Journal of Complementary and Alternative Medical Research

11(3): 1-11, 2020; Article no.JOCAMR.61839
ISSN: 2456-6276

Antioxidant activities of *Dichrostachys cinerea (L.) Wight et Arn* (Leguminosae): Correlations between the Polyphenol Level and the Antioxidant Activity

Reine Raïssa Rolande Aworet Samseny[1*], Line-Edwige Mengome[1] and Sophie Aboughe Angone[1]

[1]Institut de Pharmacopée et de Médecine Traditionnelle, Centre National de la Recherche Scientifique et Technologique Libreville, Gabon.

Authors' contributions

This work was carried out in collaboration among all authors. Authors RRRAS, SAA and LEM performed the experimental studies and drafted the manuscript. Authors RRRAS and SAA played roles in the writing and editing of the manuscript. Author SAA conceived of the study, participated in the design and coordination of the study, supervised the study and revised the manuscript. All authors have read and approved the final manuscript.

Article Information

DOI: 10.9734/JOCAMR/2020/v11i330185
Editor(s):
(1) Dr. Sahdeo Prasad, Texas Tech University Health Sciences Center, USA.
Reviewers:
(1) Bruna da Fonseca Antunes, Universidade Federal de Pelotas, Brasil.
(2) João Andrade, Universidade Federal da Paraíba (UFPB), Brasil.
Complete Peer review History: http://www.sdiarticle4.com/review-history/61839

Original Research Article

Received 02 August 2020
Accepted 08 October 2020
Published 22 October 2020

ABSTRACT

Aims: Free radicals are involved in many diseases. Antioxidants help control them. The overproduction of free radicals or reduction of natural antioxidants promotes the destruction of cells in our body, for example in neurodegenerative diseases. The aim of our study is to evaluate the correlation between the level of polyphenol and antioxidant activity of *Dichrostachys cinerea* barks he work was carried out at the phytochemistry department of the Institute of Pharmacopoeia and Traditional Medicine (Gabon).
Methodology: Different extracts of *Dichrostachys cinerea* barks were obtained with polarity solvent (dichloromethane, ethyl acetate, butanol, methanol and water). The antioxidant activity is evaluating using respectively FRAP method (Ferric Reducing Antioxidant Power), antiradical activity by the method of inhibition of DPPH radical (2.2-diphenyl-1-picrylhydrazyl), cations ABTS radical (2.2'-azinobis-[3-acid-6-sulfonic ethylenzothiazoline]).

**Corresponding author: E-mail: awsams@yahoo.fr;*

Results: Ethyl acetate and dichloromethane extract have the higher value for polyphenolic compounds, respectively 52,27±0,66 mg EAG / g and 49,72±0,55 mg EAG / g. Aqueous extract have the lower value 20,67±1,05 mg EAG / g. The different fractions of *Dichrostachys cinerea* have antioxidant effect, and this effect is in correlation with its constituents: polyphenols, tannins and flavonoids. Results were compared to standard antioxidants such as ascorbic acid and quercetin.
Conclusion: This study confirms higher is the level of polyphenols greater is the antioxidant power.

Keywords: Medicinal plan; phenolic compounds; oxidative stress; tannins; flavonoids.

1. INTRODUCTION

Free radicals are molecules naturally produced by our body. However, it is unstable molecules produced in excess, they degrade the cells accelerate aging. Experts suspect the free cause of many inflammatory disease's radicals. There are also external factors that increase the production of free radicals in our body: alcohol, smoking, pollution, excessive sun exposure, certain medications. Anti-free radical allies are antioxidants. They serve as a defense against free radicals and are found in many foods. Experts recommend to consuming daily and naturally in food rather than supplements. When our body is exceeded, the excess free radicals are compared with antioxidants, this is called oxidative stress. Oxidative stress is the cause of many diseases among which cognitive impairment and dementia. Oxidative stress has been shown to play a pivotal role in the pathophysiology of dementia [1]. In fact, several studies have evaluated the relationship between the levels of some oxidative stress biomarkers and cognitive function. A recent study showed that increased oxidative stress biomarkers correlated with raised levels of inflammatory cytokines and both were associated with low cognitive performance in institutionalized elderly people [2]. Oxidative stress is also responsible for the formation of cancer. Many studies have demonstrated the direct relationship between chronic inflammation and carcinogenesis [3]. The main chemical effectors of these effects of the inflammatory response are free radical species derived from oxidative stress induced by inflammation It is, therefore, important to make sure provide sufficient dose of antioxidants in our body [4,5,6]. Since it has been established that oxidative stress plays an important role in the pathogenesis of many clinical conditions and aging, several studies have been conducted to investigate the therapeutic effects of antioxidant therapy. Vitamins, selenium and polyphenols are among the compounds used as antioxidants. The search for phytochemicals with potent antioxidant continues to be of great importance in the search for remedies against free radical-mediated diseases, prevention of oxidative reactions in foods, protection against DNA damage and carcinogenesis, and possible substances with wide range of pharmacological activities such as anti-inflammatory, antibacterial, and antifungal properties. During our study we will be interested in the antioxidant properties of polyphenols, more particularly polyphenols contained in a plant widely used in traditional medicine in Gabon : *Dichrostachys cinerea*. It is a plant whose bark is widely used in Gabon to treat various pathologies. *Dichrostachys cinerea*, leguminosae, is a shrub or small tree up to eight feet tall, with branches ending in thorns [7]. The *Dichrostachys* are widespread species of the Sudanian Sahel regions of Guinea, they grow on heavy soils, and invading disturbed areas (fallow land, road embankments). They are common, locally abundant. They are found from Senegal to Cameroon, to Gabon to Sudan. It's used as herbal medicine to treat diseases like: kidney troubles, gonorrhea, cough, measles, rheumatism, asthma and dental caries. *Dichrostachys cinerea* barks have been reported to be used by population to facilitate childbirth [8]. The different extracts of *Dichrostachys cinerea* have shown an excellent anti-inflammatory activity [9]. *Dichrostachys cinerea* protects guinea pigs from anaphylactic shock caused by the histamine inhalation [10]. The plant extract has shown tracheal relaxation in guinea pig [11] and mouse [12]. It as shown also antibacterial properties [13], and have hepatoprotective activity [13]. The plant contains flavonoids and tannins [14]. Flavonoids are part of the polyphenol family. They are present in plants and are mainly used to give color to plants. Flavonoids are recognized like antitumor, anti viral, anti allergy, anti-inflammatory [15] and vascular protective more antiseptic or antibacterial. Antioxidants are documented in several publications to mitigate inflammatory processes and this plant activity has been attributed to the phenolic compounds it contains,

particulary the flavonoids [16,17,18,19]. This justifie the choice of our study on the antioxidant potential of *Dichrostachys cinerea* and to establish a correlation between the phenolic compounds and the antioxidant activity.

2. MATERIALS AND METHODS

2.1 Plant Material

The bark of *Dichrostachys cinerea* were collected at Essassa (December 2019, rain season) in Ntoum, Gabon. Plant materiel was authenticated by a botanist of Gabon National Herbarium (IPHAMETRA/CENAREST). A voucher specimen (H.P Bouroubou 387, M.S.M Sosef n°: 894, M.SM 1097) were deposed in this department. After the harvest, the plants are dried at room temperature (30-40) on drying racks in a dedicated room, for 30 days (time necessary to obtain a water content of less than 15 %). After drying, the plants are immediately crushed and stored in jars until their extractions. After this step they are kept in vials, and put in a desiccator.

2.2 Chemicals and Reagents

The chemicals and reagents used were: Acetone (PubChem CID: 180), Butanol (PubChem CID: 263), Dichloromethane (PubChem CID: 6344), Ethyl acetate (PubChem CID: 8857), petrol benzene, methanol (Sigma Aldrich, Germany), DDPH (2,2-diphenyl-1-picrylhydrazyl hydrate), ABTS (acid 2,2-azimo-bis 3-ethylbenzothiazolin-6-sulfonic). Folin–Ciocalteu reagents, potassium hexacyanoferrate [K_3 Fe (CN) $_6$]. Quercetin hydrate (PubChem CID: 5280343), Ascorbic Acid PubChem CID: 54670067). NaNO2, NaOH, $FeCl_3$, AlCl3.

2.3 Fractionation of Plant Materiel

Dichrostachys cinerea barks are cut into small piece and dried at room temperature for one month. Thereafter, the barks were crushed into powered using a crusher (Retsch SK 100 Confort Geissen Germany). 100g of the powered is defatted with petrol benzene, filtered and dried. The powered is taken with distillated water, and evaporated in a rotary evaporator (Buchi) to obtain the aqueous fraction. The same thing is doing with the methanol to obtain the methanolic fraction. In a second time 50g of powered were macerated for 24h with acetone/water (80:20). After filtration acetone is removing in a rotavapor at 40°C. The aqueous extract is taken with butanol and shakes in a funnel. Then, the butanolic fraction is evaporated in a rotavapor at 40°C to obtain the butanolic fraction. After the residue is shaken again with dichloromethane, and evaporated to obtain the dichloromethane fraction. Finally, the last residue is shaken with ethyl acetate and evaporated to obtain the ethyl acetate fraction. We thus obtain 5 extracts which we call extracts of *Dichrostachys cinerea* (EDiCi).

2.4 Determination of Polyphenolic Compounds

2.4.1 Determination of total polyphenols

The total phenolic content was determined using Folin–Ciocalteu reagents by the method of Li et al, [20] with analytical grade gallic acid as the standard. 1 ml of extract or standard solution (0–500 mg/l) was added to distilled water (10 ml) and Folin–Ciocalteu phenol reagents (1.0 ml). After five minutes, 20% sodium carbonate (2.0 ml) was added to the mixture. After being kept in total darkness for 1 h, the absorbance was measured at 760 nm using a spectrophotometer (CECIL 2041 Series UV/VIS, England). Amounts of total phenolics were calculated using gallic acid calibration curve. The results were expressed as gallic acid equivalents (GAE) g/g of dry plant matter.

2.4.2 Determination of total flavonoids

A modified method [21] was used for this part: diluted solution (1 ml) containing flavonoids, 5% (w/w) NaNO2 (0.7 ml) and 30% (v/v) ethanol (10 ml) were mixed for 5 min, and then 10% AlCl3 (w/w, 0.7 ml) was added and mixed altogether. Six minutes later, 1 mol/L NaOH (5 ml) was added. The solution was then diluted to 25 ml with 30% (v/v) ethanol. After standing for 10 min, the absorbance of the solution was measured at 415 nm with a spectrophotometer. A standard curve was plotted using quercetin as a standard. Different concentrations of quercetin were prepared in 80% ethanol and their absorbance was read at 415 nm using a spectrophotometer (CECIL 2041 Series UV / VIS, England). The results were expressed in mg quercetin/g dry weight by comparison with the quercetin standard curve, which was made under the same condition.

2.4.3 Determination of total tannins

This assay was performed according to the method proposed by the [22]. Briefly 1 ml of

extract to be assayed is mixed with 5 ml of water vortex which is added 1 ml of ferric ammonium citrate (28% Fe, 3.5 g / l) (circa 24 h) and 1 ml ammonia (8 g / l). The absorbance of the solution is measured at 525 nm after 10 min against a blank (1 ml of extract dose + 6 ml of water + 1 ml of ammonia) for three readings. The tannic acid was used as standard to plot the calibration curve.

2.5 Antioxidant Activities

2.5.1 Reducing powered with FRAP method

The FRAP method (Ferric Reducing Antioxidant Power) is based on the ability of extracts to reduce ferric ion (Fe^{3+}) by ferrous ion (Fe^{2+}). Total antioxidant capacity of each plant extract was determined by the method of Benzie and Strain [23]. And 1 ml of an aqueous solution of each extract (10 mg / ml diluted 100th for 0.1 mg / ml), ascorbic acid, was mixed with 2.5 ml of phosphate buffer (0.2 M, pH 6.6) and 2.5 ml of the aqueous solution (1%) of potassium hexacyanoferrate [$K_3Fe(CN)_6$]. After 30 min of incubation at 50 ° C, 2.5 ml of trichloroacetic acid (10%) was added. The mixture was then centrifuged at 3000 rpm / min for 10 min. 2.5 ml of the supernatant were then mixed with the same volume of water and 0.5 ml of a freshly prepared aqueous solution of $FeCl_3$ (0.1%) was added. The absorbances were read at 700 nm against a calibration curve obtained from ascorbic acid (0-100 mg / l). The reducing power is expressed in equivalent ascorbic acid (EAA) (mmol ascorbic acid / g of dry extract).

2.5.2 Antiradical activity by the method of inhibition of DPPH radical

The antiradical activity of plant extracts reflects their ability to scavenge free radicals from the body. The radical scavenging activity was evaluated on the different fractions of *Dichrostachys cinerea*. The extracts were dissolved in methanol to obtain concentrations of mothers of 10 mg / ml. This concentration is diluted to 100 for the second test. The method spectrophotometric 2,2-diphenyl-1-picrylhydrazyl (DPPH) as described by Popovici et al [24] is used with some modifications. Introduce 1.5 ml of a methanol solution of DPPH at 20mg / l in test tubes containing 0.75 ml of extracts prior to test. A control containing no plant extract is also prepared. The absorbance was read at 517 nm against a calibration curve obtained from ascorbic acid (0-200 mg / l). Each test was performed in triplicate. Antiradical power was expressed as equivalents of ascorbic acid (EAA) (mmol ascorbic acid / g of dry extract). The concentration of reducing compounds (antioxidants) in the extract is expressed in mmol ascorbic acid equivalent (ABE) / g of dry extract. We considered criteria of Scherer and Godoy [25] according to which plant extracts show poor antioxidant activity when AAI < 0.5, moderate antioxidant activity when AAI between 0.5 and 1.0, strong antioxidant activity when AAI between 1.0 and 2.0, and very strong when AAI > 2.0

2.5.3 Antiradical activity by the method of inhibition of the radical cation ABTS

The method described by Re et al [26] is used. It is based on the discoloration of a stable radical cation, $ABTS^+$ (2,2 '-azinobis-[3-acid-6-sulfonic ethylenzothiazoline]) to ABTS in the presence of antioxidant compounds at 734 nm. The radical cation $ABTS^+$ was generated by reacting an aqueous solution of ABTS (7 mM) with 2.5 mM potassium persulfate (final concentration), the mixture is kept in the dark at room temperature for 12 hours before use. The mixture was diluted with ethanol to give an absorbance of 0.70 ± 0.02 to 734 nm using the spectrophotometer. For each extract, a methanol solution (10 mg / ml) is diluted to 100th in ethanol µl of sample and 10 (solution), the reference substance (ascorbic acid) were mixed with 990 µl of fresh solution of $ABTS^+$. The set is stored away from light for 15 minutes and absorbances were read at 734 nm in a spectrophotometer against a standard curve of ascorbic acid exactly 6 min after initial mixing. The concentration of compounds having a reducing effect on the radical cation $ABTS^+$ is expressed in mmol ascorbic acid equivalent (ABE) / g of dry extract. The concentration of reducing compounds (antioxidants) in the extract is expressed in mmol ascorbic acid equivalent (ABE) / g of dry extract.

2.6 Statistical Analysis

Statistical analysis was performed with GraphPad Instat software (Microsoft California, USA), GraphPad Prism 4 (Microsoft, USA). Variance *P* is considered significant when it is less than 0.005 ($P < 0.005$).

3. RESULTS

3.1 Total Phenolic, Flavonoids and Tannins

We conducted assays on five extracts obtained with *Dichrostachys cinerea* barks (EDiCi). These

assays confirm us about the presence of phenolic chemical group. So apart from the aqueous extract which has a low rate of total phenols (20.67 ± 1.05 mg EAG / g), methanolic, butanolic, ethyl acetate and dichloromethane extract, have respectively values ranging from 42.62 ± 0.69 mg EAG / g, 45.51 ± 1.03 mg EAG/ g, 49.72 ± 0.55 mg EAG / g to 52.27 ± 0.66 mg EAG / g (Table 1). Regarding the total flavonoids, aqueous and methanolic extracts have low levels (2.38 ± 0.30 mg EQ / g and 4.63 ± 0.45 mg EQ / g; butanolic extract value is 5.02 ± 0.02 mg EQ / g; ethyl acetate extract value is 6.83 ± 0.27 mg EQ / g and dichloromethane extract value is 6.36 ± 0.67 mg EQ / g (Table 1). Like other chemicals, total tannins are present in all extracts, but only at a low rate for the aqueous extract (14.63 ± 0.67 mg EAT / g); 36.04 ± 1.00 mg EAT / g for methanolic extract, 36.20 ± 0.14 mg EAT for butanolic extract, 44.63 ± 0.12 mg EAT for ethyl acetate and 40,41±1,05 for dichloromethane extract (Table 1).

3.2 Antioxidant Activity of Extracts

The study of the antioxidant activity of our extracts showed that the DPPH method for antioxidant power of extracts is similar to that of quercetin (Table 2) except for the aqueous extract (4.86 ± 0.01 mmol EAA/g). The values obtained with quercetin (13.76 ± 0.26 mmol EAA / g) compared to the five extract types showed a similarity value (EDiCi dichloromethane: 12.40 ± 12; EDiCi ethyl acetate: 12.43 ± 0.08; EDiCi methanol: 11.17 ± 0.67 mmol EAA/g). ANOVA indicated after Dunett multiple comparison test that the values obtained in this experiment are identical except that one obtained with the aqueous extract was significantly different. By the so-called ABTS method, with ascorbic acid as reference substance, ANOVA indicates that the reference value of ascorbic acid (1.78 ± 0.58) is significantly different from other values 3.78 ± 0.37 for EDiCi dichloromethane, 3.36 ± 0.12 for EDiCi ethyl acetate, 3.00 ± 0.37 for EDiCi butanol, 2.12 ± 0.07 for EDiCi methanol, and 0.64 ± 0.02 the lowest value aqueous EDiCi (Table 2). With the reducing power by the FRAP method whose reference substance ascorbic acid, ANOVA indicates that the value of the reducing power of ascorbic acid (5.86 ± 0.51) is significantly different from that of other extracts : 2.57 ± 0.55 for EDiCi dichloromethane, 2.31±0.04 for EDiCi ethyl acetate, 2.44 ± 0.07 for EDiCi butanol, 1.82 ± 0.01 for EDiCi methanol and 0.40 ± 0.09 for the aqueous extract which has a very low reducing power.

Table 1. Total phenolics, flavonoids and tannins contents of different extracts of *Dichrostachys cinerea* root bark

Type extract	Total phenolics mg EAG / g	Total flavonoids mg EQ / g	Total tannins mg EAT / g
EDiCi dichloromethane	49.72±0,55	6.36±0,67	40.41±1,05
EDiCi ethyl acetate	52.27±0.66	6.83±0.27	44.63±0.12
EDiCi butanol	45.51±1.03	5.02±0.02	36.20±0.14
EDiCi methanol	42.62±0.69	4.63±0.45	36.04±1.00
EDiCi aqueous	20.67±1.05**	2.38±0.30**	14.63±0.67**

*** P <0.01: the difference is very significant when compared with reference substance*
** P <0.05: the difference is significant when compared with reference substance*
Nd : not determined, Mean ± S.E.M = Mean values ± Standard error of means of three experiments

Table 2. Evaluation of the antioxidant activity of extracts *Dichrostachys cinerea*, quercetin and of ascorbic acid by different methods (EAA mmol / g)

Type extract	DPPH	FRAP	ABTS
EDiCi dichloromethan	12,40±0,12	2,57±0,55**	3,78±0,37**
EDiCi ethyl acetate	12,43±0,08	2,31±0,04**	3,36±0,12*
EDiCi butanol	12,01±2,00	2,44±0,07**	3,00±0,37
EDiCi methanol	11,17±0,67	1,82±0,01**	2,12±0,07
EDiCi aqueous	4,86±0,01**	0,40±0,09**	0,64±0,02
Quercetin	13,76±0,26	Nd	Nd
Ascorbic acid	Nd	5,86±0,51	1,78±0,58

*** P <0.01: the difference is very significant when compared with reference substance*
** P <0.05: the difference is significant when compared with reference substance*
Nd : not determined, Mean ± S.E.M = Mean values ± Standard error of means of three experiments

3.3 Study of the Correlations between the Extract of *Dichrostachys cinerea* and Different Enzymatic Activities

- The study of the correlation (r) between polyphenolic compounds and the anti-radical activity of EDiCi indicates a good correlation between this activity and the presence of total flavonoids (r = 0.836). However, we observe a higher correlation for total tannins and free radical activity (r = 0.954) and even more for total phenolic compounds (r = 0.972). This indicates a good correlation between the total phenol profile and the antioxidant activity of the plant extracts, thus suggesting that the phenolic compounds are indeed responsible for the antioxidant activity of these extracts (Fig. 1).
- The study of the correlation between polyphenolic compounds and the iron reducing power of EDiCi indicates a good correlation between this activity and the presence of total flavonoids (r = 0.821). However, we observed a higher correlation for total tannins (r = 0.881) and the reducing power of iron, and even more for total phenolics (r = 0.932). This indicates a good correlation between the total phenol profile and the reducing power of iron in these extracts. (Fig. 2).
- The study of the correlation between polyphenolic compounds and the antioxidant capacity of EDiCi indicates a good correlation between this activity and the presence of total flavonoids (r = 0.916). However, we observe a stronger correlation for total tannins (r = 0.860) and anti-oxidant capacity and even more for total phenolics (r = 0.913). This indicates that total phenols have a major antioxidant capacity (Fig. 3).

4. DISCUSSION

The study of the antioxidant activity of our extracts showed that the DPPH method for antioxidant power of extracts is similar to that of quercetin (Table 2) except for the aqueous extract. For the FRAP method has reference substance ascorbic acid, ANOVA indicates that the value of antioxidant power of ascorbic acid is significantly different from that of other extracts. This value is two times less than that of ascorbic acid except for the aqueous extract which has a very low antioxidant capacity (0.40 ± 0.09). When the called ABTS method with ascorbic acid as reference substance, ANOVA indicates that the reference value of ascorbic acid is significantly different from other values (Table 2), but extracts have more power, except the aqueous extract. These results demonstrate that our extracts have excellent antioxidant capacity. It is also found

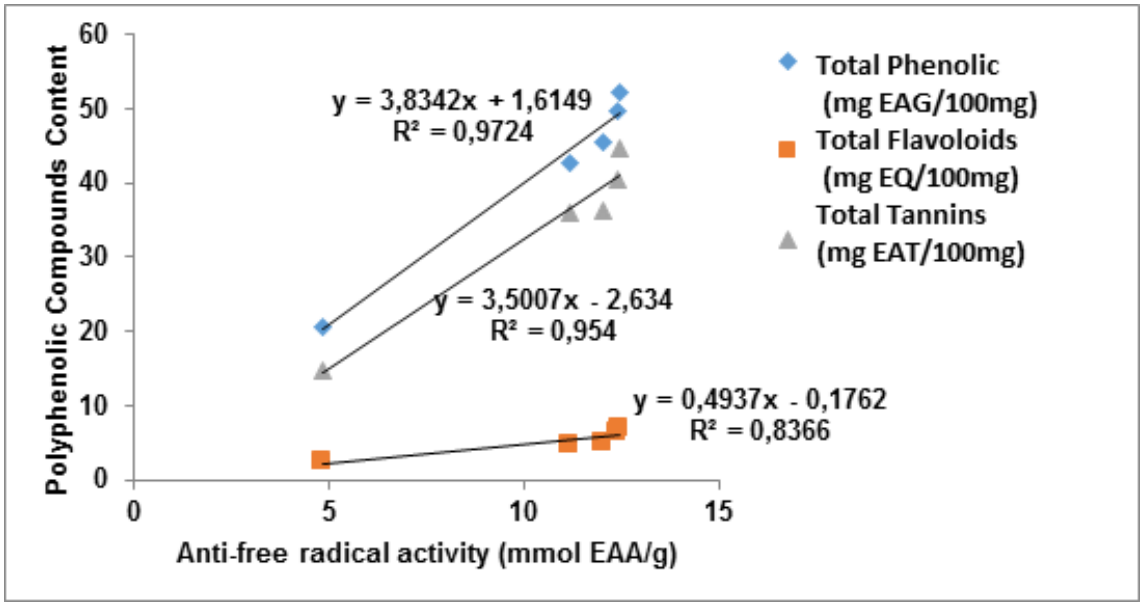

Fig. 1. Graph showing the evolution of the anti-radical activity of the three extracts (total phenolics, total flavonoids, and total tannins) as functions of the contents of polyphenolic compounds

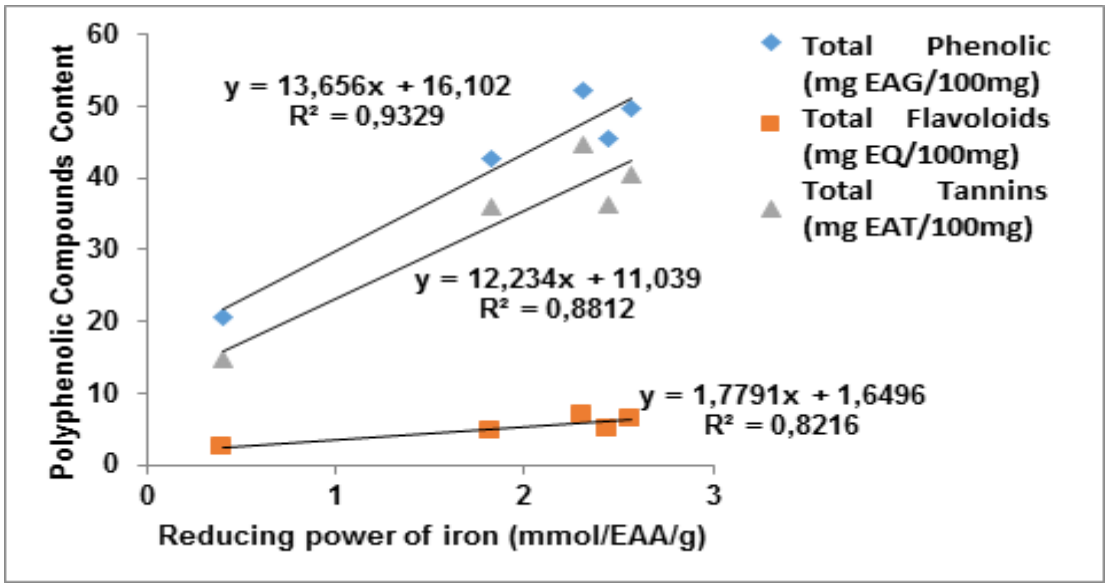

Fig. 2. Graph showing the evolution of the reducing power of iron three extracts (total phenolics, total flavonoids, and total tannins) as functions of the contents of polyphenolic compounds

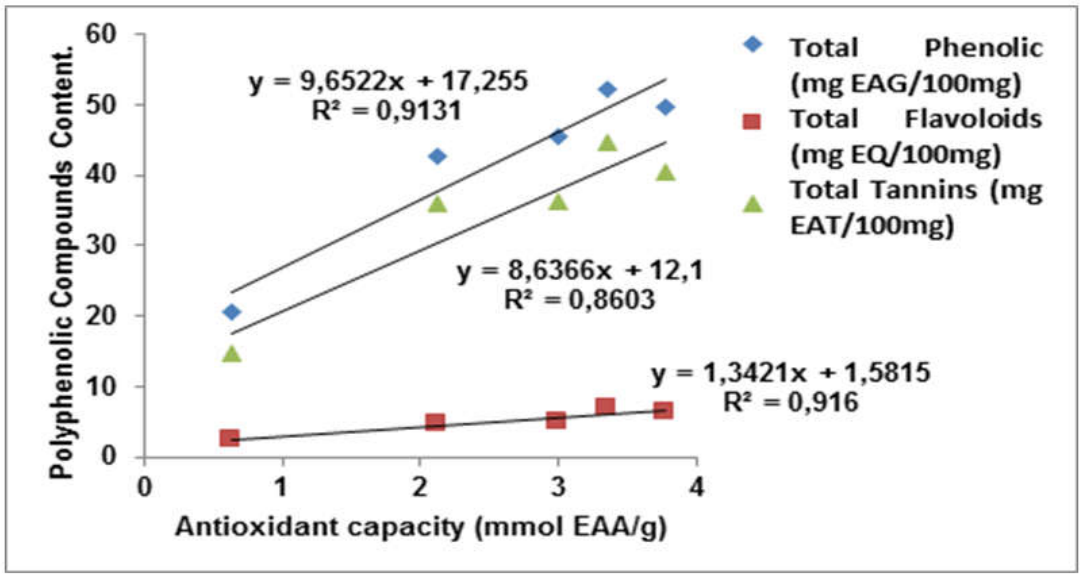

Fig. 3. Graph showing the evolution of the antioxidant capacity of three extracts (total phenolics, total flavonoids, and total tannins) as functions of the contents of polyphenolic compounds

that polar solvents dichloromethane and ethyl acetate, which are richer in phenols, flavonoids and tannins have greater antioxidant activity, which confirms the correlation between phenolic compounds and antioxidant capacity and between phenolic compounds and antiradical activity demonstrated in a large number of studies [27,28]. These results conform to those obtained by Ferrari [29] that highlighted the antioxidant properties of flavonoids and tannins. Polyphenolics compounds are known to have antioxidant activity and it is likely that the activity of the extracts is due to those compounds [30]. This activity is believed to be mainly due to their redox properties, which plays an important role in an adsorbing and neutralizing free radicals, quenching singlet and triplet oxygen, or decomposing peroxides. Phenols or polyphenols

have an antioxidant role, able to neutralize free radicals. Bae et al [31] have shown that free radicals are responsible for many inflammatory diseases. Polyphenols are becoming increasingly important, in particular thanks to their beneficial effects on health [32]. Indeed, their role as natural antioxidants is arousing more and more interest in the prevention and treatment of cancer [33], inflammatory [34], cardiovascular [35] and neurodegenerative diseases [36]. They are also used as additives for the food, pharmaceutical and cosmetic industries [37]. In most extracts studied the antioxidant power is close to that of quercetin [38]. In addition, the polar fractions, dichloromethane and ethyl acetate are richer in phenol, flavonoids and tannins. A positive correlation between antioxidant activity of the plant extracts and their polyphenol content is well established. Flavonoids can prevent injury caused by free radicals in various ways and one way is the direct scavenging of free radicals. Flavonoids are oxidised by radicals, resulting in a more stable, less-reactive radical. In other words, flavonoids stabilise the reactive oxygen species by reacting with the reactive compound of the radical. Because of the high reactivity of the hydroxyl group of the flavonoids, radicals are made inactive [39]. Hanasaki et al [40] found that some of the flavonoids can directly scavenge superoxides, whereas other flavonoids can scavenge the highly reactive oxygen-derived radical called peroxynitrite. Alzheimer's disease (AD) is the most prevalent neurodegenerative disease and most common cause of dementia. However, there is no known way to halt or cure the neurodegenerative disease. Oxidative stress is a cardinal hallmark of the disease and has been considered as therapeutic target for AD treatment. Numerous studies demonstrated that natural polyphenolic compounds protect against various neurotoxic insults in vitro and in vivo AD models. In these studies, dietary polyphenolic compounds exhibit neuroprotective effects through scavenging free radicals and increasing antioxidant capacity. Furthermore, they could facilitate the endogenous antioxidant system by stimulating transcription. Some epidemiological and clinical studies highlighted their therapeutic potential for AD treatment [41]. Irawan et al, [42] in their research of natural antioxidant compounds, show that the inibition of the DPPH radical by their plant extracts compared to ascorbic acid is very important. So the most antioxidant activities against DPPH were displayed by the ethanol extracts of Ficus variegata stem bark, Leucosyke quadrinerva root and Clausena excavata leaves showing 91 %, 91 % and 86 % inhibition at the concentration of 50 ppm. In comparison, the standard ascorbic acid showed 92 % DPPH inhibition in the assay [42]. This is consistent with several reports, including that of Olamide et al [43], whose study revealed that the ethanolic extract of Grewia carpinifolia contains a substantial amount of phenolic compounds which are responsible for its marked antioxidant activity as tested by various in vitro models used in their study. A close relationship between total phenolic content and the antioxidant activity of fruits, plants and vegetables [44,45,46], has also been highlighted. More importantly, various flavonoids have been found to inhibit different targets of coronaviruses SARS and MERS [47], such as blocking the enzymatic activites of viral proteases like 3-chymotrypsin- like protease (3CLpro), papain-like protease (PLpro) and helicase or interfering with spike (S) proteins. A few flavonoids were shown to suppress the activity of angiotensin-converting enzyme (ACE). More recently, another important issue presents the anti-inflammatory potential of flavonoids in viral diseases, such as activating and stimulating the host immune response to viral infections [48], goal also suppressing overwhelming inflammatory reactions, which are often associated with a higher mortality rate of SARS-CoV-2 infections [49]. For instance, some flavonoids have been reported to interfere with the activation of NLRP3 inflammasome [50] which upregulates the production of inflammatory cytokines, and thus can cause respiratory distress syndrome that frequently occurs within SARS coronavirus diseases [51], and SARS-CoV-2 infections [52].

5. CONCLUSION

The study of the antioxidant activity of the extracts has shown that the extracts have excellent antioxidant activity. The determination of total phenolics, flavonoids and tannins carried out with the various extracts reveals that these compounds are predominantly present in the ethyl acetate and dichloromethane extracts. The aqueous extract has the lowest content of phenolic compounds, the lowest antioxidant activity and the lowest reducing power of iron while the ethyl acetate, dichloromethane, butanol and methanol extracts have the best values. The evaluation of the antioxidant activity of our extracts indicates that the dichloromethane and ethyl acetate extracts have a high antioxidant activity compared to the other extracts. These studies show that there is a correlation between total phenolic levels and anti-radical activity,

between total phenolics and antioxidant activity. These results could justify, the use of *Dichrostachys cinerea* extract in traditional medicine in the treatment of inflammatory diseases due to oxidative stress caused by the over production of radical. The screening and characterization of antioxidants derived from natural sources has gained much attention and efforts have been put into identifying compounds as suitable antioxidants to replace synthetic ones. Further investigation on isolation and characterization of bioactive compounds derived from natural extracts is in progress across the world. The outcome of the various works would make it possible to make available on the market new molecules against diseases due to oxidative stress.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

ACKNOWLEDGEMENTS

This work was supported by IPHAMETRA/CENAREST Gabon. All procedures in this study are in accordance with the Gabonese laws and ethical rules.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Chen L, Liu B. Relationships between stress granules, oxidative stress, and neurodegenerative diseases. Oxid Med Cell Longev. 2017;2017:1809592.
2. Baierle M, Nascimento Sn, Moro Am, et al. Relationship between inflammation and oxidative stress and cognitive decline in the institutionalized elderly. Oxid Med Cell Longev. 2015;2015:804198.
3. Khansari N, Shakiba Y, Mahmoudi M. Chronic inflammation and oxidative stress as a major cause of age-related diseases and cancer. Recent Pat Inflamm Allergy Drug Discov. 2009;3(1):73–80.
4. Beliveaud R. Le cancer, une maladie inflammatoire. Le journal de Montréal, votre vie. (23 juin), 2008. Available:https://www.journaldemontreal.com
5. Rock E. Stress oxydant, fruits et légumes & santé. INRA – CRNH. Unité Maladies Métaboliques et Micronutriments Theix/Clermont-Ferrand; 2003.
6. Gibault T. Des interactions bénéfiques entre les antioxydants. Flash: Santé News (APRIFEL). 2000. Available:https://www.aprifel.com.
7. Walker A, Sillans R. Les Plantes utiles du Gabon. 1995;9:241.
8. Aworet Samseny RRR. Aboughe Angone S, Koumba Madingou N, Boukandou Mounanga M, Yao Datté J. Study of pharmacological properties of the methanolic extract of Dichrostachys cinerea bark (L.) Wight et Arn (Leguminosae) in isolated myometrium from pregnant rats. Journal of Ethnopharmacology. 2015;169(1):195-199.
9. Aworet Samseny RRR, Konaté K, Aboughe Angone S, Koumba Madingou N, Yao Datte J. Potential on Anti-Inflammatory Activity and Protection, in Anaphylactic in Conscious Guinea Pig. International Journal of Pharmacognosy and Phytochemistry. 2014;29(1). ISSN: 2051-7858.
10. Aworet Samseny RRR, Souza A, Kpahé F, Konaté K, Yao Datté J. Dichrostachys cinerea (L.) Wight et Arn (Mimosaceae) hydro-alcoholic extract action on the contractility of tracheal smooth muscle isolated from guinea-pig. BMC Complementary and Alternative Medicine. 2011;11:23.
11. Irié-N'guessan G, Champy P, Kouakou-Siransy G, Koffi A, Kablan BJ, Leblais V .Tracheal relaxation of five Ivorian anti-asthmatic plants: Role of epithelium and K^+ channels in the effect of the aqueous-alcoholic extract of Dichrostachys cinerea root bark. Journal of Ethnopharmacology; 2011.
12. Eisa MM, Almagboul AZ, Omer MEA, Elegami AA. Antibacterial activity of Dichrostachys cinera. Fitoterapia. 2000;71:324-327.
13. Babu PS, Krishna V, Maruthi KR, Shankarmurthy K, Babu RK. Evaluation of acute toxicity and hepatoprotective activity of the methanolic extract of Dichrostachys cinerea (Wight and Arn.) leaves. Pharmacognosy Research. 2011;3(1):40.
14. Omukhulu NJ, Mweu MC, Kariuki NP, Wangari MC. Phytochemical characterization, antibacterial screening

and toxicity evaluation of Dichrostachys cinerea. Int J Med Plant Res. 2012;1:32-7.

15. H Hadi M. La quercétine et ses dérivés : molécules à caractère peroxydant ou thérapeutiques. Thèse de doctorat; 2004.

16. Pereira NA, Pereira BMR, DO Nascimento MC, Parente JP, Mors WB. Pharmacological screening of plants recommended by folk medicine as snake venom antidotes; IV. Protection against Jararaca venom by isolated constituents. Planta Med. 1994;60:99-00.
17. Hutt M J, Houghton PJ. A survey from the literature of plants used to treat scorpion stings. J Ethnopharmacol. 1998;60:97-10.
18. Sakai A, Hirano T, Okazai R, Okimoto N, Tanaka K, Nakamura T. Large-dose ascorbic acid administration suppresses the development of arthritis in adjuvant – infected rats. Arch Orthop. Trauma Surg. 1999;119:121-126.
19. Ozgov AS, Hermanek J, Gut I. Different antioxidant effects of polyphenols on lipid peroxidation and hydroxyl radicals in the NADPH-, Fe-ascorbate- Fe-microsomal systems. Biochem. Pharmacol. 2003;66:1127-1137.
20. Li HB, Cheng, KW, Wong CC, Fan KW, Chen F, Jiang Y. Evaluation of antioxidant capacity and total phenolic content of different fractions of selected microalgae. Food chemistry. 2007;102:771-776.
21. Bahorun T, Gressier B, Trotin F, Brunete C, Dine T, Vasseur J, Gazin JC, Pinkas M, Luycky M, Gazin M. Oxygen species scavenging activity of phenolic extracts from hawthorn fresh plant organs and pharmaceutical preparations. Arzneimittel forschung. 1996;46(11):1086-9.
22. Determination of tannins in herbal drugs. 2.8.14. European Pharmacopoeia. Council of Europe (COE) - European Directorate for the Quality of Medicines (EDQM) 5th edition. Strasbourg: Council of Europe. 2005;221.
23. Benzie IFF, Strain JJ. The ferric reducing ability of plasma (FRAP) as a measure of ‘‘antioxidant power”: The FRAP assay. Analytical Biochemistry. 1996;239:70–76.
24. Popovici C, Saykova I, Tylkowski B. Evaluation de l’activité antioxydant des composés phénoliques par la réactivité avec le radical libre DPPH. Revue de Génie Industriel. 2009;4:25-39.
25. Scherer R, Godoy HT. Antioxidant Activity Index (IAA) by the 2,2-diphenyl-1-picrylhydrazyl Method. Food Chemistry. 2009;112:654-658.
26. Re R Pellegrini N, Proteggente A, Pannala A, Yang M, RICE EVANS C. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radical Biology and Medicine. 1999;26:1231-1237.
27. Iacopini P, Baldi M, Storchi P, Sebastiani L. Catechin, epicatechin, quercetin, rutin and resveratrol in red grape: Content, in vitro antioxidant activity and interactions. Journal of Food Composition and Analysis. 2008;21:589-598.
28. Baydar NG, Özkan G, Yaoar S. Evaluation of the antiradical and antioxidant potential of grape extracts. Food Control. 2007;18(9):1131-1136.
29. Ferrari J. Contribution à la connaissance du métabolisme secondaire des Thymelaeaceae et investigation phytochimique de l’une d’elles : Gnidia involucrata Steud.ex A. Rich. Thèse de doctorat. Université de Lausanne. 2002;225.
30. Minker C. Myrtille et autres fruits rouges-Un concentré de bienfaits pour votre santé et votre beauté. Eyrolles. 2011;44.
31. Bae SC, Jung WJ, Lee EJ, Yu R, Sung MK. Effects of antioxidant supplements intervention on the level of plasma inflammatory molecules and disease severity of rheumatoid arthritis patients. Journal of the American College of Nutrition. 2009;28(1):56-62.
32. Wainapel Stanley F, Fast A. Antioxidants and the Free Radical Theory of Degenerative Disease, Alternative Medicine and Rehabilitation. 2003;266.
33. Di C, Kenyon GD, Deborah JK, Aslamuzzaman K, Mohammad B, Lianhai L, Zhigang W, Sheng Biao W, Wai Har L, Tak Hang C, Q Ping D. Green tea and tea polyphenols in cancer prevention, Frontiers in Bioscience. 2004;9:2618-31.
34. Laughton MJ, Evans PJ, Moroney MA, Hoult JR, Halliwell B. Inhibition of mammalian 5-lipoxygenase and cyclo-oxygenase by flavonoids and phenolic dietary additives. Relationship to antioxidant activity and to iron ion-reducing ability, Biochemical. Pharmacology. 1991;42:1673-168.
35. Frankel EN, German JB, Kinsella JE, Parks E, Kanner J. Inhibition of oxidation of human low-density lipoprotein by phenolic

substances in red wine », Lancet. 1993;341:454-457.
36. Orgogozo JM, Dartigues JF, Lafont S, Letenneur L, Commenges D, Salamon R, Renaud S, Breteler MB. Wine consumption and dementia in the elderly: A prospective community study in the Bordeaux area, Revue Neurologique. 1997;153(3):185-92.
37. Third International Conference on Polyphenols applications in Nutrition and Health Satellite Symposium on Natural Antioxidants and Polyphenols : Valorization from Fruit and Vegetable Wastes St Julian. 2006;26-27.
38. Boots AW, Li H, Schins RPF, Duffin R, Heemskerk JWM, Bast A, Haenen GRMM. The quercetin paradox. Toxicology and Applied Pharmacology, Elsevier. 2007;222(1):89-96.
39. Korkina L, Afanasev I. Antioxidant and chelating properties of flavonoids. Adv Pharmacol. 1997;38:151–163.
40. Hanasaki Y, Ogawa S, Fukui S. The correlation between active oxygens scavenging and antioxidative effects of flavonoids. Free Radic Biol Med. 1994;16:845–850.
41. Dong-Young Choi, Young-Jung Lee, Jin Tae Hong, Hwa-Jeong Lee. Antioxidant properties of natural polyphenols and their therapeutic potentials for Alzheimer's disease. Brain Research Bulletin. 2012;87(2–3):144-153
42. Irawan Wijaya Kusuma, Murdiyanto, Enos Tangke Arung, Syafrizal, Yong-ung Kim. Antimicrobial and antioxidant properties of medicinal plants used by the Bentian tribe from Indonesia. Food Science and Human Wellness. 2014;3(3–4):191-196.
43. Olamide E Adebiyi, Funsho O Olayemi, Tan Ning-Hua, Zeng Guang-Zhi. In vitro antioxidant activity, total phenolic and flavonoid contents of ethanol extract of stem and leaf of Grewia carpinifolia. Beni-Suef University Journal of Basic and Applied Sciences. 2017;6:10–14.
44. Albayrak S, Aksoy A, Sagdic O, Hamzaoglu E. Compositions, antioxidant and antimicrobial activities of Helichrysum (Asteraceae) species collected from Turkey. Food Chem. 2010;119:114–122.
45. G. Biju J, Sulaiman CT, Satheesh G, Reddy VRK. Total phenolics and flavonoids in selected medicinal plants from Kerala. Int. J. Pharm. Pharm. Sci. 2014;6(1):406–408.
46. H Baba SA, Malik AS. Determination of total phenolic and flavonoid content, anti-microbial and antioxidant activity of a root extract of Arisaema jacquemonti Blume. J. Taibah Univ. Sci. 2015;9:449–454.
47. Yang Y, Islam MS, Wang J, Li Y, Chen X. Traditional chinese medicine in the treatment of patients infected with 2019-new coronavirus (SARS-CoV-2): a review and perspective. Int J Biol Sci. 2020;16:1708–1717.
48. Dong W, Wei X, Zhang F, Hao J, Huang F, Zhang C, Liang W. A dual character of flavonoids in influenza A virus replication and spread through modulating cell-autonomous immunity by MAPK signaling pathways. Sci Rep. 2014;4:7237.
49. McKee DL, Sternberg A, Stange U, Laufer S, Naujokat C. Candidate drugs against SARS-CoV-2 and COVID-19. Pharmacol Res. 2020;157:104859.
50. Lim H, Min DS, Park H, Kim HP. Flavonoids interfere with NLRP3 inflammasome activation. Toxicol Appl Pharmacol. 2018;355:93–102.
51. Chen IY, Moriyama M, Chang MF, Ichinohe T. Severe acute respiratory syndrome coronavirus viroporin 3a activates the NLRP3 inflammasome. Front Microbiol. 2019;10:50.
52. Shah A. Novel coronavirus-induced NLRP3 inflammasome activation: a potential drug target in the treatment of COVID-19. Front Immunol. 2020;11:1021.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sdiarticle4.com/review-history/61839