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Phytochemical Characterization of Anti Gastric Ulcer Properties of Aqueous Leaf Extract of *Dacryodes edulis* (Plum Leaf)

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Abstract

In Cameroon, the study of traditional medicine and treatment by plants is of particular interest. This country is known for the diversity of its climate, the nature of its soil and the wealth of its medicinal flora. *Dacryodes edulis* (plum) a versatile plant in African ethno medicine is rich in nutrients such as lipids, vitamins and proteins. Its various parts are employed to treat several diseases especially in traditional medicine for the treatment of ailments such as Skin diseases, dysentery, and Leprosy. Generally, the study sought to assess the phytochemical characterisation of anti-gastric Ulcer Properties of aqueous leaf extract of *Dacryodes edulis* and specifically sought to extract the active component of *Dacryodes edulis*, screen aqueous leaf extract of *Dacryodes edulis* for secondary metabolites and to evaluate the antioxidant activity (*in vitro*) of aqueous leaf extract of *Dacryodes edulis*. The research design used was experimentation (*in vitro*). The extraction yield of the extract of *D. edulis* is 10% in milligram obtained by decoction. The current study revealed through phytochemical screening and the study of some antioxidant properties, antiulcer activity and the reactions of characterization to identify the chemical constituents of *Dacryodes edulis* showed that it contains polyphenols; flavonoids and tannins which existed in significant amount ($p < 0.05$). Also, the extract of *Dacryodes edulis* showed a significant radical scavenging capacity effect (IC_{50} 25.03 ± 0.71 µg/ml 2.76 ± 0.45 µg/ml) for DPPH and iron III (Fe^{3+}) respectively indicating that it is good antioxidant agent. These anti-oxidant properties are related to the presence of secondary metabolites detected in the plant. This justifies the traditional usage of the leaves and seeds as remedy for stomach problems and their prevention.

Keywords

Dacryodes edulis, Phytochemicals, Anti-Gastric Ulcer, Traditional Uses

1. Introduction

Plants have been used for several centuries to cure various illnesses and literature has revealed the potential use of many plants in the treatment of gastric ulcers. For this reason, it is important to research the use of medicinal plants with traditional use in the treatment of gastric diseases.

The leaves of *Dacryodes edulis* are used in traditional medicine for the treatment of gastric ulcer among other ailments in many parts of Africa. Plants have been the basis of many traditional medicine systems throughout the world for thousands of years and still remain as the main sources for the development of innovative drugs [1]. Medicinal plants are found worldwide. *Dacryodes edulis*, (plum) tree a versatile plant in African ethno medicine is rich in nutrients such as lipids, vitamins and proteins. Its various parts are employed to treat several diseases especially in traditional medicine for the treatment of ailments such as Skin diseases, dysentery, Leprosy, etc. [2]. The generic name *Dacryodes* is derived from the Greek word Dakruon meaning tear, referring to resin droplets on the bark surface of its members, while edulis means edible, emphasizing the importance of the nutritious fruit in the plant's cultivation [3]. The plant belongs to the family Burseraceae whose members are characterized by an ovary of 2 to 5 cells, prominent as in ducts in the bark, wood and intra staminal disk [4].

In Cameroon, the extracts of 42 medicinal plants used for anaemia, diabetes, AIDS, malaria and obesity were investigated for phytochemical substances and antioxidant properties. The leaves of *Dacryodes edulis* elicited very high antioxidant effect when analyzed against three assay methods: Folin (Folin Ciocalteu Reagent), FRAP (Ferric Reducing Antioxidant Power) and DPPH (1,1-diphenyl-2-picrylhydrazyl), ranking second behind *Alchornea cordifolia* [5]. This antioxidant property was attributed to the presence of flavonoid in the plant. The essential oil of the plant resin also demonstrated good antioxidant activity. In a DPPH test system, the IC50 value of *D. edulis* oil was reported to be 68.5 ± 2.29 $\mu g \cdot mL^{-1}$, while oxidation of linoleic acid was effectively inhibited by the plant (70%) in the β-carotene-linoleic acid test [6].

The Plum tree (*Dacryodes edulis*) is an indigenous fruit tree in the Gulf of Guinea and Central African countries [7], but due to popularity of the nutritious fruit for consumption, the plant is widely cultivated, extending its area of distribution to Sierra Leone, Uganda, Angola, *Dacryodes edulis* is a dioecious shade loving species of non-flooded forests in the humid tropical zone. in the treatment of diverse diseases. The decoction of the bark is taken orally to treat leprosy. It is also used as a gargle and mouth-wash to treat tonsillitis. The bark is comminuted with meleguetta pepper to cure dysentery, anaemia, spitting blood and as an em-

menagogue; when mixed with palm oil, it is applied topically to relieve pains, debility, stiffness and skin diseases. The leaves are chewed with kola nut as an anti-emetic. The leaf sap is used as an ear drop to treat ear trouble while the leaf decoction is prepared to produce vapour that treats fever and headache. In Cameroon, the leaves are boiled with those of Lanata camara, *Cymbopogon citratus* and *Persea americana* in water to form a decoction for treating malaria. A steam bath can also be taken from the decoction to treat the same ailment. Boiling the leaves with those of P. americana alone can be used to treat headache, antalgic and cephalgy [8]. *Dacryodes edulis* is a versatile plant in African ethnomedicine, as its various parts are employed to treat several diseases. The bark of the plant has long been used to cicatrize wound in Gabon. In this case, the bark is pulped and then applied directly to the wound. In Democratic Republic of the Congo, the plant is employed for the treatment of diver's ailments. The decoction of the bark is taken orally to treat leprosy. It is also used as gargle and mouth-wash to treat tonsillitis [9]. The bark is comminuted with meleguetta pepper to cure dysentery, anaemia, spitting blood and as an emmenagogue; when mixed with palm oil, it is applied topically to relieve pains, debility, stiffness and skin diseases. The leaves are chewed with kola nut as an antiemetic. The leaf sap is used as ear drop to treat ear trouble, while a leaf decoction is prepared to produce vapour that treats fever and headache.

1.1. Biological Action of Plum Leaves

The presence of bioactive compounds such as saponins, tannins, alkaloids and flavonoids identified in the plant has been suggested to be responsible for the various uses of *D. edulis* in traditional medicine to cure ringworm, wound, scabies, skin diseases and inflammation [10]. In addition, the potential health-related functions of dietary plants were found to include antibiosis, immunostimulant, nervous system action, detoxification, anti-inflammatory, antigout, antioxidant, glycaemic and hypolipidemic properties [11]. Among the 13 Congolese plants examined for antidrepanocytary activity, the aqueous and ethanol extracts of *D. edulis* leaves were discovered to normalize the SS blood erythrocytes, following the deoxygenation of haemoglobin in anaerobic condition, thus validating their use in traditional medicine [12].

1.2. Peptic Ulcer Disease (PUD)

Peptic ulcer is one of the most common diseases of the gastrointestinal tract (mostly the stomach shown in **Figure 1**, with an increasing incidence and prevalence. Indeed, it can lead to serious complications and even death in some cases. It affects about 10% of the world's population with an estimated mortality rate of 15,000 deaths per year. This condition is characterised by an imbalance between factors that damage and those that protect the integrity of the gastric mucosa. It is multi factor, but the highest incidence is usually seen in the context of Helicobacter pilori infection, and the use of non-steroidal anti-inflammatory drugs [13].

Peptic ulcer, including both gastric and duodenal ulcers, have been a major threat to the world's population over the past two centuries. Chronic inflammation due to colonisation of Helicobacter pylori and nonsteroidal anti-inflammatory drugs account for the large majority of peptic ulcer disease [14]. Peptic ulcer can be delineated as the presence of a deep destruction of the stomach lining or mucosa and/or duodenum, reaching beyond the muscularis mucosa, specifically to the muscle layer owing to the environmental gastric acid synthesis. The two consummate ubiquitous etiological antecedents are the chronic infection with Helicobacter-pylori (Hp) and the use of NSAIDs, involving of course, the ASA. There are distinctive less ubiquitous antecedents that can cause a PU, which are thought-out together, responsible for <5% of cases. Zollinger-Ellison syndrome (ZES) or gastronomy is one amid them which is a neuroendocrine tumor, often located at the head of the pancreas or in the duodenal wall, overactive and gastrin secretory [15].

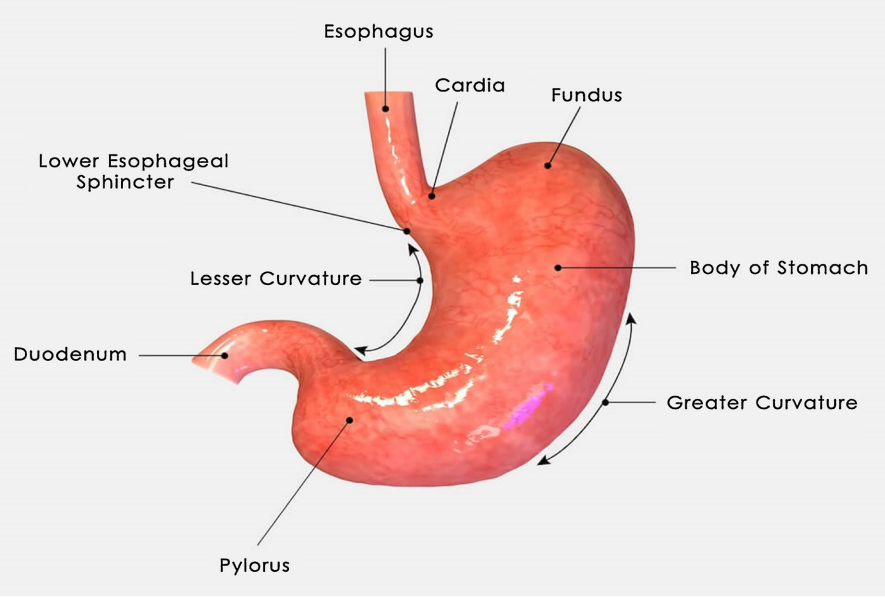

Figure 1. The human stomach anatomy source: [16].

1.3. Analysis of Aqueous Leaf Extract of *Dacryodes edulis*

Percentage composition of moisture, crude fibre, ash, crude fats, crude proteins and carbohydrates were determined for the chloroform and ethanol layers of the chloroform-ethanol extract of the leaves of *D. edulis*). Phytochemical screening (quantitative and qualitative) of aqueous leaf extract of *Dacryodes edulis*. Phytochemical screening of the plant extract was carried out using the methods outlined by Harbone [17]. The phytochemical composition was represented using (+) for presence and (-) for absence.

2. Materials and Methods

2.1. Research Design

This study was carried out using an *in vitro* experimental design. Fresh plum

leaves (*Dacryodes edulis*) were gotten from the farm in Kumba and kept in a dark room to dry. Later it was ground using a manual grinding miller to powder and the ground sample was then taken to the Institute Universitaire de Technology (IUT) science laboratory university of Douala where it was analysed for phytochemical and its antioxidant properties.

2.2. Preparation of the *Dacryodes edulis* Extract

The *Dacryodes edulis* extract was prepared by decoction method. 100 g of dried leaf powder was introduced into a 1000 ml volumetric flask containing a volume of 500 ml of distilled water. The mixture was boiled for 30 minutes using a reflux heating system mounted on a heating mantle. After heating, the mixture was filtered through a filter paper No. 1 filter paper. Then, the decoction was dried at 40˚C in a sand bath for 2 days to obtain the crude extract. The crude extract obtained was weighed and the extraction yield was determined using the formula below:

$$\text{Yield}(\%) = \frac{\text{Mass of extract}(\text{g})}{\text{Mass of the plant matter}(\text{g})} \times 100$$

2.3. Qualitative and Quantitative Analyses on Phytochemical Constituents.

The qualitative analysis of phytochemical constituents was conducted following the methods described by Harbone [17] in a book entitled: "A guide to modern techniques of plant analysis". The quantitative determination of total phenols, total flavonoids, and flavonol content was evaluated using the methods described by Ramde-Tiendrebeogo *et al.* 2012, and Chang *et al.* 2002 [18] [19]. These phytochemical analyses were based on the appearance of different colours and the formation of precipitates or products in the final solution.

2.3.1. Analysis of Polyphenols.

For phenol determination, 0.1 gram of the aqueous extract was solubilized in 3 mL of ethanol. The solution was treated with 3 drops of iron chloride III 10% (V/V), and the appearance of a blue-violet color indicated the presence of polyphenols.

2.3.2. Analysis of Flavonoids

Few drops of 1% NH_3 solution was added to 0.1 gram of the aqueous extract contained in a test tube. The appearance of a yellow color indicated the presence of flavonoid compounds

2.3.3. Analysis of Tannins

For tannins identification, 0.1 gram of the aqueous extract was boiled in 20 mL of distilled water in a test tube and then filtered. The filtration method used here is the normal method, which includes a conical flask and filter paper. Three drops of 0.1% $FeCl_2$ were added to the filtered samples and observed for a brownish green or a blue-black colour, which indicates the presence of tannins.

2.3.4. Determination of Total Polyphenols

10 mg of extract was diluted in 1 ml of methanol, then 100 µl of the solution was diluted in 900 µl of distilled water and 1 ml of Folin's reagent was added. After incubation for 30 min, the solution was saturated with sodium carbonate and the optical densities were read at 650 nm against a blank consisting of 1 ml of Folin's reagent in 1 ml of distilled water.

A calibration curve established from a gallic acid dilution series (10.00, 3.33, 1.11, 0.37, 0.12 mg/ml) was treated in the same way as the extracts. The results were expressed in milligram equivalent of gallic acid per gram of extract. The polyphenol content was obtained from the regression line of the gallic acid calibration curve.

2.3.5. Determination of Flavonoids

We proceeded to stabilize tannins with BSA (Bovine Serum Albumin) Thus, in each extract, 30 ml were taken and 3 g of BSA were added to it, then the solutions obtained were stirred and then filtered with cotton. In the filtrates obtained, the flavonoids were assayed according to the same protocol described above for the polyphenols.

2.3.6. Determination of Tannins

The tannin content was obtained by taking the difference of total polyphenols and flavonoids (non-tannin polyphenol), that is total polyphenols.

2.3.7. Antioxidant Activity (*in Vitro*) of aqueous Leaf Extract of *Dacryodes edulis*

The method used for the extraction of plant materials. *D. edulis* leaf and were washed, chopped into pieces and dried at room temperature for about 14 days to a constant weight. The samples were coarsely powdered, each of the coarsely powdered samples was weighed, placed in a big glass jar and extracted by maceration with 80% ethanol for 72 hours (h) at room temperature. Also, wet samples were extracted by maceration with water for 8 h. Figure 2 represents the flow process as described above.

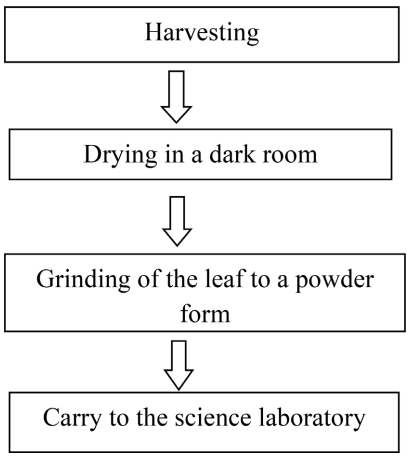

Figure 2. Flow chart for plum leaf.

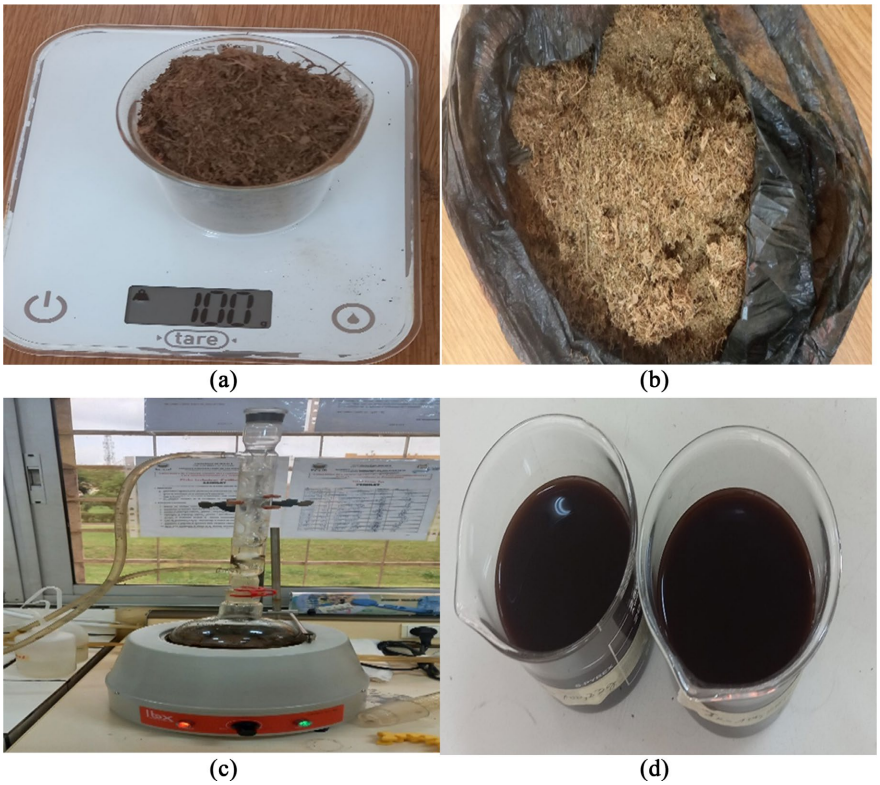

Figure 3. Various stages in the processing of *D. edulis* extract Redox activity of iron III (Fe^{3+}) ions.

The Fe^{3+} reduction test was performed according to the procedure described by Berker *et al.* 2007 [20] with some modifications. The various stages of the extraction process are shown in **Figures 3(a)-(d)** respectively. **Principle**: this method is based on the ability of a substance to reduce Fe^{3+} ions to Fe^{2+} ions which, in the presence of 1,10-phenanthroline, forms a red-orange complex whose optical density can be measured at 505 nm. The colour intensity is proportional to the quantity of Fe^{3+} ions converted by the extract.

2.3. Method

2.3.1. Preparation of Solutions

- **Fe^{3+} solution**

This was prepared to a concentration of 1.2 mg/ml. To do this, 1.2 mg of $FeCl_3$ powder was dissolved in 1 ml of distilled water so as to obtain 100 µg of Fe in 120 µl of the collected solution, which was then diluted by the extract solution to 50 µg.

- **Ortho-Phenanthroline solution**

Ortho-Phenanthroline solution prepared at 0.2% was used to measure the amount of Fe^{2+} formed. To do this, 200 mg of powder was weighed for every 100 ml of ethanol.

- **Extract stock solution**

The extract stock solution was prepared at a concentration of 1 mg/ml. To do this, 10 mg of *D. edulis* extract was weighed and dissolved in 10 ml of water. In a

96-well plate template, 120 µl of water was introduced into all the wells except those of the first line (A) where 240 µl of each extract was introduced, and a series of dilutions of the order 2 was carried out. Thereafter 120 µl of Fe^{3+} solution was added and the plates were incubated for 15 min at room temperature. After this incubation, 60 µl of the ortho-Phenantroline solution was added and the plates were re-incubated for 15 min, still at room temperature. At the end of this incubation, the optical density of the content of the wells was read at 505 nm with a plate reader (Opus). The test was performed twice. The extract's blank solution consisted of the extract, ortho-Phenanthroline and distilled water in place of the Fe^{3+} solution, treated under the same conditions. The solvent blank solution consisted of 120 µl distilled water and 180 µl ethanol. The wells containing 120 µl solvent, 120 µl ethanol and 60 µl of ortho-phenanthroline served as an absorbance control for ortho-Phenantroline. The negative control corresponding to 0% reduction consisted of 120 µl solvent, 120 µl Fe^{3+}, 60 µl ortho-phenanthroline and the positive control corresponding to 100% reduction was considered for a maximum optical density of 4. The following formula was used to calculate the reduction percentages of Fe^{3+}.

$$\%RFe^{3+} = \left(\frac{\text{OD Test}}{4} \times 100\right)$$

2.3.2. To Evaluate the Antioxidant Activity (*in Vitro*) of Aqueous Leaf Extract of *Dacryodes edulis*

The method is based on the capacity of the antioxidant (AH) contained in the sample to donate a single electron to the synthetic radical DPPH of violet colour to stabilize it to DPPH of pale-yellow colour.

- The DPPH solution was prepared by completely dissolving 20 mg of DPPH in 100 ml of 95˚ ethanol. It was stored away from light before use.
- Stock solution of extracts: the stock solution of extracts was prepared to a concentration of 1 mg/ml. 10 mg of each extract were weighed and dissolved in 10 ml of solvent.

In a 96-well plate template, 50 µl of methanol was introduced into all the wells except those of the first line where 100 µl of each extract or gallic acid were introduced, and a series of dilution of order 2 was carried out ranging from line 1 to line 11. Line 12 constituted the negative control. Then, 150 µl of an ethanolic solution of DPPH was added to all the wells of the microplate. After 30 min of incubation in the dark and at room temperature, the absorbance was measured with a spectrophotometer at 517 nm. Each test was performed twice. The measurement of the optical densities of the different tubes enabled us to calculate the percentage trapping of free radicals of each range of concentration of extracts and of gallic acid. The trapping percentage (SC: scavenging concentration) was calculated using the following formula:

$$SC = \frac{\text{Aref} - \text{Ames}}{\text{Aref} - \text{A100}} \times 100$$

Aref = Absorbance at t = 60 min of the negative control solution (DPPH solu-

tion + methanol)

Ames = Absorbance at t = 60 min of the DPPH solution containing the anti-radical.

A100 = Absorbance at the end of the reaction (close to 0) for total entrapment.

The data obtained were subjected to one-way analysis of variance (ANOVA). One – way ANOVA is used to determine whether there are statistically significant differences between the means of three or more independent variables. This helps us find out if the independent variables had a significant effect on the dependent variable. Significant differences are observed at $p < 0.05$. The results are expressed as means ± standard errors of means (SEM). The analysis was done using the computer software known as Statistical Product and Service Solutions (SPSS), version 21.

3. Results

3.1. Extraction of the Active Component of *Dacryodes edulis*

The extraction yield of the extract of *D. edulis* obtained by decoction was determined by relating the mass of the extract to the mass of leaves initially extracted. The ratio calculation showed that the extraction yield is 10% in milligrams. To screen aqueous leaf extract of *Dacryodes edulis* for secondary metabolites, Preliminary phytochemical screening for secondary metabolites of *Dacryodes edulis* of the 10% extract showed the presence of polyphenols, flavonoids and tannins. The concentrations were determined using optical density techniques and the values present in mean ± standard deviation.

3.2. Quantitative Determination of Polyphenols, Flavonoids and Tannins

Figure 4 below shows the variation curve of the optical density values obtained at different concentrations of Gallic acid.

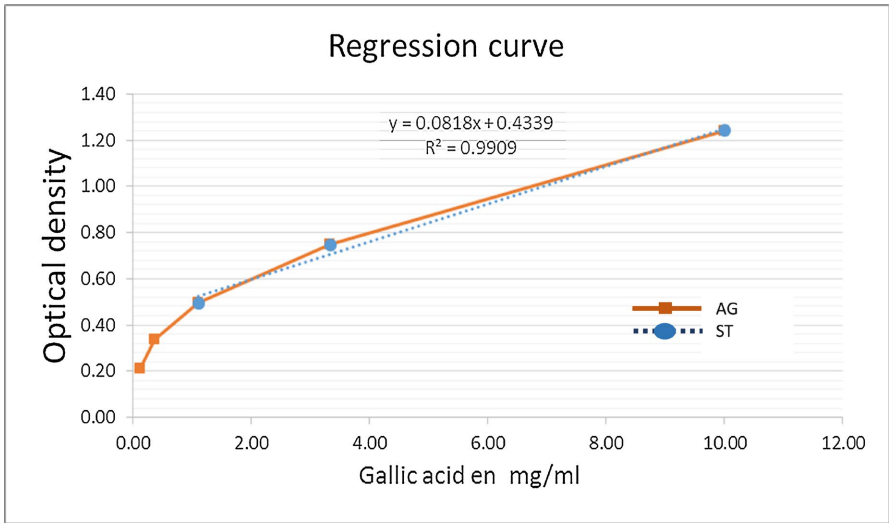

Figure 4. The variation curve of the optical density values obtained at different concentrations of Gallic acid.

From this curve, the regression equation obtained made it possible to determine the total content of polyphenols (meq g of Gallic acid/ml) and of flavonoids (meq g of Gallic acid/ml) contained in the extract of *D. edulis*. One Way Analysis of Variance (ANOVA) was used to determine if the concentration of polyphenols, flavonoids and tannings were significant at 0.05 significant level in the 10 % extract. The results obtained were presented in Table 1 and Table 2 below:

Table 1. Polyphenol content in the *D. edulis* extract in meq g of Gallic acid/ml.

	Polyphenol		Mean	p-value.
OD (650 nm)	0.835	0.860	0.848 ± 0.00013	
Concentration of extracts (mg/ml)	4.952	5.260	5.106 ± 0.048	0.001*
Polyphenols in meq g of Gallic acid/ml	24.759	26.302	25.531 ± 0.240	

Table 2. Flavonoid content in *D. edulis* extract in meq g of Gallic acid/ml.

	Flavonoids		Mean	P-value
OD (650 nm)	0.509	0.513	0.511 ± 0.002	
Concentration	0.927	0.977	0.952 ± 0.024	0.003*
Flavonoid in meq g of Gallic acid/ml	4.635	4.885	4.76 ± 0.12	

Determination of tannins

Tannins are obtained by differentiating between the contents of total polyphenols and flavonoids (non-tannin polyphenol). Table 3 below presents the value of the tannins present in the extract of *D. edulis*.

Table 3. Content of tannins contained in the extract of *D. edulis* in meq g of gallic acid/ml.

	Tannin content		Mean	P-value
Concentration	4.025	4.283	4.154 ± 0.130	0.0015
Tannins in meq g of gallic acid/ml	20.125	21.415	20.770 ± 0.648	

The concentration of polyphenols in the extract of *Dacryodes edulis* is (5.106 ± 0.048 mgGAE/gE) as shown in Table 1. A one-way ANOVA shows that the concentration is significantly high ($p = 0.001 < 0.05$) to carry out the polyphenol's activities in *Dacryodes edulis*. Also, the concentration of flavonoid in the extract is (0.952 ± 0.024 mgGAE/gE) and this amount is significant ($p = 0.003 < 0.05$) to show the activities of flavonoids in the extract. Moreover, the concentration of tannins in the extract were significantly high (4.154 ± 0.130 mgGAE/gE, $p = 0.0015 < 0.05$) to carry out the activities of tannings in the extract.

3.3. Evaluation of Antioxidant Activity (*in Vitro*) of Aqueous Leaf Extract of *Dacryodes edulis*

The antiradical activity of the *D. edulis* extract was determined by its DPPH rad-

ical scavenging capacity (DPPH radical scavenging is an accepted mechanism for screening the antioxidant activity of plants in DPPH essays, violet colour DPPH solutions is reduced to yellow colour product Diphenyl hydrazine) by the addition of the extract in a concentration dependent manner. Thus, a free radical scavenger (plat material) reacts to DPPH to form DPPHH) marked by the change of the purple colour to yellow at different concentrations of the extract (Figure 5 below).

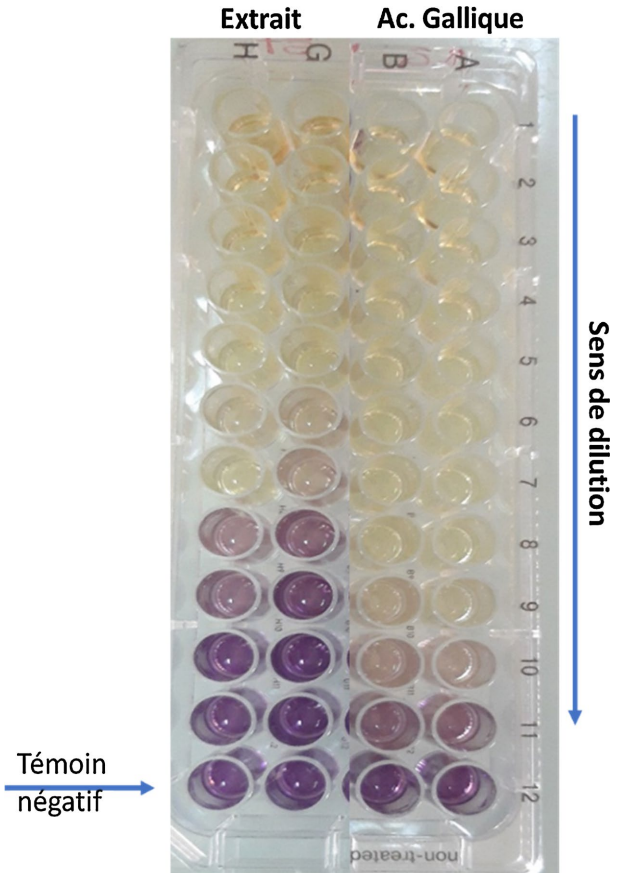

Figure 5. Well plate template obtained after incubation of DPPH radical solution in the presence of *D. edulis* extract and Gallic acid at different concentrations. The content of each cupule read with the spectrophotometer made it possible to obtain the optical density (OD) values which enabled calculation of the percentage (%) trapping of the DPPH radicals at different concentrations of the *D. edulis* extract and Gallic acid. Figure 6 below presents the curves of variation of DPPH radical scavenging percentages at different concentrations of each antioxidant.

It appears from Figure 6 below that a small increase the concentration of *D. edulis* significantly affects the DPPH trapping percentage. Further increase in the concentration show no significant increase in DPPH trapping percentage Thus, *D. edulis* extract has a significant DPPH trapping percentage in their antioxidant properties.

From these different curves, the regression equations were established, which made it possible to determine the IC_{50}s of 0.37 ± 0.00 and 2.76 ± 0.45 µg/ml respectively for Gallic acid and *D. edulis* extract.

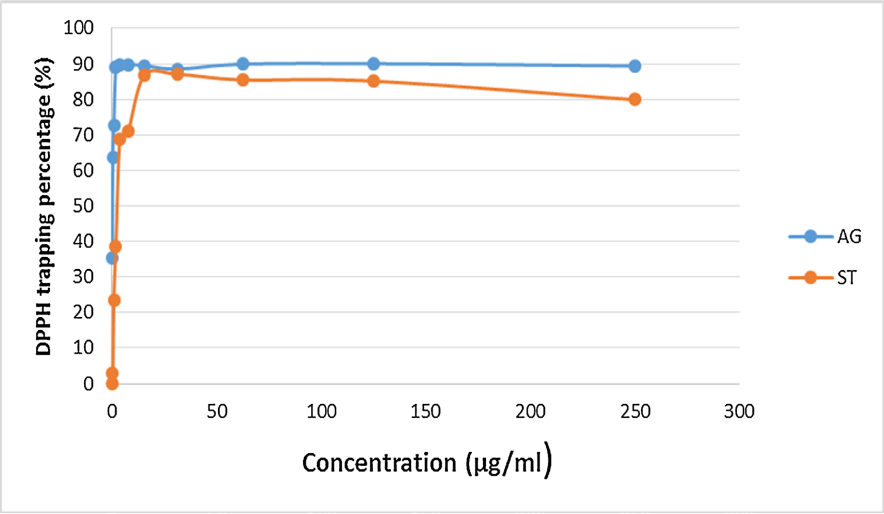

Figure 6. Percentages of DPPH radical scavenging at different concentrations of each antioxidant, with AG = Gallic acid and ST = extract from *D. edulis*.

3.4. Iron III Ions (Fe^3) Radical Scavenging Capacity

Figure 7 below shows the variation curve of the iron III (Fe^{3+}) radical scavenging percentages as a function of the concentrations of *D. edulis* extract and Gallic acid.

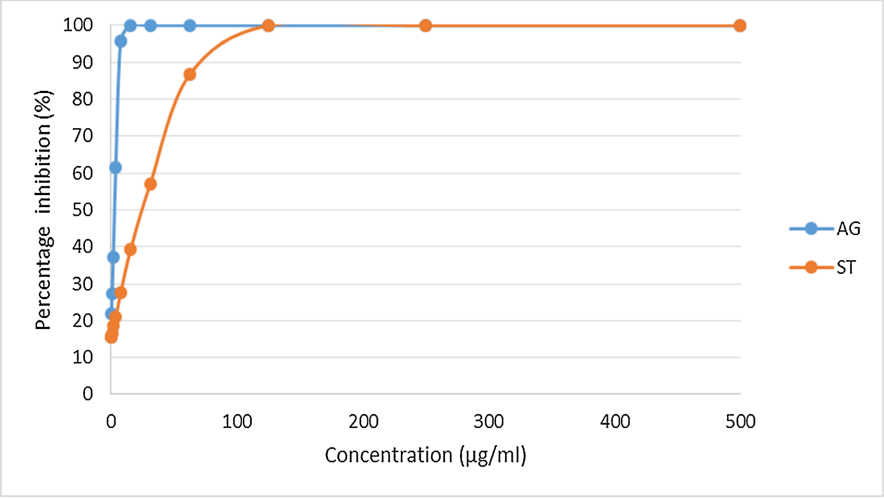

Figure 7. Percentage reduction of Iron III ions (Fe^{3+}) radicals in the presence of the extract and gallic acid, with AG = gallic acid and ST = extract of *D. edulis*.

It appears from Figure 7 above that when the concentrations of *D. edulis* extract increases, the percentage inhibition effect increases exponentially until a point where an increase in their concentration results to no change in percentage inhibition. Thus, *D. edulis* extract significantly affects the percentage reduction of Fe^{3+} in its antioxidant properties. From these different curves, the regression equations were established, which made it possible to determine the (IC_{50} It is (depicts) half the maximum inhibitory concentration (IC_{50}). It is the most widely used and informative measure of a drug's efficacy. Thus, it indicates how much drug is needed

to inhibit a biological process by half (Provides measure of potency). IC_{50} of 29.97 ± 0.05 and 25.03 ± 0.71 µg/ml respectively for Gallic acid and *D. edulis* extract.

4. Discussions

The purpose of this study was to assess the phytochemical characterization of anti-gastric ulcer properties of aqueous leaf extract of *Dacryodes edulis*. Specifically, the study sought to extract the active component of *Dacryodes edulis*, phytochemically screen (quantitative and qualitative) aqueous leaf extract of *Dacryodes edulis* for secondary metabolites and evaluate the antioxidant activity (*in vitro*) of aqueous leaf extract of *Dacryodes edulis*. The extraction yield of the extract of *D. edulis* obtained by decoction was determined by relating the mass of the extract to the mass of leaves initially extracted. The ratio calculation showed that the extraction yield is 10% in milligrams equivalents. This finding collaborates with that of [21] who carry out the same extraction procedures in their research work using the aqueous extract of *D. edulis* leaves in the Universty of Dschang, Cameroon and obtained a percentage yield of 9.67% milligrams equivalents. The quantitative determination of total phytochemical content was evaluated using the methods described by Ramde-Tiendrebeogo *et al.* (2012), Chang *et al.* (2002), and Almaraz-Abarca *et al.* (2007) [19]-[21] These phytochemical analyses were based on the appearance of different colours and the formation of precipitates or products in the final solution. Preliminary phytochemical screening for secondary metabolites of Dacroydes edulis leaves of the 10% extract showed the presence of polyphenols, flavonoids and tannins. These results are similar to those obtain by Omonhinmin and Uche [21] [22] which showed that the aqueous extract of *D. edulis* contains various secondary metabolites such as alkaloids, phenols, flavonoids, triterpenoids, tannins, saponins, anthocyanins, and anthraquinones. The results are also similar to those obtained by Ogboru *et al.*, (2015) [23] in their research work in Nigeria in which they used the aqueous extract of the stem bark of *D. edulis*. In Ivory Coast, the fruitcake of *D. edulis* was also found to contain same groups of molecules [24]. The chemical composition of exudate extracted from the stem of *D. edulis* was the subject of several studies. These studies revealed the presence of several bioactive compounds like saponins, alkaloids, tannins, flavonoids, and phenolic compounds as reported by Koudou *et al.* (2008) [15]. However, contrary to these studies, alkaloids, triterpenoids, saponins, anthocyanins, anthraquinones were grouped under polyphenols and sterols were not detectable in the aqueous extract of *D. edulis* leaves in the present research work. These results suggest that the harvest site and the part of the plant used have a direct impact on the presence of secondary metabolites in the plant.

The quantitative analysis revealed that the aqueous extract of *D. edulis* leaves contains significant amounts of total polyphenols, flavonoids, and Tannins. The results are similar to that obtained by Omonhinmin and Uche (2013) [21] [22]. which shows that aqueous extract of *D. edulis* leaves contains significant amounts of total phenols, flavonoids, and flavonols. The proportions were different (infe-

rior) from those reported by Ogboru *et al.* (2015) [23] in Nigeria who worked with the stem bark and Ano-Aka *et al.* (2018) [24] in Ivory Coast who worked with the fruitcake of the same plant. These results suggest that the harvest site and the part of the plant used also have an impact on the number of secondary metabolites in the plant.

The antioxidant activity (*in vitro*) of aqueous leaf extract of *Dacryodes edulis* was investigated using DPPH radical scavenging capacity and Iron III ions (Fe^{3+}) radical scavenging capacity. The result from this study revealed that aqueous leaf extract of *Dacryodes edulis* show significant antioxidant power or effect due to its low "Half-maximal inhibitory concentrations" (IC50s) of 2.76 ± 0.45 µg/ml DPPH and 25.03 ± 0.71 µg/ml for Iron III ions (Fe^{3+}). The results of this study tie with results from several studies that have shown the antioxidant activity of *D. edulis* leaves. The leaves of the plant showed a significant antioxidant effect with different methods [5] [25]. These antioxidant effects were attributed to the presence of flavonoids in the plant. tropical plum leafs are rich in protein, minerals, and vitamins. (The *Dacryodes edulis* extract studied possessed high protein (07 ± 0.26 g/100 g) which is higher than the range of protein content of various species of plum leaf 1.6 - 2.1 g/100g as reported by Nguefack (2009) [25]. The protein content was an appreciable amount suggesting that the plum leaf was a good source of protein. The extract was also seen to be low in total fat (0.60 ± 0.28 g/100 g) and was rich in magnesium, Phosphates and calcium but poor in Iron and lipids as seen in other research works. From overall analysis, it was seen that the mushroom was nutritionally good. In our search for the biological activity with respect to peptic ulcers, the ulcers were induced by using absolute ethanol. Ethanol readily penetrates the gastric mucosa due to its ability to solubilize the protective mucous and expose the mucosa to the proteolytic and hydrolytic actions of hydrochloric acid and pepsin causing damage to the membrane.

The treatment of ulcers using pharmaceuticals is mostly aimed at either stimulating the mucosal defences or counteracting aggressive factors with goals to relieve pain, heal the ulcer and prevent reoccurrence. Researchers have evaluated a large number of different spices and herbs for their anti-ulcer activities. Many plants have been shown to possess gastro-protective activities such as Azadirachta indica (commomly known as the Neem plant), Alo vera, Carica papaya, Ginseng and many more. These plants are seen as potential therapeutic applications because of their high efficacy and low toxicity. Studies of these medicinal plants showed that they could prevent gastric ulcers in rats in a dose-dependent manner. Like results were seen in the case of *Agariscus* campestris.

5. Conclusion

The findings of this study showed how local medicinal knowledge working hand in glove with modern science can isolate characterise and standardise the active constituents of herbal source and produce better anti-ulcer drugs with fewer side effects. This work will also help locals prioritize their local plum leaf in diets to

help treat and prevent gastritis. It causes them to engage in the active cultivation of the plant rather than just relying on what sprouts out when the first rains come. This will go a long way to increase the country's revenue. *Dacryode edulis* is a tree cultivated in most rural communities by the peasant farmers, mostly for its edible fruits. But all parts of this tree, including leaves, bark, roots, resin, seeds and fruit pulp are used for medicinal purposes. The fruit confers huge economic value on the plant through local and international trade. Phytochemical screening for secondary metabolites of *Dacroydes edulis* leaves extract showed the presence of free polyphenols, flavonoids and tannins. The anti-oxidant screening of DPPH and iron III ions scavenging capacity of the extract of *D. edulis* showed percentage trapping, a property of a good anti-oxidant agent. These anti-oxidant properties are related to the presence of secondary metabolites detected in the plant. This justifies the traditional usage of the leaves and seeds as remedy for stomach problems and their prevention. Further research is on-going to isolate and characterize the medicinal principles of *Dacroydes edulis.*

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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