and hydroethanolic extracts

Extracts	Total phenols (mg EAA/g dw)	Primary antioxidants (mg EAA/g dw)	Secondary antioxidants (mg EAA/g dw)
(Ac ₁ +Ak ₁)	270.8±0.1**	8.33± 0.34*	94.2±2.03*
(Ac ₂ +Ak ₂)	262.41±0.3**	8.49± 0.3*	97.4±4.16*

Legend: (Ac₁+Ak₁): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v); (Ac₂+Ak₂) hydroethanolic extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v). Values that have the same symbols are statistically identical ($p > 0.05$). Statiscaly differences between total polyphenols content of combined aqueous and hydroethanolic extracts of *Alchornea cordifolia* and *Antrocaryon klaineum* (** $p < 0.001$).

In this table, it can be seen that the aqueous extract (270.8±0.1 mg AAE/g dw) composed of extracts of equal proportions of *Alchornea cordifolia* and *Antrocaryon klaneum*, had a higher total phenolic content than the hydroethanolic extract (262.41±0.3 mg AAE/g dw) composed of extracts of same proportion of the same plants. It was also noted that the extracts had a higher amount of preventive antioxidant with the content of 94.2±2.03 mg AAE/g dw and 97.4±4.16 mg AAE/g dw respectively for the combined aqueous extract of *Alchornea cordifolia* and *Antrocaryon klaineum* and combined

hydroethanolic extract of *Alchornea cordifolia* and *Antrocaryon klaineum* than that of the curative antioxidant (8.33± 0.34 mg AAE/g dw and 8.49±0.3 mg AAE/g dw) respectively for the combined aqueous extract of *Alchornea cordifolia* and *Antrocaryon klaineum* and combined hydroethanolic extract of *Alchornea cordifolia* and *Antrocaryon klaineum*.

Kinetics of DPPH scavenging

A kinetic study preceded the antiradical analysis by the DPPH method (fig. 1):

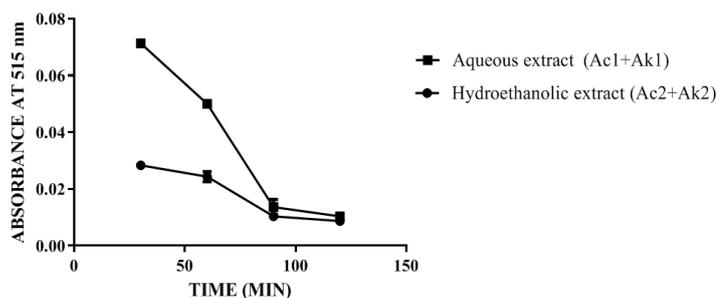


Fig. 1: Absorbance curves of the extracts as a function of time

Legend: (Ac1+Ak1): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v); (Ac2+Ak2) hydroethanolic extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v)

It follows from this curve that the decay of the kinetics curve of the DPPH continues until the 120th min and its stationary state is still not reached.

DPPH antiradical assay

The Ascorbic acid calibration curve for the DPPH test obtained is shown in fig. 2 below:

This curve made it possible to obtain the concentration value for trapping 50% of free radicals (SC_{50}) of Ascorbic acid, which is 0.015 ± 0.000 mg/ml.

The following curve represents the percentage of free radical trapping by the extracts as a function of the concentration (fig.3):

This curve made it possible to obtain the concentrations at which the extracts trap 50% of free radicals (SC_{50}) from which the effective concentrations and the antioxidant power were deduced. The values obtained are recorded in the following table 2.

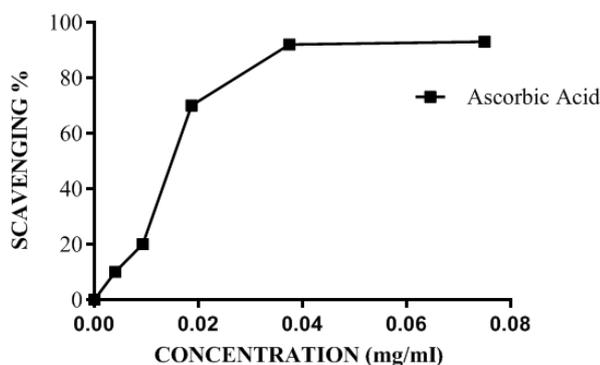


Fig. 2: Ascorbic acid calibration curve for DPPH

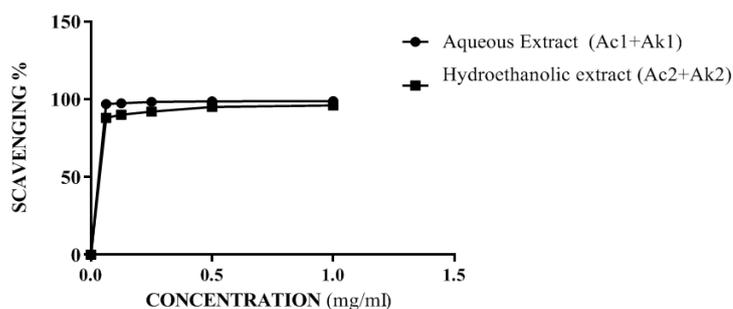


Fig. 3: Curves of the percentages of trapping of free radicals by the extracts, Legend: (Ac₁+Ak₁): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v); (Ac₂+Ak₂) hydroethanolic extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v)

Table 2: Anti-free radical efficacy of DPPH by extracts compared to ascorbic acid

Extracts	SC_{50} en mg/ml	EC_{50}	PA
Aqueous extract (Ac ₁ +Ak ₁)	$0.47 \pm 0.2725^*$	1.175*	0.851*
Hydroethanolic extract (Ac ₂ +Ak ₂)	0.048 ± 0.8143	1.2*	0.83*
Ascorbic Acid	0.015 ± 0.000	0.375	2.66

Legend: (Ac₁+Ak₁): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v); (Ac₂+Ak₂) hydroethanolic extracts *Alchornea cordifolia* and *Antrocaryon klaineum* (v/v). Values that have the same symbols are statistically identical ($p > 0.05$).

The two extracts had substantially the same antiradical activity with statistically identical power of 0.81 and 0.851 respectively for the aqueous and hydroethanolic extracts of *Alchornea cordifolia* and *Antrocaryon klaineinum*, but Ascorbic acid with antiradical power of 2.66 had a antiradical activity greater than that of the two extract plants.

β -carotene bleaching assay

The fig. 4 represents the reducing capacity of the aqueous and hydroethanolic extracts by the β -carotene method

This curve made it possible to obtain the IC₅₀ and reducing power values and the values are recorded in the following table 3:

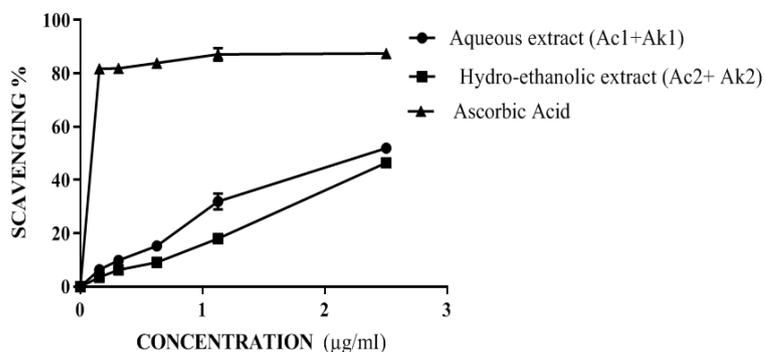


Fig. 4: Curve of the percentages of entrapments as a function of the concentration of the extracts

Legend: (Ac1+Ak1): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineinum* (v/v); (Ac2+Ak2) hydroethanolic extracts *Alchornea cordifolia* and *Antrocaryon klaineinum* (v/v)

Table 3: Reductive capacity of *in vitro* extracts by the β -carotene test

Extracts	IC ₅₀ µg/ml	PR mg EAA/g dw
Aqueous (Ac ₁ +Ak ₁)	2.64±0.11*	94.2±2.03*
Hydroethanolic (Ac ₂ +Ak ₂)	2.86±0.0655*	97.4±4.16*
Ascorbic Acid	0.53±0.0053	

Legend: (Ac1+Ak1): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineinum* (v/v); (Ac2+Ak2) hydroethanolic extracts *Alchornea cordifolia* and *Antrocaryon klaineinum* (v/v). Values that have the same symbols are statistically identical ($p > 0.05$).

Extracts and Ascorbic Acid have statistically different reducing capacities at $p = 0.0001 < 0.005$. Indeed, the reducing capacity of Ascorbic Acid (0.53±0.0053 µg/ml) is higher than that of the two recipes of aqueous and hydroethanolic extracts (between 2.64±0.11 and 2.86±0.0655 µg/ml) respectively. In addition, the reducing capacity of the recipe composed of same proportions of aqueous extracts of *Alchornea cordifolia* and *Antrocaryon klaineinum* (2.64±0.11 µg/ml) is greater than that of the hydroethanolic extracts of the same plants (2.86±0.0655 µg/ml).

Anti-inflammatory assay

The fig. 5 below shows the curve of the percent inhibition as a function of the concentration of the aqueous (*Alchornea cordifolia*

and *Antrocaryon klaineinum*, v/v) and the hydroethanolic (*Alchornea cordifolia* and *Antrocaryon klaineinum*, v/v) extracts compared to the anti-inflammatory reference, Diclofenac®:

The IC₅₀ was determined from the regression line $Y = ax+b$.

The denaturation test of BSA reveals that the two extracts studied have substantially the same activity as Diclofenac at $p < 0.05$ with an IC₅₀ of 50.21 µg/ml.

The fig. 6 shows percentage inhibition of proteinase extracts and acetylsalicylic acid versus concentration.

This curve made it possible to determine the IC₅₀ and the results recorded in the following table 4:

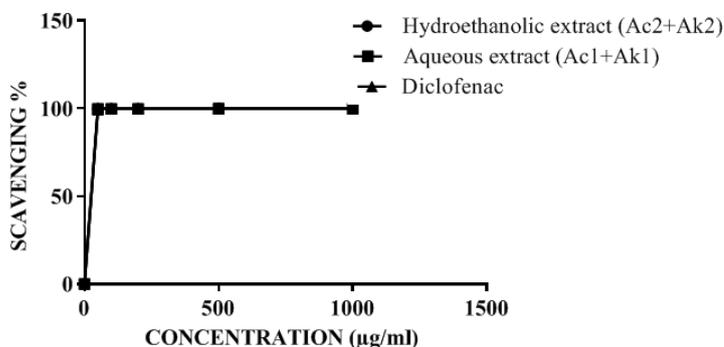


Fig. 5: Calibration curve of the extracts and Diclofenac and capacity to denature the BSA

Legend: (Ac1+Ak1): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineinum* (v/v); (Ac2+Ak2) hydroethanol extracts *Alchornea cordifolia* and *Antrocaryon klaineinum* (v/v)

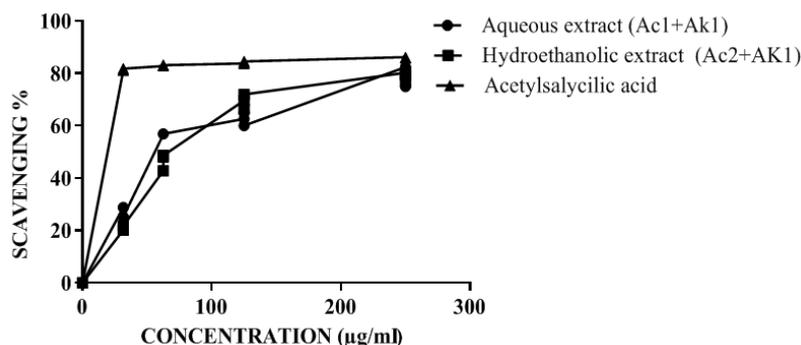


Fig. 6: Curves showing percentages of proteinase inhibition by extracts

Legend: (Ac1+Ak1): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineanum* (v/v); (Ac2+Ak2) hydroethanol extracts *Alchornea cordifolia* and *Antrocaryon klaineanum* (v/v)

Table 4: Percentage inhibition of proteinase by extracts and acetylsalicylic acid

Echantillons	IC ₅₀ en µg/ml
Aqueous Extract (Ac ₁ +Ak ₁)	60±1.52*
Hydroethanolic Extract (Ac ₂ +Ak ₂)	75±2.08*
Ascorbic Acid	10±0.57

Legend: (Ac1+Ak1): aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineanum* (v/v); (Ac2+Ak2) hydroethanol extracts *Alchornea cordifolia* and *Antrocaryon klaineanum* (v/v). Values that have the same symbols are statistically identical ($p > 0.05$).

The IC₅₀ values obtained vary from 10±0.57 µg/ml for Acetylsalicylic acid to 60±1.52 µg/ml for the aqueous extract composed in equal proportions of the aqueous extracts *Alchornea cordifolia* and *Antrocaryon klaineanum* 75±2.08 for the hydroethanolic acid extract composed in equal proportions of the hydroethanolic extracts of *Alchornea cordifolia* and *Antrocaryon klaineanum*. The two extracts

have substantially the same activity ($p > 0.05$), although different from that of acetylsalicylic acid at the $p < 0.05$ threshold.

Chemical screening

Table 5 below shows the families of bioactive compounds present in the extracts studied:

Table 5: Chemical screening

Plant	Extract	Alc.	Anth.	Anthr.	Coum.	Flav.	Gluc.	Phe.	Sapo.	STS	Tan.
<i>A. cord.</i>	Aq.	+	+	+	+	+	+	+	+	+	+
	He	+	+	+	+	+	+	+	+	+	+
<i>A. klain.</i>	Aq.	+	-	-	-	+	+	+	+	+	+
	He	+	-	-	-	+	+	+	+	+	+

Legend: Plant: *A. cord.*: *Alchornea cordifolia*; *A. klain.*: *Antrocaryon klaineanum*; secondary: Secondary metabolites; Anthr.: Anthraquinones; Sapo.: Saponins; STS.: Sterols, Terpenoids and Steroids; Flav.: Flavonoids; Alc.: Alkaloids; Coum.: Coumarines; Phe.: Phenols; Tan.: Tannins; Glucosides. Extract; aq: aqueous; he: hydroethanolic; (+): presence; (-): absence.

Thus, the compound mainly revealed and present in all the extracts are alkaloids, flavonoids, carbohydrates, phenols and tannins.

DISCUSSION

The determination of the amount of total phenols content of the two extracts recipes previously made showed that the aqueous extract (270 mg AAE/g dw) composed extracts of same proportions of *Alchornea cordifolia* and *Antrocaryon klaineanum*, contained more total antioxidants than the hydroethanolic extract (262.41 mg AAE/g dw) composed of extracts of same proportions of the two plants. In addition, both extracts had statistically the same amount of primary antioxidants on the one hand at $p = 0.74$. On the other hand, the aqueous extract (77±1 mg AAE/g dw) was richer than the hydroethanolic extract (9.9±1.595 mg AAE/g dw) as antioxidants secondary to the threshold of significance ($p = 0.0008 < 0.05$).

The preponderance of secondary antioxidants compared with the primary of one of the two extracts studied refers to a preventive role in the treatment of diseases caused by oxidative stress and would justify the use of these plants in the preventive treatment of a pathology that can degenerate into cancer. In several authors

highlight, the presence in large quantities of secondary antioxidants in the ethanolic extracts of leaves and bark of cameronian plants reinforce the use of this plant as indicated in ethnobotany for the treatment of breast cancer [33, 29].

A kinetic study was carried out before the actual test and it was noted that for the two extracts studied, the decay continued until the 120th minute, the stationary state was still not reached and as Nyegue demonstrated in his work on the therapeutic potential of essential oils, such kinetics where the plateau is still not reached in the 120th minute is said to be slow [27]. The reactivity time of the extracts with the DPPH is thus progressive and these extracts could act on the long term on the free radicals at the origin of oxidative stress.

Ascorbic acid SC₅₀ was 0.018±0.001 mg/ml and aqueous and hydroethanolic extracts 0.047±0.2725 and 0.048±0.8143 mg/ml, respectively. In the same time, the aqueous extract alone of *Alchornea cordifolia* reported an EC₅₀ of 0.026 mg/ml [34]; Although the recipe showed the ability to trap free radicals, its antiradical potential is lower than that of the aqueous extract of *Alchornea cordifolia*, this result reinforces the idea that the combination of

plants in traditional medicine has not always the potentiating effect sought for a specific activity.

The reducing power expressed in mg of Ascorbic acid equivalent per g of dry weight of extract was statistically higher in the aqueous extract composed in the same proportions of the aqueous extracts of *Alchornea cordifolia* and *Antrocaryon klaineianum* (70.75±0.34), compared to the reducing power of the hydroethanolic extract composed of hydroethanolic extracts of the same plant species (62.67±0.46) at the significance level $p < 0.05$. Both extracts studied demonstrated a good reducing activity of ferric iron. Nevertheless, the extracts and Ascorbic acid have statistically different reducing capacities at $p = 0.0001$. Indeed, the reducing capacity of Ascorbic Acid (0.53±0.0053 µg/ml) was higher than that of both extracts. Furthermore, the reductive capacity of the aqueous extract (2.64±0.11 µg/ml) composed in same proportions of the aqueous extracts of *Alchornea cordifolia* and *Antrocaryon klaineianum* was greater than that of the hydroethanolic extract (2.86±0.0655 µg/ml) composed in 50: 50 proportions of hydroethanolic extracts of *Alchornea cordifolia* and *Antrocaryon klaineianum*. An ethnobotanical, phytochemical and pharmacological review of *Alchornea cordifolia* has also highlighted its antioxidant properties both *in vitro* and *in vivo* [35], reinforcing its therapeutic anti-infective potential. Agbor et al. [33] Report that the extracts of this plant have a high antioxidant activity because of their ability to reduce iron and to trap free radicals. These results are in harmony with those found in the literature that phenolic compounds, most especially flavonoids, possess a strong reducing power [36].

The denaturation test of Bovine Serum Albumin (BSA) revealed that the two extracts studied had substantially the same activity as Diclofenac® with an IC₅₀ of 50.21 µg/ml. With respect to the antiproteinase assay, the IC₅₀ values obtained ranged from 10±0.57 µg/ml for acetylsalicylic acid to 60±1.52 µg/ml for the aqueous extract composed of 50:50 proportions of extracts aqueous solution of *Alchornea cordifolia* and *Antrocaryon klaineianum* and 75±2.08 for the hydroethanolic extract compound with same proportions of hydroethanolic extracts of the same plants. Both extracts had substantially the same activity, although it was different from that of Acetylsalicylic acid at $p < 0.05$. The methanolic extract of *Alchornea cordifolia* has, in the past, demonstrated anti-inflammatory activity [34].

Inflammation is the main producer of reactive forms of oxygen, so it may be interesting to combine anti-inflammatory treatment with antioxidant treatment [37]. Vitamin A, like β-carotene, is a lipophilic antioxidant whose role is to protect the body against reactive forms of oxygen, and many studies tend to demonstrate their involvement in the regulation of inflammation genes [38]. In humans, the addition of antioxidants seems, to a certain extent, to have preventive or beneficial effects on the evolution of several pathologies [39]. Few studies report the effect of antioxidants in equine medicine; however, it appears that antioxidants can be used beneficially in inflammatory conditions [40]. In addition, as part of the prevention of degenerative diseases of the nervous system in horses, it seems that vitamin A such as β-carotene are unavoidable elements.

The chemical screening carried out on the 5 types of plant extracts gave an insight into the different groups of metabolites found there. Indeed, the compounds present in the aqueous extracts were alkaloids, flavonoids, carbohydrates, phenols and tannins. The extracts of *Alchornea cordifolia* are those containing all ten groups of secondary metabolites sought. This result is the same as those obtained by a large number of studies where *Alchornea cordifolia*, thanks to its wide range of active ingredients, would be used as a panacea in several African countries and for the treatment of diseases of different orders [41, 35]. In contrast to this study, saponins and alkaloids are absent in the aqueous extract of *Alchornea cordifolia* [34].

Several factors have been updated to justify the difference in the chemical composition of extracts from several individuals of the same species; the most incriminated being the place and the period of harvest, the treatment suffered by the harvested organ, the mode of drying, the nature of the solvents and many others. Chemical groups such as flavonoids, polyphenols have been the subject of previous studies and the antioxidant and anti-inflammatory

properties have been recognized; this would justify the use of these plants in the treatment of infectious and degenerative diseases [29].

CONCLUSION

Alchornea cordifolia and *Antrocaryon klaineianum* are two cameroonian plants whose virtues have been studied for each one of them. These plants whose active compounds are quite diversified are used in traditional medicine in the treatment of recurrent tropical diseases. In this study, they were associated with the same concentrations and their estimated antioxidant and anti-inflammatory potential. Thus, prepared in two ways, the extracts of the same plant revealed the presence of the same bioactive compounds. The antioxidant potential evaluated was found to be important, although lower than that of the standard molecule. Regarding the anti-inflammatory potential of the extracts studied, it was identical to that of Diclofenac® thus enhancing their use in the management of inflammatory diseases. The anti-inflammatory activity of these extracts, which is substantially the same as that of Diclofenac® supplemented with their antioxidant potential, would be of therapeutic importance for the management of diseases related to oxidative stress.

ACKNOWLEDGMENT

Authors are very grateful to Cameroon National Herbarium (Yaoundé) for the identification of the plant and to Dr. Mokale Aristide for his technical assistance and advice.

FUNDING

Nil

AUTHORS CONTRIBUTIONS

Ndjib Rosette Christelle, Djova Steve Valdi, Kom Wayoué Christelle and Amina Mamat carried out the experiments. Dibong Siegfried Didier and Agbor Gabriel Agbor designed the study. Nyegue Maximilienne Ascension supervised the work and provided the facilities for the study. All authors read and approved the final manuscript.

CONFLICTS OF INTERESTS

The authors declare that there are no conflicts of interest regarding this paper.

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