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Determination of Total Phenolic, Total Flavonoid and Antioxidant Activities of the Leaf Extracts of *Capparis tomentosa* (Gumero)

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Abstract

Capparis tomentosa is one the varieties of *Capparis* species commonly used traditional medicinal plants for various ailments like headache, pneumonia and malaria. The study was conducted to determine TPC, TFC and anti-oxidant activities of different solvent extracts of *Capparis tomentosa*. The results indicated that all extracts have reducing power of DPPH radical. Methanol and ethyl acetate leaf extracts of *Capparis tomentosa* have the highest total reducing power (90.38% and 82.193% respectively at concentration of 125 ppm for DPPH assay) and petroleum ether showed the lowest reducing power (0.392% at the lowest concentration 25 ppm) and for FRAP assay the same is true as DPPH assay; methanol extract has the highest reducing power (85.574%) next to the standard ascorbic acid (89.866%) at a concentration of 200 ppm and the lowest was petroleum ether extract (76.533%) at the same concentration. When the concentration of the standard and the extract decrease its anti-oxidant activities also decrease. Qualitative phytochemical screening proves the existence of alkaloids, flavonoids, steroids, terpenoids, saponins and tannins. Quantitative analysis of the extract for total phenolic and total flavonoid content revealed that the total phenolic content ranged from 53.93 to 195.97 mg/g of dry weight of extracts which expressed as gallic acid equivalents. The total TFC ranged from 58.4 mg/g (petroleum ether extract) to 175.104 mg/g (methanol extract) of dry weight of which was expressed as catechin equivalents.

Keywords: *Capparis tomentosa* • Antioxidant activity • Total phenolic content • Total flavonoid contents

Introduction

Organic chemistry because it stands nowadays has evolved in large part from the chemistry of herbal merchandise. The have a have a take a study of herbal merchandise has been and remains a first-rate deriving stress with inside the improvement of the fields of natural and medicinal chemistry. Natural merchandise are expected to play outstanding function as re-assets of recent drugs with inside the years due to their incomparable structural diversity, quite small dimensions of pretty some them, and their drug like properties, because of this that their functionality to be absorbed and metabolized. During the three hundred and sixty five days 2000 to 2010 50% of the small pharmaceutically crucial molecules have been found that is related to the arena of herbal merchandise. Obviously herbal merchandise will live quite crucial as re-assets of medicine [1].

Medicinal flowers had been applied for lots of years as most important re-assets of treatments for humans in the course of the world. In specific components of Ethiopia, peoples had been the usage of conventional natural remedy for addressing their number one health-care want and to deal with numerous illnesses and bodily damages. And maximum of the humans of Ethiopia are exceedingly depending on plant primarily based totally conventional clinical practices. Many medicinal flowers had been observed *via* way of means of humankind *via* way of means of trial and error. In Turkey, medicinal fragrant flowers are generally utilized in each day existence to deal with plenty of sicknesses. Turkey is an appealing supply of medicinal flowers due to its numerous flowers and interest to a variety. It is thought that plant extracts and additives showcase critical organic activities, especially antimicrobial antifungal antibacterial and antioxidant activities. Most drug treatments are actually synthetic pharmacologically, with natural origins accounting for 25% of them. Free radicals are generated in the course of the frame *via* way of means of the poisonous air we breathe in the course of the day, toxic compounds in spoiled foods, additives, subconscious eating, and inactivity. Oxygen atoms damaged off *via* way of means of those dangerous results from outdoor flow into freely with inside the frame, breaking down hydrogen atoms and inflicting tissue damage. Free radicals specially assault the mobileular and immune system. Molecules that limit and block the impact of unfastened radical's with inside the frame and save you chain reactions which could motive many sicknesses and untimely getting old are called "antioxidant" substances. Prevention of unfastened radical-mediated reactions that result in difficult-to-deal with troubles which include getting old, cancer, and diabetes is most effective viable with the assist of antioxidant compounds. As is thought, antioxidants are often observed in inexperienced and red leaved flowers. At the equal time, nutrients A, C, and E display herbal antioxidant properties.

Capparis tomentosa belongs to the own circle of relatives capparaceae generally known as Woody Caper Bush in English. It is an indigenous South African plant that grows certainly with inside the savanna Forest of Western, Eastern and Southern Africa. It is a scrambling shrub, now and again maturing right into a tree that could develop as excessive as 10 meters tall and is protected with scattered spines. It is nicely branched and the branches are generally protected with thick yellow hairs; even the robust, recurved spines are frequently hairy. The twigs and leaves are yellow-inexperienced in shadeation and are protected in soft, velvety hairs. The square leaves are approximately 50 × 20 mm, with more than one sharp, hook like thorns at the junction of the stem and leaf base. The white and

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red flora has a couple of stamens. The fruit is red to orange in shadeation, spherical and stalked. The seeds are surrounded with the aid of using fleshy, gray fruit pulp. In natural and conventional medicine, *C. tomentosa* used to deal with rheumatism, madness, snakebite, chest pain, jaundice, malaria, headache, coughs, pneumonia, constipation, infertility and to save you abortions. It is used to deal with leprosy, tuberculosis and gonorrhea. The roots are boiled in water and 1/2 of a cup of this infusion is under the influence of alcohol 3 instances in keeping with day to manipulate cough and chest pain. Despite the continuing use of *C. tomentosa* in control and remedy of many illnesses amongst many communities, it's been suggested to be poisonous to livestock [2].

Capparis tomentosa is one of the nice recognized woody species with magico-medicinal properties, and its miles typically utilized in ritual ceremonies. This plant is historically used to deal with numerous ailments consisting of madness, snakebite, headache, impotence, sterility and etc. In spite of its huge software in conventional medicine, little is thought approximately the photochemistry and pharmacological sports of *Capparis tomentosa*. Preliminary screening confirmed said antimicrobial activity, which warrants greater special studies. The reviews at the toxicity of numerous plant elements are contradictory, and greater studies are wanted earlier than strategies for secure utilization as medicine, forage and meals flora.

Objectives

The important goal of this have a look at changed into to perform phytochemical investigations and assessment antioxidant sports of leaf extracts of *Capparis tomentosa*. The unique goal of this have a look at to decide the entire phenolic contents, the entire flavonoid contents and to decide its antioxidant sports of the exclusive solvent extract of *Capparis tomentosa* through the usage of FRAP and DPPH assay. The use of herbal merchandise as drugs has been defined in the course of records with inside the shape of conventional drugs. The locating of this have a look at offers trace approximately antioxidant sports of the goal plant which intern informs as approximately its antimicrobial sports. This have a look at shows that there's opportunity to isolate capacity antimicrobial tablets from this medicinal plant. The locating of this have a look at May also trace that capacity lead molecule may be remote from this medicinal plant that may be a base for synthesis of powerful antimicrobial tablets [3].

Materials and Methods

Study design and study area

Bahir Dar city is positioned on the southern shore of lake tana that is located north western part of Ethiopia approximately 565 km from Addis Ababa, is the capital of Amhara National Regional State (ANRS). The Experiment became completed at Bahir Dar University department of chemistry post graduate research laboratory, that is located on variety 11°59' N and longitude 37°39' E, at an altitude of 1840 m above sea level.

Plant materials

The fresh leaves of *Capparis tomentosa* were collected from Bahir Dar city around Lack Tana in 2018/2019 and were ready for investigation.

Instrument and apparatus

Beaker, volumetric flask, plastic containers, analytical balance (RADWAG; PS 360/C/1, China), sieve 1 mm, heating mantle, rotary evaporator (RE-2S-VD, German), pH meter (HI 99161, China), UV spectrophotometer Cary 60 Agilent technologies (China).

Chemicals and reagents

The analytical grade chemicals and reagents used for this study were distilled water, ethyl acetate, 99.9% methanol, 99.9% petroleum ether, 10% Ferric Chloride ($FeCl_3$) (British drug house Ltd. England), Wagner's reagent (Iodine in potassium iodide), Aluminum Chloride ($AlCl_3$), Sodium Nitrite ($NaNO_2$) (Blulux Laboratories (P) Ltd.-121001),37% hydrochloric acid (Bluluxlaboratories(p) Ltd., India), 99% sulphuric acid (Bluluxlaboratories (p) Ltd., India), sodium hydroxide pellets AR 98% (Breckland Scientific Supplies), Nitric Acid (HNO_3), sodium carbonate (Blulux Laboratories (P) Ltd.-121001), NaH_2PO_4, Na_2HPO_4, trichloroacetic acid (Blulux Laboratories (P) Ltd.-121001), potassium hexacyanoferrate (II), iron chloride, ascorbic acid (Blulux Laboratories (P) Ltd.-121005), folin-ciocalteu reagent, gallic acid, DPPH, quercetin/catechin, 30% ammonia solution. 99.9% petroleum ether (Blulux Laboratories (P) Ltd.-121001), 98.8% acetone, 99.8% chloroform.

Instrumentation

The necessary apparatus and instruments used for this study were electronic beam balance for mass measurement, rotary evaporator for concentrating the filtrate to dryness, electrical shaker to mix the mixture well, UV-visible spectrophotometer for the absorbance measurement, digital pH meter for pH measurement, volumetric flask, beaker, conical flask with different, Whiteman no.1 filtrate paper, aluminum foil, micropipette, incubated agar, cuvettes, and others were used for different purposes.

Methodology

Sample preparation: The selected samples were thoroughly washed with tap water to remove all the dust particles. Then the cleaned samples were air dried in the room temperature (25°C) with aluminum foil until to remove the moisture present. The dried samples were ground and homogenized by using mortar and pestle and sieved using 1 mm sieve. Finally, the powdered samples were kept in an airtight container until used for analysis [4].

Extraction of *Capparis tomentosa* leaf: 200 g powdered plant sample was successively extracted with petroleum ether, ethyl acetate and methanol using maceration technique for 48 hrs in each solvent. The extract was filtered and residual solvent from each extract was removed using rotary evaporator. The resulting semidried mass of each fraction was stored in shade area until used for experiments (Figure 1).

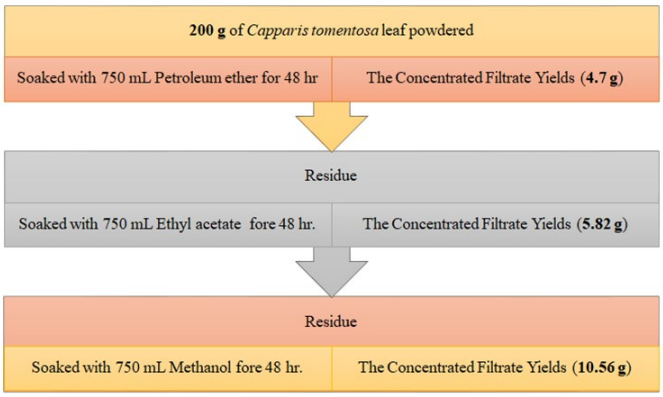

Figure 1. Flow chart showing extraction steps of the leaves of *Capparis tomentosa* plant.

Standard solution preparation

1 g of gallic acid was dissolved in 100 mL of methanol to get 1% solution of gallic acid (10 mg/mL) termed as standard 1 solution for TPC determination. Similarly 1 g of quercetin/catechin was dissolved in 100 mL of methanol separately to get 1% solution of catechin (10 mg/mL) termed as standard 2 solution for TFC determination. These standard stock solutions were used to prepare series of diluted standard solution that contained different concentration used for constructing calibration curve for each method.

Determination of Total Phenolic Content (TPC)

The total phenolic content of the crude extract was determined by

using Folin-Ciocalteu method. A standard gallic acid curve was constructed by preparing the dilutions in methanol from stock solution of gallic acid. A standard gallic acid curve was constructed by preparing the dilutions of (20, 40, 60, 80 and 100 ppm) in methanol from standard 1 solution of gallic acid. 1 mL of each of these dilutions were mixed with 5 mL of Folin-Ciocalteu reagent and allowed to stand for 6 minutes. Then 4 mL of 10% sodium carbonate were added to the reaction mixture. The absorbance was recorded after it was mixed and allowed to stand for 30 minute at 765 nm spectrometrically. The total phenolic content of the extracts of petroleum ether, ethyl acetate and methanol was calculated as gallic acid equivalents (mgGAE/g) [5].

Determination of Total Flavonoid Content (TFC)

Aluminum chloride complex forming assay was used to determine the total flavonoids content of the extracts. Catechin was used as standard and flavonoids content was determined as catechin equivalent. A calibration curve for catechin was drawn for this purpose. From the standard 2 catechin solution the dilutions of (25, 50, 100,200 and 400 ppm) concentrations were prepared in methanol. 0.25 mL of each of the catechin dilution was mixed with 1.25 mL of distilled water and then with 0.075 mL of 5% sodium nitrite and allowed to stand for 5 minutes for mixing. Then 0.15 mL of 10% aluminum chloride solution was added and allowed to stand for 6 minutes at room temperature after which 0.5 mL solution of 1 M sodium hydroxide was added sequentially. Total flavonoids content was calculated as Catechin equivalents (mg CE/g). The absorbance of this reaction mixture was recorded at 510 nm on UV-spectrophotometer Cary 60 agilent technologies [6].

Antioxidant capacity assay

DPPH radical scavenging assay: The free radical scavenging activities of the extracts was determined by using 2, 2-Diphenyl-1-Picrylhydrazyl (DPPH) free radical scavenging method. DPPH is very stable free radical. Unlike *in vitro* generated free radicals such as the hydroxyl radical and superoxide anion, DPPH has the advantage of being unaffected by certain side reactions. A freshly prepared DPPH solution exhibits a deep purple color with an absorption maximum at 517 nm. This purple color generally fades when antioxidant molecule quench DPPH free radicals yellow (*i.e.* by providing atoms or by electron donation, conceivably *via* a free-radical attack on the DPPH molecule) and convert them into a colorless/bleached product (*i.e.* 2,2-diphenyl-1-hydrazine, or a substituted analogous hydrazine), resulting in a decrease in absorbance at 517 nm band. DPPH in oxidized form gives a deep violet color in methanol to yellow. An antioxidant compound donates the electron to DPPH thus causing its reduction and in reduced from its color changes from deep violet to yellow. A fresh 0.002% solution of DPPH was prepared in methanol. 25, 50, 75, 100, and 125 ppm ascorbic acid and plant extracts were prepared and ascorbic acid used as reference. 1.5 mL of the ascorbic acid and extracts (25, 50, 75, 100, and 125 ppm) was mixed with 3 mL solution of DPPH and allowed to stand in darkness for 15 minutes. Control was prepared by taking 3 mL of DPPH and 1.5 mL methanol and its absorbance was recorded at 517 nm. The assay was performed in triplicates. Percentage inhibition of free radical DPPH was calculated based on control. The absorbance was again recorded at 517 nm. The percentage inhibition of DPPH by extracts was calculated by using equation 3 as shown in the data analysis part [7].

Ferric Reducing Antioxidant Power (FRAP) assay

The reducing power of the crude extracts was determined according to the standard method with slight modification. 2.5 mL of different concentrations (50,100,150 and 200 ppm) of plant extracts/standards was mixed with phosphate buffer solution (PH=6.6, 0.2 M) and potassium Hexacyanoferrate (K_3Fe (CN) 6) (1%). The mixture was incubated at 50°C for 20 min in water bath. Then Trichloroacetic acid (10%) was added to the mixture to terminate the reaction. 4 mLof the reaction mixture was mixed with 5 mL of distilled water and 0.5 mL of $FeCl_3$ solution (0.1%). The reaction mixture was incubated for 10 minutes at room temperature and the absorbance was measured at 700 nm by using UV-spectrophotometer against a blank solution [8].

Method of data analysis: The total phenolic content of the extracts of petroleum ether, ethyl acetate and methanol was calculated as gallic acid equivalents (mgGAE/g) by using the formula given below.

$$T=C \times V/M \quad (1)$$

Where;

T is the total phenolic contents in mg/g of the extracts as GAE.

C is the concentration of gallic acid established from the calibration curve in ppm.

V is the volume of the extract solution in mL and M is the weight of the extract in g.

The total flavonoid content of the extracts of petroleum ether, ethyl acetate and methanol was calculated as catechin equivalents (mg CE/g) as the formula given below:

$$T=C \times V/M \quad (2)$$

Where;

T is the total flavonoid contents in mg/g of the extracts as CE.

C is the concentration of catechin established from the calibration curve in ppm.

V is the volume of the extract solution in mL.

M is the weight of the extract in g.

The free radical scavenging activity of each extract and the standard ascorbic acid was calculated by the following formula from the absorbance at 517 nm. The percentage scavenging activity of DPPH was calculated by using equation 3.

$$\% \text{ scavenging activity} = ((A-B)/A) \times 100 \quad (3)$$

Where;

A is the absorbance of pure DPPH in oxidized form.

B is the absorbance of sample taken after 30 minutes of reaction with DPPH.

Ferric Reducing Antioxidant Power (FRAP) assay were also calculated by using equation 4.

$$\%\text{Reduction power} = (\text{Asample-lank})/\text{Asample} \times 100 \quad (4)$$

Where: Asample=Absorbance of sample

A blank=Absorbance of blank

Statistical data analysis

The statistics for antioxidant sports had been expressed because the common of 3 measurements and all of the final statistics had been expressed as mean ± standard deviations of triplicates, the usage of MS Excel 2010, origin eight software [9].

Results and Discussion

The finely divided powder of the leaf of *Capparis tomentosa* (200 g) was subjected to successive extraction by petroleum ether, ethyl acetate and methanol respectively. The extracts were analyzed for total phenolic, total flavonoid, antioxidant (Ferric Reducing Antioxidant Power (FRAP) and antioxidant activities by (DPPH). A number of assays were conducted to analyze antioxidant activities of the extracts since antioxidant activities of different types of substances involved different mechanisms.

Yield of the extracts of *Capparis tomentosa*

Successive extraction two hundred g leaf powder of *Capparis*

tomentosa gave the very best yield with methanol (10.5 g) accompanied with the aid of using ethyl acetate (5.82 g) and petroleum ether (4.709 g) respectively (Table 1).

Solvent system	Mass of extracts (g)	Percent yield (%)
Etroleum ether	4.7	2.352
Thyl acetate	5.82	2.91
Ethanol	10.56	5.28

Table 1. Yields of each extract.

Quantitative analysis for total phenolic and total flavonoid contents

Total phenolic contents: Five mg of every extract of *Capparis tomentosa* plant became weighed and dissolved in a separate 100 mL (distilled water) volumetric flask. 1 mL of prepared solution was then taken into three different test tubes and combined with 5 mL of distilled water after which 0.5 mL of folinciocalteu reagent became introduced. After five min 1.5 mL of Na_2CO_3 answer became introduced and the extent became mad up to 8 mL with distilled water and ultimately the consequent solution became incubated for 60 min in room temperature and the absorbance became measured at 765 nm. A collection of diluted solution (20, 40, 60, 80, and 100 ppm) of widespread gallic acid has been organized from the inventory solution through the use of the subsequent dilution procedure [10].

After guidance of the solution the absorbance became measured at a wavelength of 765 nm through the use of UV-VIS spectroscopy. The instrument gave records in spectra of absorbance vs. wavelength (nm). The absorbance at a wavelength of 765 nm became used to devise calibration curve of absorbance *vs.* concentration (Figure 2). From this calibration curve the unknown concentration of the extract was determined through the use of the equation derived from the standard. The results have been given in the following (Table 2).

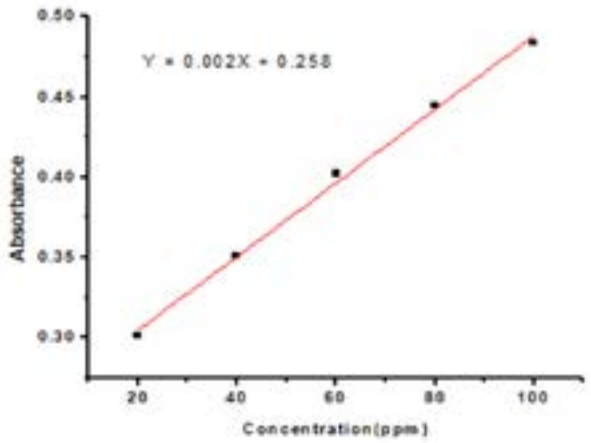

Figure 2. Gallic acid standard calibration curve for TPC determination.

Concentration (ppm)	Absorbance (Mean)
	λ_{max}=765 nm
20	0.30107
40	0.38096
60	0.41452
80	0.44488
100	0.47364

Table 2. Absorbance of standard compound (gallic acid).

From the graph of gallic acid standard calibration curve the following equation was derived. TPC of the tested plant extracts become determined by the use of the Folin-Ciocalteu reagent). The result was expressed in terms of Gallic acid equivalent (the standard curve equation: Y=0.002x+0.258, R^2=0.9951). The TPC of the extracts of *Capparis tomentosa* was evaluated and the results were presented in the following Table 3. The TPC result was ranged from 53.93 to 195.97 mg GAE/g. The highest concentration of phenolic compound was found in methanol and ethyl acetate extracts but not on petroleum ether extracts. According to the previous reports, high solubility of phenols in polar solvents provides high concentration of phenolic compounds in the extracts obtained using polar solvents of the target plant the extracts. The volume of the each plant extract used (10 mL).

No	Solvent system	Absorbance at 765nm	Total Phenol Content (TPC)
1	Methanol extract	0.45397	195.97 ± 0.94
2	Ethyl acetate extract	0.3727	114.3 ± 1.61
3	Petroleum ether extract	0.31193	53.3 ± 0.67

Table 3. Total phenolic content of *capparis tomentosa.*

Total flavonoid content: Aluminum chloride complex forming assay was used to determine the total flavonoid content of the extracts. Catechin was used as standard to determine TFC as catechin equivalents. The standard solution catechin was prepared from the stock solution (solution 2 in procedure 2.6.3.3) by dilutions of it with distilled water (25, 50, 100, 200 and 400 ppm) concentrations were prepared (Table 4). After performing all the procedure described before, the absorbance was measured at a wavelength of 510 nm with their respective concentration (ppm) (Figures 3 and 4) [11].

No.	Concentration (ppm)	Absorbance at
		λ_{max}=510 nm
1	25	0.023
2	50	0.062
3	100	0.123
4	200	0.251
5	400	0.51

Table 4. Absorbance of standard compound (*Catechin*).

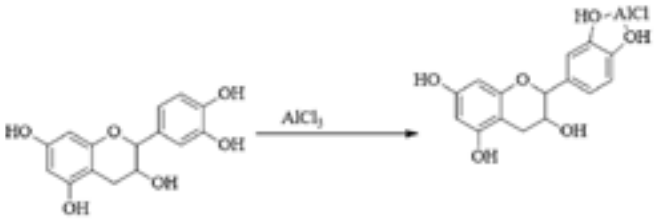

Figure 3. Complexation reaction of catechin (reference compound) with aluminum chloride.

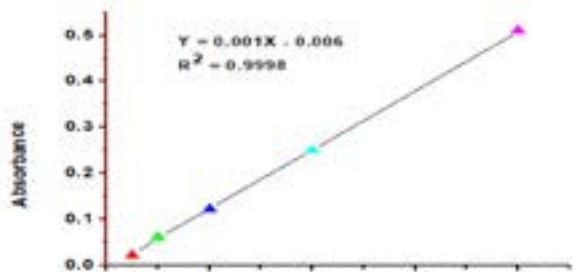

Figure 4. Absorbance *vs.* concentration calibration curve of catechin standard to determine TFC in (mg CE/g).

From the graph of catechin standard the following calibration curve equation was derived and from this equation the value of C(X) was calculated and TFC in (mg CE/g) was also analyzed. The absorbance of standard catechin at a wavelength of 510 nm was used for calculating the unknown concentration of the extract sample for determining total phenolic content as the result given in the Table 5 below.

No	Solvent system	Mean Absorbance at 510 nm	Total Flavonoid Content (TFC)
1	Petroleum ether	0.0232	58.400 ± 1.89
2	Ethyl acetate	0.041886	95.772 ± 2.31
3	Methanol	0.081552	175.104 ± 1.47

Table 5. Total flavonoid content of *C. tomentosa* leaf extract.

The concentration of flavonoids in extracts of the *Capparis tomentosa* was determined by using spectrophotometric method with aluminum chloride. The standard calibration curve equation that was used in calculation was (y=0.001 × -0.006, R^2=0.9998). The concentration of flavonoids in the extracts ranged from 58.4 to 175.104 mg CE/g. Methanol extract contained the highest flavonoid concentration whereas the lowest flavonoid concentration was found in petroleum ether extract. The concentration of flavonoids in plant extracts depends on the polarity of solvents used in the extract preparation. The Total Flavonoid Content (TFC) of the different extracts of *Capparis tomentosa* was determined in terms of catechin equivalents (mg CE/g), and the results were shown in Table 6 below. Methanol extract had the highest TFC followed by ethyl acetate extract and the petroleum ether extract had the least the TFC value. As mentioned above three solvent system extracts of *Capparis tomentosa* leaf were tested for the determination of total phenols and total flavonoids contents. The total phenolic content of all the three extracts varied from 53.93 ± 0.67 to 195.97 ± 0.94 mg/g of gallic acid equivalent [12].

Solvent system	Total phenolic content (mg/g) of GAE	Total flavonoid content (mg/g) of CE
Methanol extract	195.97 ± 0.94	175.104 ± 1.47
Ethyl acetate extract	114.3 ± 1.61	95.772 ± 2.31
Pet ether extract	53.93 ± 0.67	58.4 ± 1.89

Table 6. Total Phenolic and Flavonoid content of methanol, ethyl acetate and petroleum ether extracts of *C. tomentosa* leaf.

Methanol extract contained the best flavonoid contents while the petroleum ether extract showed the lowest flavonoid contents. The total flavonoid content of the three solvent extract of capparis tomentosa leaf tested varied from 175.104 ± 1.04 to 58.4 ± 1.89 mg/g of Catechin equivalent. Maximum flavonoids were found in methanol extract. Table 7 showed the comparison of TPC and TFC of *Capparis tomentosa* plant extract.

No	Concentration in ppm	Absorbance at 517 nm			
		Ascorbic acid	Methanol extract	Ethyl acetate extract	Petroleum ether extract
1	25	0.03547	0.23727	0.54185	0.59964
2	50	0.0145	0.18706	0.43088	0.5461
3	75	0.00436	0.14104	0.33541	0.38766
4	100	0.00178	0.08907	0.24443	0.30477
5	125	0.00048	0.05791	0.1072	0.13328

Table 7. Absorbance and concentration of Std. and *C. tomentosa* leaf extract on DPPH.

Antioxidant capacity assay

DPPH radical scavenging activity: Antioxidants are the additives of the plant life which can be able to enacting the visually significant quenching of the purple-colored DPPH radical to the yellow-colored DPPH. The impact of an antioxidant on DPPH radical scavenging is because of their hydrogen donating potential or radical scavenging activity. When an answer of DPPH is blended with that of a substance which could donate hydrogen atom, then this offers upward thrust to the decreased shape DPPH with the lack of its deep violet color at 517 nm. When an answer of DPPH is blended with that of a substance which could donate hydrogen atom, then this offers upward thrust to the decreased shape DPPH with the lack of its deep violet color at 517 nm This color step by step modified whilst combined with *Capparis tomentosa* plant extract with recognized concentration. After addition of 1 mL of DPPH strategy to 3 mL of every extract plant pattern and the same old ascorbic acid the color of DPPH answer became modified to mild yellow color. The absorbance of widespread and every extracts of *Capparis tomentosa* have been recorded at 517 nm as proven with inside the desk nine below [13].

By using this formula percentage scavenging activity of standard and *C. tomentosa* leaf extract on DPPH was calculated and tabulated in the below. The DPPH radical scavenging activities of selected medicinal plants were presented in Table 8 below. All the plant extracts showed concentration dependent increases in radical scavenging capacity. The greatest DPPH radical scavenging potency was recorded for methanol extract of C. tomentosa (90.38%), followed by ethyl acetate extract (82.193%) and petroleum ether extract (77.86%) at a concentration of 125 ppm but the standard ascorbic acid outweigh all of the extracts with their respective concentration (99.92%). Among the three plant extract methanol extract of *Capparis tomentosa* showed the highest antioxidant activity next to the standard ascorbic acid antioxidant activities. Ethyl acetate extract had also the higher antioxidant activity as compared to petroleum ether extract. Petroleum ether extract had the lowest antioxidant activity as compared with methanol and ethyl acetate extract of *Capparis tomentosa* leaf.

No	Concentration in ppm	% of Inhibition			
		Ascorbic acid	Methanol extract	Ethyl acetate extract	Petroleum ether extract
1	25	94.108	60.586	9.992	0.392
2	50	97.591	68.927	28.425	9.286
3	75	99.276	76.571	44.284	35.605
4	100	99.704	85.204	59.397	49.374
5	125	99.92	90.38	82.193	77.86

Table 8. Percentage scavenging activity and concentration of Std. and *C. tomentosa* leaves extract (Abs control=0.601) for DPPH.

This was confirmed by quantitative test which informs as methanol extract had a high content of phenolic compound which intern supported by total phenolic content as calculated above (195.97 as expressed in mg/g of gallic acid equivalence) while the other extract ethyl acetate and petroleum ether has TPC of 114.3 and 53.93 mg/g of gallic acid respectively in which their antioxidant activity is lower than methanol extract and the standard ascorbic acid. The previous report on the root part of this plant were also had the following results The radical scavenging activity of the *C. tomentosa* roots, according to the DPPH method was found to be 35.50 ± 0.02% compared to the ascorbic acid pure standard which had 96.50 ± 0.02% at concentration of 1000 ppm. This was lower than the current research conducted in the leaf part of *Capparis tomentosa* as the result mentioned above [14]. On the other hand *C. tomentosa* leaf proven brilliant scavenging hobby. The methanol extract had the better scavenging capability than aqueous extract. The maximum quantity of loose radical scavenging hobby changed into observed with inside the methanol plant extracts in comparison to the aqueous extract. Compared to petroleum ether extract,

methanol extracts and ethyl acetate extract of *C. tomentosa* confirmed better diploma of loose radical scavenging capability. The extracts of *Capparis tomentosa* proven brilliant radical scavenging capability. The end result suggests that the extract may be used for prevention of degenerative sicknesses which includes Parkinson's, Alzheimer's, cancer, diabetes, coronary heart disease, arthritis, osteoporosis etc. They additionally offer crucial fitness advantages with inside the shape of antioxidant hobby through stopping aging. This excessive scavenging capability is attributed to the phytochemicals gift with inside the plant: Alkaloids, anthranoids, flavonoids, glycoside, polyphenols, saponins and steroids. The medicinal movements of polyphenols are generally attributed to their antioxidant capability which performs a great position in, loose radical scavenging, chelation of redox lively steel ions, modulation of gene expression and interplay with the mobileular signaling pathways. Other file in this plant changed into additionally deceived a barely better loose radical scavenging hobby of the methanol extract possibly indicated the presence of a better content material of exceptionally polar flavonoids and phenols with inside the methanol than the ethyl acetate and petroleum ether extracts, facilitating hydrogen atom switch to take place. These entire file confirms the present day have a look at on *Capparis tomentosa* plant [15].

Ferric Reducing Antioxidant Power (FRAP) assay

The reducing power of the crude extracts was determined according to the standard method, using ascorbic acid as a standard. A series of diluted solution (50, 100, 150 and 200 ppm) of standard and *Capparis tomentosa* leaf extracts were prepared based on the procedure described before. After preparing the solution the absorbance were measured for each plant extract and standard at 700 nm. Table 9 gave the absorbance of the three solvent system extract (methanol, Ethyl acetate and petroleum ether) and the standard ascorbic acid at a wavelength of 700 nm.

No	Concentration in ppm	Absorbance at 700 nm			
		Ascorbic acid	Methanol	Ethyl acetate extract	Petroleum ether
1	50	0.2521	0.2236	0.1938	0.1562
2	100	0.4352	0.343	0.29375	0.2553
3	150	0.731	0.4632	0.42962	0.39243
4	200	0.9812	0.68926	0.54619	0.4237

Table 9. Absorbance and concentration of ascorbic acid standard and the three extracts of *C. tomentosa.*

The below showed that the absorbance of each extract and standard increase as the concentration of each extract and standard increases. 200 ppm had the highest absorbance in all cases of extracts and standards. The standard ascorbic acid had the highest absorbance and so have the highest antioxidant activity. Methanol Extract (ME) showed the highest absorbance as compared with the other solvent extract of *C. tomentosa* leaf and so it had pronounced antioxidant activity than ethyl acetate and petroleum ether extracts while petroleum ether extract of *C. tomentosa* had the lowest absorption in all the cases and so showed the least antioxidant activity. This is due to the amount of phenolic and flavonoid content present in each extracts, because of methanol extract had the highest content of TPC and TFC while petroleum ether had the lowest TPC and TFC value. Ferric Reducing Antioxidant Power (FRAP) assay was calculated and tabulated as follows.

Reducing power (antioxidant capacity) of methanol, ethyl acetate and petroleum ether extracts of *Capparis tomentosa* leaf was determined and the results were shown in the Table 10 below. The methanol extract was displayed the highest reducing power whereas the ethyl acetate was moderate and petroleum ether was the lowest reducing power. All three extracts showed almost similar increasing trend in reducing power with the increase in extracts concentration. In this assay, the presence of reducers (*i.e.*, antioxidants) causes the reduction of the Fe^{3+} ferric cyanide complex to the ferrous form. Therefore, measuring the formation of Perl's Prussian blue at 700 nm can monitor the Fe^{2+} concentration. The methanol extract which had the highest TPC and TFC, also displayed the highest reducing power. The correspondence supported to the fact that phenolic and flavonoids possess high antioxidant potential [16].

No	Absorbance at 700 nm				
	Concentration (ppm)	AA	ME	EA	PE
1	50	60.559	55.532	48.694	36.34
2	100	77.153	71.012	66.151	61.053
3	150	86.398	78.534	76.856	74.66
4	200	89.866	85.574	80.552	76.533

Table 10. Percentage reduction of power standard and *tomentosa* extract.

Other report on this plant root part using methanol and water as a solvent was compared with the current study. It was observed that the reducing power of the aqueous roots extracts of *C. tomentosa* increased with concentration. This could be explained by the fact that the extracts contained chemical substances capable of reacting with potassium ferric cyanide (Fe^{3+}) to form potassium ferric cyanide (Fe^{2+}), which then reacts with ferric chloride to form ferric ferrous complex that has an absorption maximum at 700 nm. The observation that the reducing power was linearly proportional to the concentration is explained by the fact that the amounts of antioxidants increased with an increase in the amount of crude extracts. Reducing power is associated with antioxidant activity and indicates that primary and secondary antioxidants with ability to donate electrons and reduce oxidized intermediates of lipid peroxidation processes are present in the extracts. In the assay, the yellow color of the test solutions changes to various shades of green and blue and this depend on the reducing power of the extracts concentrations. Similarly, the radical scavenging activity by DPPH assay indicates that the roots extract contains antioxidant compounds. All the previous report written above was support the results of the current study [17].

Conclusion

The powdered leaf of *Capparis tomentosa* was subjected to successive extraction with petroleum ether, ethyl acetate and methanol respectively and the solvent were removed by Rotary evaporators (40°C-60°C). Phytochemical works were then conducted on the three extract sample of the leaf of *Capparis tomentosa*. The results of phytochemical work on the extract revealed the presence of alkaloids, phenols, flavonoids, tannins, terpenes, saponins, and glycosides on the extract sample. All of the extracts (petroleum ether, ethyl acetate and methanol) of the leaf part of *Capparis tomentosa* possess antioxidant activity, evidenced by the free radical scavenging property (DPPH) and Ferric Reducing Antioxidant Power property (FRAP), which may be due to the presence of phenolic and flavonoid components in the extracts. It is noticed that the highest concentration of phenolic and flavonoid compounds in the extracts were obtained using solvents of high polarity (methanol and ethyl acetate extract) manifested greater power of phenolic and flavonoid compounds from the leaf part of *Capparis tomentosa*. Methanol extract had a high contents of phenolic and flavonoid compounds (195.97 mgGAE/g TPC and 175.104 mgCE/g TFC) and so methanol extract had highest antioxidant activity as compared to the other solvent extract and petroleum ether extract have the lowest TPC and TFC and so had the lowest antioxidant activity. Significant linear correlation between the values of the concentration of phenolic and flavonoid compounds and antioxidant activity indicated that these compounds contribute to the strong antioxidant activity. Methanol extract had the highest antioxidant activity next to the standard as compared to ethyl acetate and petroleum ether extract.

Recommendations

- Isolation and characterization of bioactive compounds on the crude extracts of the stems, flowers, root, leaf and fruits of Capparis tomentosa plant using different solvent system should be done.
- Anti-microbial studies by using other kinds of bacteria and fungi on the crude extracts and pure compounds should be done.
- Determination of the cytotoxity level of the extracts from all parts of the plant should also be done.

Authors' Contribution

HGA was performed the whole work as well as prepare the manuscript and the remaining Co-authors were helping HGA in doing laboratory experiment and give the necessary feedback for edition. All authors contribute for manuscript finalization. All authors were read and approve the final manuscript.

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Competing Interest

All the authors declare that they have no competing interests

Consent for Publication

All author consent to the publication.

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