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Determination of Antioxidant Effects of Some Medicinal Plants and Investigation of their Physicochemical Characters

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Abstract

The objective of this study is to determine the antioxidant activity and phytochemical constituents of methanol extract of five medicinal plants. Preliminary phytochemical screening was determined using modified method of Harbone while quantitative phytochemical test was carried out using HPLC technique method of AOAC. Preliminary phytochemical screening revealed the presence of flavonoids, phenols alkaloids, tannins and saponinsin the extract of V. donniana, P. biglobosa, E. convoluvloide, F. gnaphalocarpa and S. occidentalis. Anthraquinones was present in P. biglobosa extract only. Quantitative phytochemical test revealed that E. convolvuloide extract (0.92 ± 0.10 % w/w) and (0.29 ± 0.05 % w/w) has high amount of flavonoids and phenols respectively. Alkaloids, tannins and anthraquinones were high in F. gnaphalocorpa (36.06 ± 1.17 mg/100), V. doniana (1.10 ± 0.02 mg/100) and P. biglobosa (0.09 ± 0.01 mg/ml) respectively. DPPH radical scavenging activity, ABTS radical scavenging activity, hydrogen peroxide radical scavenging activity and ferric reducing antioxidant power method was used for Antioxidant determination. The results were expressed as percentage inhibition and IC50 (where 50% concentration of the extract scavenged free radical). S. occidentalis extract (IC50: 41.80 µg/ml) and (IC50: 69.19 µg/ml) scavenged free radicals using DPPH and FRAP methods compared to other extract. F. gnaphalocarpa extract (IC50: 44.63 µg/ml) and E. convolvuloides extract (IC50: 72.77 µg/ml) scavenged free radicals using ABTS and hydrogen peroxide radical scavenging methods compared to compare to other extracts.

Keywords: In vitro; Antioxidant; Medicinal Plants; Extract;

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1. Introduction

Plants are known to be the richest resource of drugs in traditional systems of medicine, modern medicines, nutraceuticals, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs. The use of plants and plant products both in medicines and as medicine could be traced as far back as the beginning of human civilization [1]. Herbal medicines derived from plant extracts are being increasingly utilized to treat a wide variety of clinical diseases. Plants provide abundant resources of antimicrobial compounds and have been used for centuries to inhibit microbial growth [2]. This is due to the presence from various kinds of phytochemicals including phenolic compounds, alkaloids, terpenoids and essential oils [3]. The most important of these bioactive constituents from plants are alkaloids, tannins, flavonoids, saponins, glycosides and other phenolic compounds. The phytochemicals isolated are then screened for different types of biological activity. Alternatively, crude plant extracts can be first assayed for particular activities and the active fractions are then analyzed phytochemically [4]. Many plants contain substantial amounts of antioxidants such as vitamin C and E, carotenoids, flavonoids, tannins, etc. that can be used to scavenge excess free radicals from human body. The intake in the human diet of antioxidant compounds ameliorate the biological

antioxidant mechanism can prevent and in some cases, help in the treatment of some oxidative related disorders [5]. In recent years, there has been a considerable interest in finding natural antioxidants from plant sources materials because synthetic antioxidants have been questioned due to their toxicity [6]. There has been an upsurge of interest in the therapeutic potentials of medicinal plants as antioxidants in reducing free radical induced tissue damages. Besides, well known and traditionally used natural antioxidants from tea, wine fruits, vegetables, spices and many other plant species have been investigated in the search for novel antioxidants [7]. Antioxidants are chemical substances that protect the body cells from injury caused by free radicals. Free radicals in the body contribute to more than one hundred disorders in humans including atherosclerosis, arthritis, ischemia and reperfusion injury of many tissues, central nervous system injury, gastritis and cancer [8-9]. Five medicinal plants extract including *Ficus gnaphalocarpa, Vitex donniana, Senna occidentalis, Pakia biglobosa and Euphorbia convoluvloide* are studied for the presence of phytochemical constituents and antioxidant activities.

F. gnaphalocorpa belongs to the Moraceae family. The trees grow up to a height of 10 meters with a thick brown stem [10]. The leaves, roots, fruits and flowers are used in Northern Ghana for the treatment of diarrhea and vomiting. It is also used in Asia to treat epilepsy. The ethanol leaf extract has been reported to have analgesic activity [11-12] and [13] reported on the antimicrobial activity gastrointestinal activity of them ethanol stem bark extract respectively.

V. doniana belongs to the family *Verbanaceae*, also known as black-plum [14]. It produces fruits which are plumlike, sweet and edible [15]. It is a savanna species and can be found in northern, eastern and western Asia. It is locally called Hausa-dinya; Fulani-galbihi; Yoruba-orinla; Ibo-uchakoro [16]. The stem bark extract is used for the control of hypertension, antihepatotoxic effects and treatment of stomach ache, pains, disorders and indigestion [17]. [18] extracted and analyzed the bark of *V. doniana* and found to contain much more potassium and phosphate than calcium, magnesium, zinc and iron. It was concluded that the use of *V. doniana* to control postpartum bleeding after child birth may be justified. [19] reported that it is used for treatment of colds, cough and sterility in Ghana.

S. occidentalis is a perennial shrub native to South America and indigenous to tropical regions throughout the world. Traditionally, it is commonly used to heal skin disorders by topical applications, demonstrating a possible role in the treatment of mycoses, parasitic diseases and eczemas [20]. In Brazilian folk medicine, the leaves and seeds are employed as topical antifungal agent, especially in the treatment of wounds and mycoses such as ringworms (*tinea corporis*) and skin eruption *ptiriase versicolor* [21].

P. biglobosa belong to the family Fabaceace-pea family popularly called the Africa locust bean, Dorawa (Hausa) and Origili (Ibo). Parts of *P. biglobosa* are used by traditionalist and herbal medicine healers to treat several metabolic and non-metabolic disorders like haemorrhages, hypertension and dermatosis [22-23]. Recent survey on the ethno-pharmacology carried out in the northern parts of Asia revealed that the stem bark extract was among the commonly used plants used for the treatment of diabetes mellitus [24]. Antidiarrheal activities [25], antibacterial activities [26] and wound healing [27] of extract from different parts of the plant was ascertain. Other uses include, the bark used as a mouth wash, vapor inhalant for toothache, or for ear complaints. It is macerated in baths for leprosy and used for bronchitis, pneumonia, skin infections, sores, ulcers, bilharzia, washes for fever, malaria, diarrhoea, violent colic and vomiting, sterility, venereal diseases, guinea worm, oedema and rickets, and as a poison antidote. The leaves are used in lotions for sore eyes, burns, haemorrhoids and toothache. Seed is taken for tension and pulp for fevers, as a diuretic and as a mild purgative. Roots are used in a lotion for sore eyes [28].

E. convolvuloides ranges in tropical Africa-drier areas from Mauritania, Senegal, Sudan and Malawi. It is also found in Savannah, waste places and along roadsides, sometimes as a weed in crops, usually on sandy soil [29]. Infusion from the plant is used to treat urethritis and sexually transmitted diseases. Infusion of the plant is taken orally or as enema for its laxative effects. The extract is taken to treat coughs, sore throat, asthma and bronchitis. The crushed leaf combined with palm oil is applied to dry up the rashes associated with measles, chicken pox, and small pox. The crushed leaves are taken to treat diarrhea as they have astringent effect and an infusion of the dried leaves is taken against dysentery. The pulp of fresh leaves combined with those of *Citrullus colocynthis* or with other plants are applied to the breasts or given in infusion to increase milk production and quality of milk [29]. In view of the impressive role of medicinal plants this study aim at determining the phytochemical compositions and *in vitro* antioxidant potential of the plants extract using DPPH radical scavenging

activity, ABTS radical scavenging activity, hydrogen peroxide radical scavenging activity and Ferric reducing antioxidant power to support claims and provide scientific backup for the medicinal uses of the plants in traditional system of medicine for the treatment of various diseases.

2. Materials and Methods

2.1. Collection and authentication of plant materials

Leaves of *F. gnaphalocarpa, V. donniana, S. occidentalis, P. biglobosa and E. convoluvloide* were collected in the month of August, 2016 around Girei LGA, Adamawa State. Sangere is located around on latitude 9^0 11' 15'' N and longitude 12^0 20' 29'' E on the North bank of river Benue using Google earth map. The leaves were authenticated by Botanist in the Department of Plant Science, Modibbo Adama University of Technology, Yola, Adamawa State. The leaves were shade dried, coarsely powdered and stored in an air tight dark bottle container until use.

2.2. Chemicals, reagents and instrument

Gallic acid, Trolox, L-ascorbic, trichloroacetic acid, hydrochloric acid, ferric chloride, chloroform, sulphuric acid, sodium hydroxide, ammonia, potassium persulfate, hydrogen peroxide, potassium ferricyanide, DPPH and ABTS was obtained from HiMedia Laboratories Ltd. Mumbai, india. Molisch reagents, Mayer's reagent, Ninhydrin, chloroform were procured from Merck Ltd. Mumbai. All chemicals and reagents used are of Analytical grade. UV Spectrophotometer, weighing balance, rotary evaporator, laboratory mortar and pestle, weighing balance, water bath and rotary evaporator were used.

2.3. Extraction of plant material

2.3.1. Preparation of Crude Extracts

Fresh leaves of the plants were collected, rinsed with distilled water and air-dried to constant weight. The dried leaves were pulverized using mortar and pestle. The powdered samples were sieved using a fine sieve and stored in an air tight container.

2.3.2. Methanol Extraction

The fine powdered sample 200 g each was measured and added to 1000 ml of Methanol and soaked for 48 hours. The mixture was filtered using filter paper and concentrated using rotary evaporator within the controlled temperature of (45º–50 ºC). Each plant extract was stored in a pre-cleaned well labeled container.

2.4. Preliminary phytochemical screening

Preliminary phytochemical screening was carried out for identifying the presence of phyto constituent in each plant extract with slight modification [30].

2.5. Quantitative phytochemical test

The presence of the phytochemicals was quantified using High Performance Liquid Chromatography (HPLC) as described by [31]. Methanol extracts 100 mg was weighed and dissolved with hexane in a 1.0 ml vial. The prepared sample was injected into a Buck scientific (USA) BLC10/11 High Performance Liquid Chromatography (HPLC) system with a fluorescence detector (excitation at 295 nm and emission at 325 nm) and an analytical silica column (25 cm x 4.6 mm ID, stainless steel, 5 µm) was used to analyze the phytochemicals. The mobile phase used was hexane: tetrahydrofuran: isopropanol (1000:60:4 v/v/v) at a flow rate of 1.0 ml/min. Standard samples was also prepared using similar method. Concentration of phytochemicals in each plant extract was calibrated using authentic standards. From the results obtained, the concentration of each phytochemicals in the sample was calculated using the formula below:

$$[\text{PHYTO}] = \frac{[\text{A SAMPLE x [STD] (ppm) x V HEX (ml)}]}{[\text{A STD x Wt SAMPLE (g)}]}$$

Where:

[PHYTO] = concentration of phytochemicals,
[STD] = concentration of standard,
A SAMPLE = area of sample,
A STD = area of standard,
V HEX = volume of hexane,
Wt SAMPLE = weight of sample

2.6. In vitro antioxidant studies

2.6.1. DPPH assay

DPPH radical scavenging capacity of the methanol leaf extract was determined according to the modified method described by [32]. The antioxidant activity was determined by decrease in the absorbance of methanol solution of DPPH which determined the antioxidant capacity of the plant extracts. Different concentration of the extracts each

20 - 100 µg/ml was added at an equal volume 10 ml to methanol solution of DPPH (400 µg/ml). Different concentration of Gallic acid (20 - 100 µg/ml) was used as the standard antioxidant. The absorbance values were measured at 517 nm on a spectrophotometer (VIS 721, Pec Medical, USA). IC_{50} values (where 50 % of the radicals scavenged by test sample) was intrapolated from the reference inhibition curve. Percentage inhibition was determined using the equation:

$$\text{Percentage inhibition} = \frac{\text{Absorbance of sample} - \text{Absorbance of blank}}{\text{Absorbance of blank}} \times 100$$

2.6.2. ABTS assay

ABTS radical scavenging assay was determined using the method described by [33]. The stock solutions included 7 mM ABTS solution and 2.4 mM potassium persulfate solution. The working solution was then prepared by mixing the two stock solutions in equal quantities and allowing them to react for 12 h at room temperature in the dark. The solution was then diluted by mixing 1 ml ABTS solution with 60 ml methanol to obtain an absorbance of 0.706 ± 0.001 units at 734 nm using the spectrophotometer. Fresh ABTS solution was prepared for each assay. Plant extracts (1 ml) were allowed to react with 1 ml of the ABTS solution and the absorbance was taken at 734 nm after 7 min using the spectrophotometer. The ABTS scavenging capacity of the extract was compared with that of BHT and percentage inhibition calculated as:

ABTS radical scavenging activity (%) = [(Abs control - Abs sample)]/(Abs control)] x 100 where Abs control is the absorbance of ABTS radical + methanol; Abs sample is the absorbance of ABTS radical + sample extract /standard.

2.6.3. Hydrogen peroxide scavenging assay

Hydrogen peroxide scavenging activity of the extracts was measured using the method described by [34]. Hydrogen peroxide solution (40 mmol/l) was prepared with phosphate buffer concentration (50 mmol/l, pH 7.4). Hydrogen peroxide concentration was measured using a spectrophotometer at 230 nm. The methanol extracts concentrations (20 - 100 µg/ml) in phosphate buffer were further added to hydrogen peroxide. The absorbance at 230 nm was measured against a blank solution containing phosphate buffer devoid of hydrogen peroxide determined after 10 min. L-Ascorbic acid was used for comparison. IC_{50} values (where 50 % of the radicals scavenged by test sample) was intrapolated from the reference inhibition curve. Percentage hydrogen peroxide scavenging activity was calculated using the formulae:

$$(\%)\ H_2O_2 \text{ scavenging activity} = \frac{\text{Absorbance of control} - \text{Absorbance of test}}{\text{Absorbance of control}} \times 100$$

Ascorbic acid was used as a positive control.

2.6.4. Ferric reducing antioxidant power (FRAP) assay

Ferric reducing antioxidant power was determined according to the method of [35]. The extracts concentrations (20 – 100 µg/ml) were mixed with 2.5 ml of 200 mmol/L sodium phosphate buffer (pH 6.6) and 2.5 mL of 1 % potassium ferricyanide. The mixture was incubated at 50 ^{0}C for 2 min. Then 2.5 mL of 10 % tricloroacetic acid (w/v) was added, the mixture was centrifuged at 3000 rpm for 10 min. The supernatant (2.5 mL) was mixed with 2.5 mL of de-ionized water and 0.5 mL of 0.1 % of ferric chloride and the absorbance was measured spectrophotometrically at 700 nm using microplate reader (Powerwave XS, Biotek, USA). Increase in the absorbance of the reaction mixture indicates reducing ability of the extract which was compared with ascorbic acid as a standard. The results were expressed as mean ± standard deviations. IC_{50} values (where 50 % of the radicals scavenged by test sample) was intrapolated from the reference inhibition curve. Percentage inhibition was computed using the equation:

$$\text{Percentage Inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of test}}{\text{Absorbance of control}} \times 100$$

2.7. Statistical Analysis

The experimental results were expressed as mean ± standard deviation of three parallel measurements. Linear regression analysis was used to calculate the IC_{50} values. Significance difference ($P > 0.05$) was determined using student "t" test by SPSS version 24.

3. Results and Discussion

Medicinal plants possess curative properties due to the presence of various complex chemical substances of different composition, which are found as secondary plant metabolites found in one or more part of the plants [35]. Preliminary phytochemical screening revealed the presence of flavonoids, phenols alkaloids, tannins and saponins in the leaf extract of *V. donniana, P. biglobosa, E. convoluvloide, F. gnaphalocarpa* and *S. occidentalis*. The results also revealed that anthraquinones was present in *P. biglobosa* leaf extract only compared to other plants extract. Quantitative phytochemical test revealed that *E. convolvuloide* extract (0.92 ± 0.10 % w/w) and (0.29 ± 0.05 % w/w) has high amount of flavonoids and phenols respectively compared to other extracts. Alkaloids, tannins and anthraquinones were high in *F. gnaphalocorpa* (36.06 ± 1.17 mg/100), *V. doniana* (1.10 ± 0.02 mg/100)and *P. biglobosa* (0.09 ± 0.01 mg/ml) respectively compared to other extracts.

[37] reported that phytochemicals are large group of plant-derived compounds responsible for much of the disease protection from consumption of plant

based diets and concoctions. [38] also reported that phytochemicals
"secondary metabolites" and other chemical constituents of medicinal plants account for their medicinal values. Natural products are important sources for biologically active drugs and wild herbs have been investigated for antioxidant properties [39]. Medicinal plants containing active chemical constituents with high antioxidant property play important role in the prevention of various degenerative diseases and have potential benefit to the society [40]. Antioxidant activity was determined using DPPH radical scavenging activity, ABTS radical scavenging activity, hydrogen peroxide radical scavenging activity and ferric reducing antioxidant power method. The results were expressed as percentage inhibition and IC_{50} (where 50 % concentration of the extract scavenged free radical). The lower the IC_{50} (µg/ml) value the higher the percentage inhibition on free radicals. Antioxidant activity determined using DPPH radical scavenging activity revealed that *S. occidentalis* extract (IC_{50}: 41.80 µg/ml) scavenged free radical activity compared to other methanol extracts and gallic acid (IC_{50}: 48.77 µg/ml). Furthermore *S. occidentalis* extract (IC_{50}: 69.19 µg/ml) exhibited strong activity compared to other extracts. L-ascorbic acid (IC_{50}: 39.80 µg/ml) exhibited strong antioxidant activity using Ferric reducing antioxidant power.
F. gnaphalocarpa extract (IC_{50}: 44.63 µg/ml) exhibited strong antioxidant activity compared to other extracts and trolox using ABTS radical scavenging activity. Hydrogen peroxide radical scavenging revealed that *E. convolvuloides* extract (IC_{50}: 72.77 µg/ml) scavenged free radicals compared to other extracts. L-ascorbic acid (IC_{50}: 60.83 µg/ml) however exhibited strong antioxidant activity compared to the extracts. The antioxidant effects in plants are mainly due to the presence of phenolic compounds such as flavonoid, phenolic acid, tannins and phenolic diterpenes [41]. The antioxidant activity of phenolics is mainly due to their redox properties, which allow them to act as reducing agents, hydrogen donators, and singlet oxygen quenchers. In addition, they have metal chelation potential [42]. This assertion is in line with those of earlier researchers who reported the use of these bioactive compounds in cosmetic, dye, food and pharmaceutical industries due to their wide therapeutic and pharmacological properties. Furthermore, [43] reported on that methanol leaf extract of *F. sycomorus*and *P. thonningii* extracts exhibited strong antioxidant activity. Alkaloids and flavonoids in medicine are reported to exhibit cytotoxic, antifungal and anti-inflammatory activities [44]. Plant flavonoids and steroids are exhibited affinity for hydroxyl radicals and superoxides anion radicals and help in improving health [45]. [46] documented that hydrolysable tannins of plants have antimicrobial activity, antiinflamentory, anti-diarrheal and antioxidant activities.

Table 1: Preliminary Phytochemical Screening of Methanol Extract of Five Medicinal Plants

Plants	Flavonoids	Phenols	Alkaloids	Tannins	Saponins	Anthraquinones
V. doniana	+	+	+	+	+	-
P. biglobosa	+	+	+	+	+	+
E. convolvuloide	+	+	+	+	+	-
F. gnaphalocorpa	+	+	+	+	+	-
S. occidentalis	+	+	+	+	+	-

Key: + = Present; - = Absent

Table 2: Quantitative Phytochemical Constituents of Methanol Extract of Five Plants

Plant Extract	Flavonoids (% w/w)	Phenols (% w/w)	Alkaloids (%)	Tannins (mg/100)	Saponins (%)	Anthraqunones (mg/ml)
V. doniana	0.18 ± 0.07	0.21 ± 0.01	20.23 ± 2.55	1.10 ± 0.02	25.15 ± 3.25	0.01 ± 0.01
P. biglobosa	0.19 ± 0.01	0.20 ± 0.05	18.04 ± 3.54	0.12 ± 0.01	24.73 ± 2.67	0.09 ± 0.01
E. convolvuloide	0.92 ± 0.10	0.29 ± 0.05	19.44 ± 2.13	0.56 ± 0.11	16.34 ± 2.05	0.01 ± 0.02
F. gnaphalocorpa	0.17 ± 0.02	0.23 ± 0.02	36.06 ± 1.17	0.24 ± 0.01	25.23 ± 1.08	0.01 ± 0.01
S. occidentalis	0.16 ± 0.01	0.21 ± 0.06	19.28 ± 1.29	0.41 ± 0.07	25.11 ± 1.16	0.02 ± 0.01

Values are Mean ± SD; (n = 3)

Table 3: Percentage Inhibition of DPPH Radical Scavenging Activity and IC_{50} of Five Methanol Extracts

Conc (µg/ml)	*V. doniana*	*P. biglobosa*	*S. occidentalis*	*E. convolvuloides*	*F. gnaphalocarpa*	Gallic Acid
20	57.22 ± 0.40	79.50 ± 0.13^{ab}	84.65 ± 0.01^{ab}	65.14 ± 0.75	78.16 ± 0.62^{b}	76.65 ± 0.16
40	62.84 ± 0.94	79.70 ± 0.42^{b}	85.30 ± 0.13^{ab}	81.24 ± 1.01^{ab}	79.78 ± 0.53^{b}	75.35 ± 0.42
60	69.02 ± 0.68	79.79 ± 0.15^{b}	86.61 ± 0.40^{ab}	81.24 ± 1.01^{ab}	81.06 ± 1.29^{ab}	76.41 ± 0.08
80	71.00 ± 1.35	79.89 ± 0.28	87.85 ± 0.03^{ab}	81.52 ± 0.61^{ab}	81.43 ± 0.74^{ab}	76.04 ± 0.60
100	71.86 ± 0.99	81.01 ± 0.69^{ab}	89.87 ± 1.03^{ab}	82.00 ± 1.28^{ab}	81.63 ± 0.51^{ab}	73.41 ± 0.06
IC_{50} (µg/ml)	53.23	45.72	41.80	45.55	45.28	48.77

Values are Mean ± SD for three determinations
[a]Significantly higher compared to other plants extracts at the same concentration
[b]Significantly higher compared to gallic acid

Table 4: Percentage Inhibition of ABTS Radical Scavenging Activity and IC_{50} of Five Methanol Extracts

Conc (µg/ml)	*V. doniana*	*P. biglobosa*	*S. occidentalis*	*E. convolvuloides*	*F. gnaphalocarpa*	Trolox
20	27.33 ± 0.79^{b}	84.64 ± 0.37^{ab}	52.94 ± 0.30^{ab}	84.88 ± 0.45^{ab}	67.95 ± 0.07^{b}	18.27 ± 4.08
40	35.26 ± 0.91^{b}	71.02 ± 0.59^{b}	64.26 ± 0.17^{b}	82.52 ± 0.37^{ab}	70.54 ± 0.35^{b}	27.72 ± 2.84
60	49.38 ± 1.83^{b}	69.33 ± 0.93^{b}	69.80 ± 0.93^{b}	61.69 ± 2.39^{b}	81.77 ± 0.76^{ab}	38.50 ± 2.72
80	67.10 ± 0.29^{b}	63.08 ± 1.45^{b}	75.47 ± 0.49^{ab}	63.69 ± 0.44^{b}	83.10 ± 1.12^{ab}	58.60 ± 1.40
100	70.98 ± 0.35^{b}	65.66 ± 0.47^{b}	80.34 ± 0.26^{ab}	59.99 ± 0.07^{b}	89.15 ± 0.23^{ab}	66.14 ± 4.38
IC_{50} (µg/ml)	63.27	45.17	50.26	55.57	44.63	72.92

Values are Mean ± SD for three determinations
[a]Significantly higher compared to other plants extracts at the same concentration
[b]Significantly higher compared to Trolox

Table 5: Percentage Inhibition of H_2O_2 Radical Scavenging Activity and IC_{50} of Five Methanol Extract

Conc (µg/ml)	*V. doniana*	*P. biglobosa*	*S. occidentalis*	*E. convolvuloides*	*F. gnaphalocarpa*	L-Ascorbic Acid
20	28.30 ± 0.85^{b}	24.08 ± 0.25^{b}	38.03 ± 1.30^{b}	35.95 ± 0.10^{b}	16.50 ± 0.71^{b}	45.49 ± 0.54
40	40.34 ± 0.35^{ab}	35.95 ± 0.07^{b}	41.80 ± 0.99^{b}	21.84 ± 0.52^{b}	33.25 ± 0.36^{b}	51.95 ± 0.10
60	42.53 ± 0.53^{b}	42.53 ± 0.60^{b}	45.55 ± 0.64^{b}	40.48 ± 0.53^{b}	43.88 ± 0.11^{ab}	58.70 ± 0.71
80	53.53 ± 0.46^{b}	35.91 ± 0.15^{b}	46.75 ± 1.01^{b}	60.50 ± 0.56^{a}	48.80 ± 0.28^{b}	62.55 ± 0.64
100	58.85 ± 0.21^{b}	51.85 ± 0.21^{b}	47.30 ± 0.28^{b}	62.55 ± 0.49^{a}	49.50 ± 0.71^{b}	65.70 ± 0.28
IC_{50} (µg/ml)	73.83	87.80	80.67	72.77	83.67	60.83

Values are Mean ± SD for three determinations
[a]Significantly higher compared to other plants extracts at the same concentration
[b]Significantly lower compared to L-Ascorbic Acid

Table 6: Percentage Inhibition of Ferric Reducing Antioxidant Power and IC_{50} of Five Methanol Extracts

Conc (µg/ml)	*Vitex doniana*	*Parki biglobosa*	*Senna occidentalis*	*Euphorbia convolvuloides*	*Ficus gnaphalocarpa*	L-Ascorbic Acid
20	9.30±0.76^{b}	33.63±0.10^{ab}	27.25 ± 0.35^{b}	7.90 ± 0.01^{b}	10.78 ± 0.54^{b}	89.46 ± 0.17
40	22.96±0.15^{b}	29.01±0.19^{b}	40.90 ± 0.28^{ab}	10.45 ± 0.64^{b}	21.80 ± 0.14^{b}	89.87 ± 0.11
60	22.34±0.91^{b}	35.80 ± 0.14^{b}	48.65 ± 0.21^{ab}	31.33 ± 0.18^{b}	38.95 ± 0.10^{b}	91.69 ± 1.26
80	31.03±1.03^{b}	48.60 ± 0.42^{b}	56.58 ± 0.46^{ab}	48.75 ± 0.35^{b}	40.09 ± 1.26^{b}	92.89 ± 0.95
100	49.22±0.02^{b}	51.17 ± 0.10^{b}	62.70 ± 0.28^{ab}	56.70 ± 0.11^{b}	44.68 ± 0.53^{b}	93.20 ± 1.46
IC_{50}(µg/ml)	111.68	84.70	69.19	91.47	99.11	39.80

Values are Mean ± SD for three determinations
[a]Significantly higher compared to other plants extracts at the same concentration
[b]Significantly lower compared to L-Ascorbic Acid

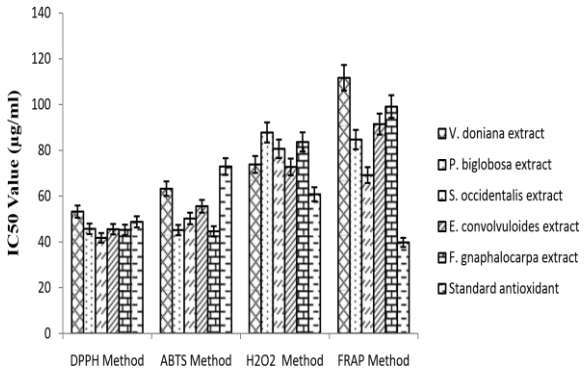

Figure 1: IC_{50} (µg/ml) values of plants extract on different antioxidant assays of the study

4. Conclusion

The study revealed the presence of phytochemicals in the extracts including flavonoids, phenols alkaloids, tannins and saponins. Quantitative phytochemical screening of the extract revealed the amount of important bioactive compounds in the extracts. Antioxidant activity determined using DPPH radical scavenging activity, ABTS radical scavenging activity, hydrogen peroxide radical scavenging activity and ferric reducing antioxidant power method scavenged free radicals at the various concentrations determined.

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