



## Original Research Article

### Antioxidant activities and estimation of the phenols and flavonoids content in the extracts of medicinal plants used to treat malaria in Ivory Coast

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#### ABSTRACT

Oxidative stress has been implicated in the pathology of many diseases such as inflammatory conditions, cancer, diabetes and the aging. This study was made to evaluate polyphenolic content and antioxidant activity, of several extract of plants. Antioxidant activity was estimated using *in vitro* models like 1,1-diphenyl-2-picryl hydrazyl (DPPH) radical scavenging, chelating ability and reducing power methods. The flavonoids and total polyphenolic contents of the extract were also determined using standard methods. Two ways ANOVA was used for statistical analyses. Phytochemical analysis revealed the presence of polyphenols, leucoanthocyanins, saponins, and flavonoids. The total phenol varied from 91.49±6.32 to 240.7±47.31mgGAE/g in the extracts. Flavonoid contents were between 6.7±1.05 and 62.18±2.037mgQE/g. 1,1-diphenyl-2-picryl hydrazyl (DPPH) radical scavenging effect of the extracts was determined spectrophotometrically. The highest radical scavenging effect was observed in *Newbouldia laevis* with IC<sub>50</sub> = 19.5µg/ml. The different extracts exhibited dose-dependent chelating abilities and reducing power. The abundance of polyphenolic compounds and antioxidant activities of the above Ivoirian medicinal plants could confirm their good therapeutic potential properties attributed by ethnotherapy.

#### Keywords

Antioxidant;  
Antiradical;  
Phyto-chemistry;  
Phenols;  
Flavonoids.

#### Introduction

Since ancient times, people explored the nature, particularly plants in search of new drugs. This has resulted in the use of large number of medicinal plants with curative properties to treat various diseases. Nearly

80% of the world's population relies on traditional medicines for primary health care, most of which involve the use of plant extracts (Sandhya *et al.*, 2006). Several chemical investigations have been

conducted to provide a scientific explanation for their use in traditional medicine (Cheong *et al.*, 2013; Santiago *et al.*, 2014). Recently there has been an upsurge of interest in the therapeutic potentials of medicinal plants as antioxidants in reducing such free radical induced tissue injury. Oxidation process is one of the most important routes for producing free radicals in food, drugs and even living systems (Pourmorad *et al.*, 2006).

During *Plasmodium* infections in humans and experimental models (primates, mouse), a high oxidative stress is generated due to metabolism of compounds rich in iron (Schwarzer *et al.*, 2003; Becker *et al.*, 2004; Hans *et al.*, 2009). Consequences of malaria are to decrease antioxidant enzymes and compounds such as catalase, Glutathione (GSH), Glutathione Peroxidase, superoxide dismutase (SOD), and an increased production of reactive oxygen species (ROS) (Selvam and Mathews, 1991; Delmas-Beauvieux *et al.*, 1995; Erel *et al.*, 1997; Luersen *et al.*, 2000).

Due to the depletion of the immune system natural antioxidants during malaria infection, consuming antioxidants as free radical scavengers may be necessary. In addition, it is quite interesting to note that plant have good antioxidant ability and are safer than the synthetic antioxidants (Halliwell, 1994; Kumpulainen and Salonen, 1999). The antioxidant activity can be attributed to various mechanisms like prevention of chain initiation, binding of transition metal ion catalysts, decomposition of peroxides, reductive capacity, and radical scavenging activity (Gayathri *et al.*, 2012).

The present study aims to quantitatively estimate total phenols, flavonoids and antioxidant potential in different extracts of plants used to treat malaria in order to find

antiplasmodial substances with good antioxidant activity. In this context, *Diospyros monbuttensis*, *Dialium dinklagei*, *Newbouldia laevis*, *Trema orientalis* *Cnestis ferruginea* widely used by traditional healers but little studied for their antimalarial activity have been selected.

*Diospyros monbuttensis* leaves have been reported to possess antibacterial and antimalarial properties (Kayode and Omotoyinbo, 2009; Anie *et al.*, 2011; Olasehinde *et al.*, 2012;). The antimicrobial activities of the plant leaves extract was due to the presence of tannins (Odelola and Okorosobo, 1988; Mallavadhani *et al.*, 1998). Other constituents that have been reported include saponins, anthraquinones, cardiac glycosides and alkaloids (Kayode and Omotoyinbo, 2009).

*Dialium dinklagei* is a tree of Caesalpiniaceae family with trunk cylindrical right. The sheets made up have an acid taste, the flowers are of color yellows and the bark is very thick (Akoué *et al.*, 2013). Not previously reported from Ivory Coast. This species is distributed in West and Central Africa and has been reported from Angola, Cameroon, D.R. Congo, Gabon, Ghana, Guinea, Ivory Coast, Liberia, Nigeria and Sierra Leone (Lock, 1989).

The species *Newbouldia laevis* is native to tropical Africa and grows from Guinea Savannas to dense forests, on moist and well-drained soils. It inhabits the secondary forest extending from Senegal to Cameroon, Gabon, Democratic Republic of Congo, Angola (Arbonnier, 2004). *N. laevis* is widely used in African folk medicine for the treatment of several diseases such as an astringent in diarrhea and dysentery. It is also employed in the treatment against worms, malaria, sexually transmitted

disease, and in the reduction of dental caries (Iwu, 2000; Eyong *et al.*, 2005). Wide diversity of secondary metabolites has also been described depending on the part of the plant used. In root bark, the presence of naphthoquinones and anthraquinones has been reported (Eyong *et al.*, 2005; Eyong *et al.*, 2006) whereas in the leaves, the presence of flavonoids, tannins and terpenes has also been mentioned (Gafner *et al.*, 1997; Samy and Gopalakrishnakone, 2008). The occurrence of quaternary alkaloids has also been identified (Kerharo and Adam, 1974).

*Cnestis ferruginea* Vahl exDC (Connaraceae) is a shrub or climber of deciduous forest and secondary scrubland widely dispersed in West Africa and other tropical parts of Africa (Burkill, 1985). Different parts and preparations of *Cnestis ferruginea* have been reported to be used in traditional African medicine such as the management of conjunctivitis, bronchitis, tuberculosis, migraines, sinusitis, and oral infection (fruits); snakebite, dysentery, syphilis, gonorrhoea, malaria, cough, dysmenorrhoea, enema, ovarian troubles and aphrodisiac (roots); abortion, constipation, fever and pain (leaves) (Gill, 1992, Okafor and Ham, 1999). Studies have shown that aqueous root extract contained alkaloids, flavonoids, saponins, anthraquinones and tannins (Yakubu *et al.*, 2011; Ishola *et al.*, 2011). The fruits have been reported to have anti-microbial effects especially against gram-positive bacteria (Yakubu *et al.*, 2011), while the aqueous root extract has been reported to possess antistress and laxative activities (Ishola *et al.*, 2007; Yakubu *et al.*, 2011). Also, the methanolic root extract has been reported to possess analgesics and anti-inflammatory activity (Ishola *et al.*, 2011). The toxicological implications of the crude alkaloidal fraction from *C. ferruginea* root on the liver function

indices of male rats as well as the cytotoxic activity of the leaves have been reported (Garon *et al.*, 2007; Atere *et al.*, 2009).

*Trema orientalis* (Blume) Linn, a tropical tree, is reported to grow on poor soils and chromite overburdens and hence, might be used as an effective cover to prevent leaching of heavy metals from the minewaste dumps to the neighbouring environs (Samantaray, 1991). It is found from South Africa to Tropical Africa and in warm regions of Asia. The leaves and the bark are used to treat cough, sore throats, asthma, bronchitis, gonorrhoea, yellow fever, toothache, and as an antidote to general poisoning (Yanes, 2007). Some pharmacological research done on the plant has focused on, hypoglycemic activity, analgesic, anti-inflammatory activities, anti-plasmodial activity, diuretic activity, laxativity effect, anti-convulsant activity, anti-helminthic activity, anti-sickling effect, anti-oxidant, and anti-bacterial activity (Adinortey *et al.*, 2013). The leaves of *T. orientalis* contain tannins, saponins, flavanoids, triterpenoid (simiarenol, simiarenone, trematol). (Adinortey *et al.*, 2013) Octacosanoic acid, 1-octacosanyl acetate, simiarenone, simiarenol, episimiarenol, and a new triterpene alcohol, trematol has been isolated from stem bark. (Rastogi and Mehrotra, 1993).

## Materials and Methods

### *Vegetal material and basis for selection:*

Vegetal material is made up of leaves of *Diospyros monbuttensis*, *Dialium dinklagei*, *Newbouldia laevis*, *Cnestis ferruginea* and *Trema orientalis* because, there are few data available on their antiplasmodial activity.

Popular pharmacopoeia methods (Betti,

1998) were used to create the list of plants used to cure malaria. This method consists of gathering data on the popular use of medicinal plants in a given area by direct, collective or individual talks with populations. The method used is based on that previously described by Fezan, et al. (2008). The questions asked to traditional healers were about their knowledge of malaria and the plants used in the preparation of antimalarial remedies, the method of preparation of the remedies, the used parts of the plants and details of administration. The investigations were conducted during November 2012 by ethnobotanical approaches near the actors of traditional medicine, especially markets herbal sellers of Abidjan area.

The leaves of *Diospyros monbuttensis* were collected during the month of January, 2013 from Talahini-Sokoura in Department of Sandegu  (North-east Ivory Coast). Leaves of *Dialium dinklagei*, *Newbouldia laevis*, *Cnestis ferruginea* and *Trema orientalis* were collected during March, 2013 in Abidjan area (South Ivory Coast). Species were identified by Professor Ake Assi of the "Centre National de Floristique", University of Felix Houphouet-Boigny, Abidjan, Ivory Coast. Material vegetal was air-dried during 7–10 days at room temperature at 25°C and powdered. From the powder obtained after spraying of leaves, different extracts have been prepared.

**Chemicals:** All chemicals used were of analytical grade. Methanol, aluminum chloride, potassium acetate, 2,2-Diphenyl-1-picrylhydrazyl (DPPH), ferrous chloride, ferrozine, potassium ferricyanide, Folin-ciocalteu reagent, standards such as L-ascorbic acid, ethylenediamine tetraacetic acid (EDTA), gallic acid, quercetin all from Sigma Chemicals Co. (St. Louis, MO, USA).

**Preparation of crude extract:** The decoction was obtained by dissolving 20g of fine dry powder in 1L of distilled water. The whole was homogenized and boiled for 30minutes. The preparation was first wrung out in a square of white cloth, and then filtered successively twice on an absorbent cotton and once on a Whatman filter paper 3mm. Filtrate 1 was thus obtained. The same operation was repeated with the residue by adding 1 liter of distilled water to obtain the filtrate 2. These two filtrates were gathered and evaporated in "Venticel" oven at 55°C. This series of operations led to the extract of decoction (Zirihi *et al.*, 2003; Bekro *et al.*, 2007).

**Phytochemical screening:** Chemical tests were carried out on the extracts for the qualitative determination of phytochemical constituents as described by Nemlin and Brunel (1995), B kro *et al.* (2007), Bruneton (2009).

#### **Antioxidant activity**

**Total phenolic content:** The total phenolic content of the extract was determined separately using the method of (McDonald *et al.*, 2001; Li *et al.*, 2007). The calibration curve was prepared by mixing methanolic solution of gallic acid (1ml; 0-100 g/ml) with 5ml Folin-Ciocalteu reagent and sodium carbonate (4ml, 1M). We measured absorbance at 765nm and drew the calibration curve. 1ml of extract (100 g/ml) was also mixed with the reagents above and after 15min, the absorbance was measured to determine plant total phenolic contents. Experimentations were carried out in triplicate. The total phenol values are expressed in terms of gallic acid equivalent (mg GAE/g of extract), which is a common reference compound.

**Total flavonoids content:** The total flavonoids content was analyzed by aluminum chloride method (Bahorun *et al.*, 1996; Chang *et al.*, 2002). Each plant extract (0.5ml of 1:100µg/ml) was mixed with 1.5mL methanol, 0.1mL of AlCl<sub>3</sub> (10%), 0.1mL of 1M potassium acetate and 2.8 mL of distilled water. The mixture was allowed to stand for 30min at room temperature (25°C) and absorbance was measured at 415nm with a double beam Perkin Elmer UV/Visible spectrophotometer (USA). The calibration curve was prepared by preparing quercetin solutions at concentrations ranging from 0 to 100µg/ml in methanol. Total flavonoids contents were expressed as mg of Quercetin equivalents (QE)/g of extract. Samples were analyzed in triplicates.

**Reducing power assay:** The reducing power was determined according to the method of Oyaizu, 1986 and Topçu *et al.*, 2007. Different concentrations of the extract (6.25–200µg/ml) in 1.0ml of deionised water were mixed with phosphate buffer (2.5ml, 0.2M, pH6.6) and potassium ferrocyanide (2.5ml, 1%). The mixture was incubated at 50°C for 20min. A portion of trichloroacetic acid (2.5mL, 10%) was added to the mixture, which was then centrifuged at 3000rpm for 10min. The upper layer of the solution (2.5ml) was mixed with distilled water (2.5ml) and FeCl<sub>3</sub> (0.5ml, 0.1%) and the absorbance was measured at 700nm and compared with control. Increased absorbance of the reaction mixture indicated an increased reducing power. Ascorbic acid was used as control, and all the tests were performed in triplicate. The sample concentration at which the absorbance is 0.5 (EC<sub>50</sub>) was estimated by interpolation.

**Chelating ability:** Chelating ability of Fe<sup>2+</sup> was determined according to the method of (Yildirim *et al.*, 2001; Le *et al.*, 2007). Fe<sup>2+</sup> was monitored by measuring the formation

of ferrous iron-ferrozine complex at 562 nm. Different concentration of extract (1ml) in 3.7ml of methanol was mixed with FeCl<sub>2</sub> (0.1ml, 2mM) and ferrozine (0.2ml, 5mM). The resulting mixture was shaken and left to stand for 10min at room temperature. EDTA was used as standard control. The absorbance of the resulting solution was measured at 562nm. The capability to chelate the ferrous iron was calculated using the following equation:

Chelating Effect (%) = [(A<sub>o</sub> - A<sub>1</sub>) / A<sub>o</sub>] X 100; A<sub>o</sub> was the absorbance of the control (containing all reagents except the test compound) and A<sub>1</sub>, the absorbance in presence of sample of extract and standard.

**Free radical scavenging activity:** Hydrogen atom or electron donating abilities of the compounds were measured from the bleaching of the purple-coloured methanol solution of 2,2-diphenyl-1-picryl hydrazyl (DPPH). This spectrophotometric assay uses the stable free radical, DPPH as a reagent (Parejo *et al.*, 2000). Different concentrations of each extract were added, at an equal volume, to methanolic solution of DPPH (100µM). After 30 min at room temperature, the absorbance was recorded at 517 nm. Test was repeated for three times. Vitamin C was used as standard control. The DPPH radical scavenging effect was calculated as inhibition of percentage (I %) using the following formula:

I % = (A Blank - A Sample / A Blank); A blank is the absorbance of the control reaction (containing all reagents except the test compound) and A sample is the absorbance of the test compound. The values of inhibition were calculated for various concentrations of the extract. IC<sub>50</sub> values denote the concentration of sample, which is required to scavenge 50% of DPPH free radicals.

## Statistical analyses

Results are expressed as mean  $\pm$ S.E.M. of three determinants. Comparisons among the groups were tested by two-way ANOVA using Graph Pad Prism, Version 5.0 (Graph Pad Software, San Diego, CA, USA). *P* values  $<0.05$  were considered significant.

## Results and Discussion

**Extractions yield:** Through observations the best yield is obtained with *Trema orientalis* (20%) and similar to those obtained with *Diospyros monbuttensis*, *Dialium dinklagei*, *Cnestis ferruginea* respectively with 18; 18.5 and 17.95%. The lowest is obtained with *Newbouldia laevis* (10.9%). The extraction yields did not differ significantly depending on the plant ( $p=0.4060>0.05$ ).

**Phytochemical screening:** Qualitative analyzes was carried out in extracts of different plants. The results show that all extracts are rich in polyphenols, leucoanthocyanins, saponins, and flavonoids. Others compounds are present or absent according to the plant tested (Table 1).

**Flavonoid and phenol contents of the extracts:** Table 2 show the contents of total phenols that were measured by Folin-Ciocalteu reagent in terms of gallic acid equivalent (mgGAE/g), standard curve equation:  $Y=0.01810 X + 0.07179$ ,  $R^2=0,9911$ . Total phenol varied from  $91.49\pm 6.32$  to  $240.7\pm 47.31$  mgGAE/g in the extract. *Trema orientalis* with total phenol contents ( $240.73 \pm 47.31$  mgGAE/g) had the highest amount in this study followed by *Diospyros monbuttensis* ( $185.59\pm 5.89$  mgGAE/g), *Dialium dinklagei* ( $136.54\pm 12.63$  mgGAE/g), *Cnestis ferruginea* ( $125.58\pm 2.01$  mgGAE/g) then *Newbouldia laevis* ( $91.49\pm 6.32$ mgGAE/g).

There was a statistically significant correlation between different values according the studied plant ( $p=0,0172<0,05$ ).

Flavonoid contents of extracts in terms of quercetin equivalent (Standard curve equation:  $Y= 0.02834X+ 0.01368$ ,  $R^2= 0.9986$  were between  $6.7\pm 1.05$  and  $62.18\pm 2.037$  (Table 2). The flavonoid contents in extracts of *Diospyros monbuttensis* ( $62.18\pm 2.037$  mgQE/gmgQE/g) and *Trema orientalis* ( $59.59\pm 6.424$ mgQE/g) were higher than that in extracts of *Cnestis ferruginea* ( $27.95\pm 2.960$  mgQE/g) and *Newbouldia laevis* ( $22.42\pm 3.85$  mgQE/g). *Dialium dinklagei* showed the lowest flavonoids contents ( $6.7\pm 1.05$ mgQE/g). There was a statistically significant correlation between different values ( $p<0.0001$ ).

### Antioxidant activity

**Chelating ability of ferrous ions:** The curves representing percentage chelating of plant extracts versus EDTA showed a strong slope which tend toward a plateau. As shown in Table 3, extract exhibited dose-dependent chelating abilities on ferrous ions. Different extracts revealed good chelating potencies of 50%. The concentrations of plant extracts and EDTA inducing 50% chelating are significantly different ( $p <0.0001$ ).

**Reducing power assay:** In this assay, yellow colour of test solution changes to various shades of green and blue colour depending upon the reducing power of each antioxidant sample. At different concentrations, plants extract showed higher activities and less than the control. These differences were statistically significant ( $p<0.05$ ). Table 3 shows reductive capabilities of plant extract compared to ascorbic acid. Extract power was increased with sample concentration.

**DPPH radical scavenging activity:** The different extracts showed dose dependent DPPH radical scavenging activity (Table 3). These results show amount of each extract needed for 50% inhibition ( $IC_{50}$ ).  $IC_{50}$  of the standard compound (vitamin C) was  $3,715 \pm 0,385 \mu\text{g/ml}$ , which is the highest radical scavenging activity. The difference between vitamin C and the others is significant ( $P < 0.0001$ ). But the radical scavenging activities of plant extracts are also important and decreased in the following order: *Newbouldia laevis* > *Cnestis ferruginea* > *Dialium dinklagei* > *Diospyros monbuttensis* and *Trema orientalis*. In general these results did not differ significantly ( $P > 0.05$ ) from each other.

**Extractions yield:** Yield obtained with *Trema orientalis* (20%) is similar to those of *Diospyros monbuttensis*, *Dialium dinklagei*, *Cnestis ferruginea* respectively with 18; 18.5 and 17.95%. The yields are high and indicate that the decoction is a good extraction method. These results are comparable to those obtained by Mahmoudi *et al.* (2013) with the decoction of flower of artichoke (*Cynara scolymus* L.) and Mohammedi and Atik (2011) with *Tamarix aphylla*. But, they are lower than those found by Ozsoy *et al.* (2008) with *Smilax excelsa* and superior to those of N'gaman, *et al.* (2009) with *Gmelina Arborea*.

**Phytochemical screening:** Phytochemical analysis is important in the evaluation of bioactive compounds from medicinal plants. Qualitative analyzes show that all the extracts are rich in polyphenols, leucoanthocyanins, saponins, and flavonoids. These results will be helpful to phytochemists and pharmacologists for identification of new active compounds from plants (Ankanna *et al.*, 2012). On the other side the others compounds are present

or absent according to the plant tested. The constituents present in these plants play a significant role in the identification of crude drug. Phytochemical screening is very important in identifying new sources of therapeutically and industrially compounds like alkaloids, flavonoids, phenolic compounds, saponins, steroids, tannins, terpenoids etc (Akindele, 2007).

**Flavonoid and phenol contents of the extracts:** The phenolic and flavonoid compounds quantified in the extracts seemed to be responsible for the antioxidant activity. Phenolic acids, flavonoids are the most commonly found polyphenolic compounds in plant extracts (Wolfe *et al.*, 2003). The antioxidant activity of phenolics plays an important role in absorption or neutralization of free radicals (Basile *et al.*, 1999). Polyphenols also enhance the level of cellular antioxidative system and induce the cytochrome P-450 resulting in detoxifying the activity of carcinogens intracellularly (De Flora, 1988). Total phenol varied from  $91.49 \pm 6.32$  to  $240.7 \pm 47.31$  mgGAE/g in the extract powder. These investigations provide a comprehensive profile of the antioxidant activity of extracts of plants with respect to their phenols and flavonoids content. Many reports of natural antioxidants of plant have been published and their importance in health, food and preventive medicine has been well documented (Halliwell *et al.*, 2005; Patil *et al.*, 2009). The abundance in polyphenol compounds would confirm the therapeutic properties that there are assigned in ethnotherapy. Indeed, several works have demonstrated that polyphenolic compounds confer to the plant several biologic activities (Kossah *et al.*, 2010).

#### **Antioxidant activity**

As results shown, extract exhibited dose-dependent chelating abilities on ferrous ions.

The different extracts revealed good chelating potencies of 50%. At different concentrations, plants extract showed higher activities and less than the control.

The reducing power of *Trema orientalis* was very potent and similar to vitamin C ( $p>0.05$ ). The others plant extract could reduce the most  $Fe^{3+}$  ions, which had a lesser reductive activity than standard ascorbic acid. Increased absorbance reaction indicated increased reducing power.

DPPH radical scavenging method is standard procedure applied to evaluation of antiradical activity. This method is easy, rapid and sensitive way to survey the antioxidant activity of a specific compound or plant extracts (Koleva *et al.*, 2002). DPPH free radicals, which are stable in methanol shows maximum a proton donating substances such as antioxidant. Radicals would be scavenged and absorbed. The different extracts showed dose dependent DPPH radical scavenging

activity.  $IC_{50}$  of the standard compound (vitamin C) was  $3,715 \pm 0,385 \mu g/ml$ , which is the highest radical scavenging activity. It is similar to the one found by Somanjana *et al.*, 2013 with vitamin C. The difference between vitamin C and the others is significant. But the radical scavenging activities in the plant extracts are also important and decreased in the following order: *Newbouldia laevis* > *Cnestis ferruginea* > *Dialium dinklagei* > *Diospyros monbuttensis* and. *Trema orientalis*. In general these results did not differ significantly ( $P>0.05$ ) from each other. The little difference may be due to difference in plant species or due to different environmental conditions (Perveen *et al.*, 2012). In some cases the extracts with strong antiradical activity are abundant in flavonoids or phenolic compounds. The effect of free radical scavenging activity of extracts on DPPH radicals is thought to be due to their hydrogen donation ability of polyphenols of extracts (Kossah *et al.*, 2010).

**Table.1** Results of phytochemical tests

Natural substances	Chemical groups												
	Steroids	Sterols and polyterpenes	Polyphenols	Flavonoids	Anthocyanins	Lucoanthocyanins	Tannins		Quinones	Alkaloids	Coumarins	Cardiotonics Glycosides	Saponins
Gallic							Catechiq						
<i>Diospyros monbuttensis</i>	+	+	+++	+	-	+	+++	-	+++	+	++	-	+++
<i>Dialium dinklagei</i>	-	+++	++	++	+	+	-	++	+	+++	++	+	+++
<i>Newbouldia laevis</i>	-	-	+	++	-	+	-	+	+	++	+	+	+++
<i>Cnestis ferruginea</i>	-	++	+++	+	++	++	+++	-	-	++	-	++	+++
<i>Trema orientalis</i>	-	++	+	+	-	++	+	-	++	-	++	+	+++

- Strong positive: +++; moderately positive: ++; Low positive: +; negative test: -
- Here Decoction extract of leaves used for all plants in present study.

**Table.2** Total phenols and flavonoids contents of extracts

Plant extracts	<i>Diospyros monbuttensis</i>	<i>Dialium dinklagei</i>	<i>Newbouldia laevis</i>	<i>Cnestis ferruginea</i>	<i>Trema orientalis</i>
<b>Phenols content (mg GAE/g)</b>	136,54± 12,63 <sup>a</sup>	185,59± 5,89 <sup>a</sup>	91,49± 6,32 <sup>a</sup>	125,58± 2,01 <sup>a</sup>	240,73± 47,31 <sup>b</sup>
<b>Flavonoids content (mg QE/g)</b>	62,18± 2,03 <sup>a</sup>	6,780± 0,61 <sup>b</sup>	22,42± 3,85 <sup>bcd</sup>	27,95± 2,96 <sup>d</sup>	59,59± 6,42 <sup>a</sup>

- a, b,c and d represent no significant aspect of Tukey statistique test. Values are expressed as mean ± SD (n = 3). Means in the same row with different letters are significantly different ( $P < 0.05$ ).

**Table.3** Antioxidant activities of extracts

Tests	Control	<i>Diospyros monbuttensis</i>	<i>Dialium dinklagei</i>	<i>Newbouldia laevis</i>	<i>Cnestis ferruginea</i>	<i>Trema orientalis</i>
<b>Chelating ability (IC50)</b>	EDTA 4,350±0,15 <sup>a</sup>	9,410±0,310 <sup>b</sup>	14,75±0,25 <sup>c</sup>	7,285±0,21 <sup>d</sup>	45,25±0,25 <sup>e</sup>	24,55±0,25 <sup>f</sup>
<b>Reducing power (EC50)</b>	Vitamine C 20,25 ±0,75 <sup>a</sup>	>200 <sup>b</sup>	133,5 ±2,50 <sup>c</sup>	148,0 ±1,00 <sup>d</sup>	121,5 ±1,00 <sup>e</sup>	24,30 ±0,80 <sup>f</sup>
<b>DPPH radical scavenging activity (IC50)</b>	Vitamine C 3,71±0,38 <sup>a</sup>	22,25±0,25 <sup>b</sup>	21,85±0,15 <sup>b</sup>	19,50±0,50 <sup>c</sup>	21,55±0,15 <sup>b</sup>	22,75±0,25 <sup>b</sup>

- a, b, c, d, e and f represent no significant aspect of Tukey statistique test. Values are expressed as mean ± SD (n = 3). Means in the same row with different letters are significantly different ( $P < 0.05$ ).

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