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CHEMICAL CHARACTERIZATION AND ANTIOXIDANT EVALUATION OF *OLDENLANDIA PUMILA* (L.F.) DC: A COMPREHENSIVE STUDY

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Keywords:

Oldenlandia pumila (L.f.) DC., Phytochemical analysis, Total phenolic content, Total flavonoid content, Antioxidant assay

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ABSTRACT: Medicinal plants are presently viewed as being of great value because of their unique qualities as a significant source of medicinal phytochemicals that might result in the creation of new medications. *Oldenlandia pumila* (L.f.) DC. belong to Rubiaceae family and found as a weed throughout India. *Oldenlandia pumila* (L.f.) DC. has antibacterial, antiviral, antioxidant, anticancer, anti-inflammatory, and antiplasmodial activities. The effects of three different solvents were investigated in the current study to determine the presence of phytochemicals, total phenolic content, total flavonoid content, and antioxidant assay. The preliminary phytochemical analysis revealed the presence of carbohydrates, flavonoids, tannins, and alkaloids whereas finding the absence of proteins and glycosides in the various extracts. Phytochemical analysis showed that *Oldenlandia pumila* (L.f.) DC. contained TPC ranging from (82.235 ± 0.005 mg to 424 ± 0.005 mg GAE /g) while TFC ranging from (38 ± 0.006 mg to 336 ± 0.006 mg GAE /g). The TAC ranges from (18.285 ± 0.007 to 89 ± 0.006 mg AE/g) and the TAC ranging from (64.8 ± 0.003 mg to 163 ± 0.004 mg TAE/g). In the DPPH assay, the aqueous extract of stem has the greatest IC_{50} value and lowest acetone extract of stem. According to the FRAP assay, aqueous extract of leaves had the lowest concentration, whereas methanol extract of leaves had the highest. The present study covers significant pharmacological research on *Oldenlandia pumila (L.f.) DC*. and phytochemical investigations that might be further studied to reach lead molecules in the quest for innovative herbal medications.

INTRODUCTION: Nature is usually an excellent illustration of the obvious cohabitation phenomenon. Natural goods derived from plants, animals, and minerals serve as the foundation for treating human ailments [1]. The word medicinal plant refers to a wide range of plants with medical characteristics.

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These plants are a rich source of chemicals that can be used in medication synthesis [2]. Different kinds of seeds, roots, leaves, fruits, flowers and even the entire plant can be utilized as a source of medicine [3]. The active chemicals in most medicinal plants have direct or indirect therapeutic effects and are used as medicines [4].

These plant bodies create, and store components known as active compounds (substances), which have physiological impacts on organisms [5]. Plants are a great source of secondary metabolites including phenolic and flavonoid chemicals, which are often used as antioxidants with redox and metal

chelating capabilities [6]. Antioxidants are chemical substances that have the capacity to decrease free radicals and their rate of formation as well as lipid peroxidation in human bodies, which cause a variety of human illnesses and ageing [7].

Hedyotis and *Oldenlandia* are two of the largest genera in the Rubiaceae family, with a total of around 515 species that are found across the tropics, particularly the entire world. The genus is distinguished by its septicidal dehiscing capsules and perennial plants or shrubs [8-9]. *Oldenlandia pumila* (L.f.) DC. is native plant to Andaman Islands, Bangladesh, India, Jawa, Malaya, Myanmar, Sri Lanka, Thailand, Vietnam [10]. *Oldenlandia pumila* has small white flowers with yellow centers, and small, black, glossy seeds. The seedlings are small and thin with long, narrow leaves. *Oldenlandia pumila* is used as an ornamental plant in gardens and as a groundcover. It is also used in traditional medicine to treat fever, cough, and other ailments [11].

In the current investigation, the effect of three different solvents (Methanol, Acetone, Aqueous) were examined in order to evaluate the presence of phytochemicals, total phenolic content, total flavonoid content, and antioxidant assay.

MATERIAL AND METHOD:

Plant Material: The *Oldenlandia pumila* (L.f.) DC. plant was collected from Law Garden in the month of January Ahmedabad, Gujarat. Fresh leaves and stem of *Oldenlandia pumila* (L.f.) DC. was used as a material for the experiment.

Preparation of Sample: The plant material (Leaves & Stem) was dried at room temperature until all water molecules vanished. Following drying, the plant material was finely ground using a mechanical blender into a powder then kept into airtight containers for later usage.

Preparation of Extraction: The plant material (Leaves and Stem) is extracted using methanol, acetone and aqueous solvent by the Soxhlet extraction method.

Qualitative analysis:

Test for Alkaloids: A small amount of the crude extract was individually dissolved in dilute hydrochloric acid and then filtered.

Mayer's Test: 1 ml of filtrate was taken and add 1 ml Mayer's reagent. Yellow colour precipitates indicate the presence of alkaloids.

Wagner's Test: 1 ml of filtrate was taken and add 1 ml Wagner's reagent. Brown/reddish colour precipitates indicate the presence of alkaloids.

Hager's Test: 1 ml of filtrate was taken and treated with 1 ml Hager's reagent. Yellow colour precipitates indicate the presence of alkaloids.

Dregendroff Test: 1 ml of filtrate was taken and add 1 ml Dregendroff reagent. The presence of orange colour precipitates indicates the presence of alkaloids.

Test for Carbohydrates: Crude extracts were dissolved in 5 ml distilled water and then filter it. Filtrate is used as a test solution.

Molish Test: Take 1 ml of extract and add 1 ml Molish reagent into it and shake it well. Then add 1 ml of concentrated sulphuric acid side by side. Red violates ring indicates the presence of carbohydrate.

Fehling Test: 2 ml filtrate were hydrolyzed with 1 ml diluted hydrochloric acid (1 N) and neutralize with 1ml alkaline solution (10% NaOH). Then heat in water bath and then add Fehling A & B solution. The formation of red precipitation indicates the presence of carbohydrates.

Barford's Test: Take 1 ml of filtrate and add 1 ml Barford's reagent. Boil the mixture for 2 min. The formation of red ppts indicates the presence of carbohydrates.

Bendict's Test: Take 1 ml of filtrate and add 1 ml Bendict's reagent. The mixture is heated in a boiling water bath for 2 min. The formation of orange red ppts indicates the presence of reducing sugar.

Test for Glycosides:

Borntrager's Test: Take 2 ml of extract and add 3 ml of chloroform and shake it well. Now add a few drops of ammonium solution. The pink color indicates the presence of glycosides.

Legal's Test: To 1 ml of extract add 1 ml of sodium nitroprussides and 1 ml of pyridine.

The formation of pink or red colour indicates the presence of cardiac glycoside.

Keller-Killiani Test: To 2 ml of test solution add 2-3 drops of glacial acetic acid and add 1%ferric chloride mixed with concentrated sulphuric acid. Formation of two-layer lower reddish brown and upper bluish green.

Test for Proteins

Millon's Test (Mercuric Nitrate Solution): Take 2 ml of extract and add 1-2 ml of Million's reagent. White ppts indicate the presence of proteins.

Biuret Test: Take 2 ml of extract and add 0.5 ml of $CuSO_4$, 1 ml of ethanol, 1 KOH pellet. The pink colour of the ethanolic layer indicates the presence of proteins.

Test for Phenols

Ferric Chloride Test: To the extract treated with 2 ml of ferric chloride solution. Formation of blue/black colour ppts indicate the presence of phenols.

Lead acetate Test: 1 ml of extract add 0.5 ml of 10% lead acetate solution. The formation of white precipitates indicates the presence of phenols in extract.

Folin-Cioculten Test: 0.5 ml of extract add 1 ml of Folin-Cioculten solution. The formation of bluish green precipitates indicates the presence of phenols in extract.

Test for Flavonoids

Alkaline Test: Take 1 ml of extract and add 1 ml of 10% sodium hydroxide solution. The formation of intense yellow colour, which disappeared after addition of 2 ml concentrated sulphuric acid, indicates the presence of flavonoids.

Lead acetate Test: To 1 ml of extract add 1 ml of 10% lead acetate solution. Formation of yellow precipitates indicates the presence of flavonoids in extract.

Test for Saponin:

Froth Test: Few mg extract diluted with 5ml in distilled water and shake it well for 15 min. Formation of 1 cm of foam indicates that saponin is present.

Test for Terpenoids

Salkowski Test: Take 1 ml of extract and then add 2 ml chloroform & 3 ml of concentrated sulphuric acid. The appearance of reddish brown colour indicates the presence of terpenoids.

Copper Acetate Test: To 1 ml of extract add 1 ml of 5% copper acetate solution. Green color precipitates indicate the presence of terpenoids.

Test for Fixed oils and Fats:

Filtered Paper Test: Take a small amount of extract and press it between the filtered paper. Leave oil strain on the filtered paper. The oil strain indicates the presence of fixed oils and fats.

Test for Cardiac Glycosides:

Legal Test: Take 1 ml of extract and add 1 ml of pyridine and 1 ml of 20% sodium nitroprusside. Pink or red colour indicates the presence of cardiac glycosides.

Test for Phytosterols

Liebermann Burchard's Test: Take 1 ml of extract and add 2-3 ml of acetic anhydride solution and 2 ml of conc. sulphuric acid. Violet or green colour indicates the presence of phytosterols.

Salkowski Test: Take 1 ml of extract and then add 2 ml chloroform & 3 ml of concentrated sulphuric acid. The appearance of reddish brown colour indicates the presence of phytosterols.

Estimation of Total Phenol Content: One ml of the extracts of different parts of plants were thoroughly mixed with 10 ml of distilled water, 1.5 ml of Folin-Ciocalteu reagent added. After 5 minutes, 4 ml of 20% sodium carbonate (Na2CO3) was added and adjusted with distilled water up to 25 ml and agitated.

Then incubated for 30 minutes at room temperature. The absorbance was measured at 765 nm against a blank having all the reagents excluding the sample using spectrophotometer. This procedure was repeated 3 times for each extract. The total phenols were quantified by the standard curve of gallic acid solution which was prepared using the similar procedure [12].

Estimation of Total Flavonoid Content: 500 µl of the extract of different parts of plants were mixed

with 1500 µl of 95% methanol and then 100 µl of Aluminum chloride (10%) and Potassium acetate (1M) was added respectively and make volume up to 10ml with distilled water and agitated. Incubation was done for 20-30 minutes at room temperature. The absorbance was assessed at 415 nm against a blank having all the reagent without the sample using spectrophotometer. Measurement was done in triplicates and the total flavonoid quantified by the standard curve of quantified by the standard curve of quercetin solution [13].

Estimation of Total Tannins Content: The tannins were determined by Folin-Ciocalteu method. About 0.1 ml of the sample extract was added to a volumetric flask (10 ml) containing 7.5 ml of distilled water and 0.5 ml of Folin-Ciocalteu phenol reagent, 1 ml of 35% sodium carbonate solution and dilute to 10 ml with distilled water.

The mixture was shaken well and kept at room temperature for 30 minutes. The reference standard solutions of tannic acid were prepared in the same manner as described earlier. Absorbance for test and standard solutions were measured against the blank at 700 nm with an UV/ Visible spectrophotometer. The estimation of the tannin content was carried out in triplicate. The tannin content was expressed in terms of mg of tannic acid equivalents/ g of dried sample. Tannic acid is used as standard [14].

Estimation of Total Alkaloid Content: The plant extract (1mg) was dissolved in dimethyl sulphoxide (DMSO), added 1ml of 2 N HCl and filtered. This solution was transferred to a separating funnel, 5 ml of bromocresol green solution and 5 ml of phosphate buffer were added. The mixture was shaken with 4-ml chloroform by vigorous shaking and collected in a 10-ml volumetric flask and diluted to the volume with chloroform. A set of reference standard solutions of atropine were prepared in the same manner as described earlier. The absorbance for test and standard solutions were determined against the blank reagent at 470 nm with an UV/Visible spectrophotometer. The total alkaloid content was expressed as mg of AE/g of extract [15].

Antioxidant Activity:

DPPH Assay (2, 2-diphenyl-1-picrylhydrazyl): The radical scavenging activity of different extracts was determined by using DPPH assay according to (Chang *et al.* 2001). The decrease in the absorption of the DPPH solution after the addition of an antioxidant was measured at 517nm. Ascorbic acid (1mg/ml) was used as reference.

Different volumes of plant extracts were made up to 1ml with respected solvent and then 1ml DPPH (0.1mM) solution was added. The reaction mixture was incubated in dark condition at room temperature for 20 min. After 20 min, the absorbance of the mixture was read at 517 nm. 3ml of DPPH was taken as control [17].

The % radical scavenging activity of the plant extracts was calculated using the following formula:

$$\% \text{ RSA} = (\text{Abs control} - \text{Abs sample}) / (\text{Abs control}) \times 100$$

Where, RSA is the Radical Scavenging Activity, Abs control is the absorbance of DPPH radical + ethanol; Abs sample is the absorbance of DPPH radical + plant extract.

Ferric Reducing Antioxidant Power Assay: The determination of the total antioxidant activity (FRAP assay) in the extract is a modified method of Benzie and Strain (1996). The stock solutions included 300 mM acetate buffer (0.3 M acetic acid and 0.3 M sodium acetate) having the 3.6 pH. 10 mM TPTZ (2, 4, 6-tripyridyl-s-triazine) solution was prepared in 40 mM HCl and 20 mM Ferric chloride ($FeCl .6H_2O$) solution.

The fresh working solution was prepared by mixing 25 mL acetate buffer, 2.5 mL TPTZ and 2.5 mL $FeCl.6H_2O$. The temperature of the solution was raised to 37°C before use. Plant extracts were allowed to react with the FRAP solution for 30 min in the dark condition.

Observation of the colored products (ferrous tripyridyltriazine complex) were taken at wavelength 593 nm. The standard curve was ranging between 100-1000 µM $FeSO_4$solution which is linear curve. Results are expressed in mM Fe (II) g dry mass.

RESULT AND DISSCUSSION:

Phytochemical Screening of Different Extract of *Oldenlandia pumila* (L.f.) DC.: Secondary metabolites are responsible for pharmacological

activities present in the plant. The dried extract of leaves and stem of *Oldenlandia pumila* (L.f.) DC. are utilized for qualitative analysis. The qualitative phytochemicals screening assay of *Oldenlandia pumila* (L.f.) DC different phytochemicals such as alkaloids, phenols, flavonoids, tannins, steroids, and carbohydrates are present while glycosides, proteins and cardiac glycosides are absent.

TABLE 1: PHYTOCHEMICAL ANALYSIS IN DIFFERENT SOLVENT OF *OLDENLANDIA PUMILA*

Sr. no	Phytochemical Test	Methanol		Acetone		Aqueous	
		Leaves	Stem	Leaves	Stem	Leaves	Stem
Alkaloids							
1	Mayer's test	+	+	+	+	+	+
2	Wager's test	+	+	+	+	+	+
3	Hager's test	+	+	+	+	+	+
4	Dregendroff 's test	+	+	+	+	+	+
Carbohydrates							
1	Molish test	+	+	+	+	+	+
2	Fehling test	+	+	+	+	+	+
3	Barford's test	+	+	+	+	+	+
4	Bendict's test	+	+	+	+	+	+
Glycosides							
1	Borntrager's test	-	-	-	-	-	-
2	Chloroform test	-	-	-	-	-	-
3	Keller-Killiani test	-	-	-	-	-	-
Protein							
1	Millon's test	-	-	-	-	-	-
2	Biuret test	-	-	-	-	-	-
Phenolics							
1	Ferric chloride test	+	+	+	+	+	+
2	Lead acetate test	+	+	+	+	+	+
3	Folin- Cioculten test	+	+	+	+	+	+
Flavonoids							
1	Alkaline test	+	+	+	+	+	+
2	Lead acetate test	+	+	+	+	+	+
Saponins							
1	Froth test	+	+	+	+	+	+
Terpenoids							
1	Salkowski test	+	+	+	+	+	+
2	Copper acetate test	+	+	+	+	+	+
Fixed oils & fats							
1	Filtered paper test	+	+	+	+	+	+
Cardiac Glycosides							
1	Legal test	-	-	-	-	-	-
Steroids							
1	Liebermann Burrchard's test	+	+	+	+	+	+
2	Salkowaski's test	+	+	+	+	+	+

Total Phenol Content: Plants possess phenolic chemicals, which are responsible for various pharmacological activities. Thus, estimation of the phenolic compounds evaluates the potential of the activities [18].

The phenolic content of plant ranged from 82.235 µg to 424 µg of GAE/g of dry extract. The methanolic extract of stem has high phenolic content (564 µg GAE/g) while the leaves have lowest concentration (82.235 µg GAE/g) in acetone extract. The stem contains 187.5 µg GAE/g and 168.70 µg GAE/g in aqueous and acetone extract. The methanolic extract of leaves has the highest total phenol content (424 ± 0.005 µg GAE/g) whereas the acetone extract has the lowest (82.235 ± 0.005GAE/g).

The methanolic extract (564 ± 0.004 GAE/g) has the greatest total phenol content in the stem, whereas acetone extract (168.70 ± 0.008 GAE/g) has the lowest.

TABLE 2: TOTAL PHENOL CONTENT OF VARIOUS PLANT EXTRACT OF *OLDENLANDIA PUMILA*

Sr. no.	Total Phenol Content					
	Methanol		Acetone		Aqueous	
	Leaves	Stem	Leaves	Stem	Leaves	Stem
1	424 ± 0.005	564 ± 0.004	82.235 ± 0.005	168.70 ± 0.008	103.05 ± 0.008	187.5 ± 0.006

Note: - Mean ± Standard deviation.

Total Flavonoid Content: The total flavonoid content in plant extracts was measured using the Aluminum chloride technique. Aluminum chloride may be used to measure the total flavonoid content in plant extract because it forms a colored complex with the C4 ke to group or the C3/C5 hydroxyl group of flavonoids. This solution can be analyzed using a spectrophotometer at 415 nm [19]. The plant's flavonoid content varied from 38.12 µg to 336.00 µg of QE/g of dry extract. The methanolic extract of leaves has highest flavonoid content (336 ±0.006 µg QE/g) but the acetone extract of leaves has the lowest content (61±0.007 µg QE/g). The methanolic extract of stem has the highest content (202 ± 0.008 µg QE/g) while aqueous extract (38.12 ± 0.006 µg QE/g) had lowest content.

TABLE 3: TOTAL FLAVONOID CONTENT OF VARIOUS PLANT EXTRACT OF *OLDENLANDIA PUMILA*

Sr. no.	Total Flavonoid Content					
	Methanol		Acetone		Aqueous	
	Leaves	Stem	Leaves	Stem	Leaves	Stem
1	336 ± 0.006	202 ± 0.008	61 ± 0.007	78.307 ± 0.006	63.60 ± 0.004	38.12 ± 0.006

Note: - Mean ± Standard deviation.

Total Tannin Content: The total tannin content in plant extract was measured using the Folin-Ciocalteu method. Tannic acid was used as standard and total tannin content was represented in tannic acid equivalents (TAE). Absorbance was measured using a spectrophotometer at 700 nm [20].

The tannic content of plant extract ranged from 64.8 µg to 163 µg of TAE/g of dry extract. The tannic content of the stem is high (163 µg TAE/g) in methanolic extract, but the stem has the lowest content (64.8 µg TAE/g) in the aqueous extract.

The total tannin content of the leaves was found to be highest in the acetone extract (163 ± 0.004 µg TAE/g) and lowest in the aqueous extract (94.8 ± 0.002 µg TAE/g). The stem's total tannin content was highest in the acetone (134.538 ± 0.004 µg TAE/g) and lowest in the aqueous extract (64.8 ± 0.003 µg GAE/g).

TABLE 4: TOTAL TANNIN CONTENT OF VARIOUS PLANT EXTRACT OF *OLDENLANDIA PUMILA*

Sr. no.	Total Tannin Content					
	Methanol		Acetone		Aqueous	
	Leaves	Stem	Leaves	Stem	Leaves	Stem
1	134.2 ± 0.008	85.2 ± 0.005	163 ± 0.004	134.538 ± 0.004	94.8 ± 0.002	64.8 ± 0.003

Total Alkaloid Content: The Dragondroff method was used to quantitively estimate the presence of alkaloids. In this technique, the interaction between bromocresol green (BCG) and alkaloid occurred.

The plant extract shows an alkaloid content ranging from 18.285 µg to 89 µg of AE/g of dry extract. The acetone extract contains lowest tannic content (18.285 ± 0.007 µg AE/g) in the stem, but a highest content (89 ± 0.006 µg AE/g) in the leaves. The result showed that the acetone extract of leaves had the highest content (89 ± 0.006 µg AE/g) whereas the aqueous extract had the lowest content (24.757 ± 0.005 µg AE/g).

The stem has highest alkaloid content in (36.846 ± 0.006 µg AE/g) and the lowest in acetone extract (18.285 ± 0.007 µg AE/g).

TABLE 5: TOTAL ALKALOID CONTENT OF VARIOUS PLANT EXTRACT OF *OLDENLANDIA PUMILA*

Sr. no.	Total Alkaloid Content					
	Methanol		Acetone		Aqueous	
	Leaves	Stem	Leaves	Stem	Leaves	Stem
1	68.67 ± 0.005	36.846 ± 0.006	89 ± 0.006	18.285 ± 0.007	24.757 ± 0.005	27.614 ± 0.006

DPPH Assay: The DPPH free radical scavenging assay is a common method for measuring antioxidant activity in plant extracts due to its simplicity, reliability, and ease to perform. The presence of antioxidants causes the DPPH radical to stabilize the molecule by gaining one extra electron or hydrogen atom and this lower UV absorbance, which shows that both manufactured substances have scavenging ability. In the present investigation results were represented as IC_{50} value *i.e.*, 50% inhibition concentration. The IC_{50} value of plant extract ranging from 438.951 to 755.956 µg/ml. In extract, maximum IC_{50} value shown in aqueous extract of leaves (755.956 µg/ml) and lowest in acetone extract of leaves (438.951 µg/ml). Lower the IC_{50} value of compound indicates stronger antioxidant potential. In the present study, we have found the IC_{50} value of sample very close to the standard, indicating stronger radical scavenging ability of the plant.

TABLE 6: DPPH SCAVENGING ACTIVITY OF VARIOUS PLANT PARTS OF *OLDENLANDIA PUMILA*

Sr. no.	Conc. (µg/ml)	DPPH Scavenging Activity (%)					
		Methanol		Acetone		Aqueous	
		Leaves	Stem	Leaves	Stem	Leaves	Stem
1	200	37.53 ± 1.86	26.32 ± 1.33	32.99± 1.17	38.85 ± 1.73	17.61 ± 2.26	33.31 ± 0.76
2	400	44.56 ± 1.27	37.31 ± 1.40	43.38± 1.22	47.39 ± 2.51	28.98 ± 1.37	43.30 ± 0.20
3	600	51.13 ± 1.86	50.18± 2.04	67.51± 1.08	54.97 ± 1.61	38.40 ± 1.15	59.21 ± 3.25
4	800	60.56 ± 2.24	59.32± 1.76	76.76± 1.24	61.21 ± 2.15	52.01 ± 3.13	76.18 ± 2.54
5	1000	67.16 ± 1.80	69.30 ± 0.96	83.18 ± 0.38	68.61 ± 1.66	66.24 ± 1.36	81.41 ± 2.73
IC_{50}Value		542.287	627.907	438.951	484.904	438.953	755.956

Frap Assay: The ferric reducing antioxidant power (FRAP) test assessed the plant extracts' antioxidant capacity. The FRAP test is used to assess the antioxidant capability of an extract based on its reducing ability. It is the only test that can quantify a sample's concentration of reductants or antioxidants. It is based on the principle of reducing the capacity of sample antioxidant to convert ferric tripyridyl triazine (Fe^{+3}-TPTZ) to a blue-colored complex ferrous tripyridyl triazine (Fe^{+2} – TPTZ) at acidic pH [21]. In methanol extract, the maximum activity is seen in leaves (234.42 ±0.006 µg Fe (II)/g) while minimum in stem (99.41 ± 0.003 µg Fe (II)/g). In acetone extract, the higher activity is seen in stem (246.8 ± 0.006 µg Fe (II)/g) while lowest in leaves (124.8 ± 0.007 µg Fe (II)/g). In aqueous extract maximum activity is seen in leaves (37.52 ± 0.004 µg Fe (II)/g) while minimum in leaves (18.48 ± 0.005µg Fe (II)/g).

TABLE 7: FRAP VALUE OF DIFFERENT EXTRACT OF *OLDENLANDIA PUMILA*

Sr. no.	Conc. (µg/ml)	FRAP ASSAY					
		Methanol		Acetone		Aqueous	
		Leaves	Stem	Leaves	Stem	Leaves	Stem
1	500	234.4 ± 0.006	99.4 ± 0.003	124.8 ± 0.007	246.8 ± 0.006	37.5 ± 0.004	18.4 ± 0.005

Note: - Mean ± Standard deviation.

Statistical Analysis: Many researchers have found that phenolic molecules are responsible for antioxidant activity. Phenolic compounds can scavenge free radicals due to the presence of certain functional groups in their molecular structures [22].

Phenolic substances have many hydroxyl groups and conjugate double bonds in their structures, allowing them to scavenge free radicals and avoid damage. The plant extracts in different solvents were associated with the parameters examined, which were TPC, TFC, TAC, TTC, FRAP and Phosphomolybdenum assay. The results showed a high positive connection between all the phenolic and flavonoid levels of plant extract using the FRAP test. In methanolic extract of leaves, positive correlation was observed between TTC & FRAP and TPC & TAC whereas TPC & TAC and TPC & Phosphomolybdenum showed positive connection in the methanolic extract of stem.

In acetone extract of leaves, a significant relationship was discovered between TAC & Phosphomolybdenum and TTC & Phosphomolybdenum however in acetone extract of stem exhibited positive connection between TFC & FRAP. TFC & FRAP and FRAP &

Phosphomolybdenum exhibited a positive association in the aqueous extract of leaves whereas TFC & TAC demonstrated a favorable relationship in the aqueous extract of stem.

TABLE 8: PEARSON CO-RELATION OF LEAVES METHANOL EXTRACT OF *OLDENLANDIA PUMILA*

	TPC	TFC	TAC	TTC	FRAP	Phosphomolybdenum
TPC	1.000					
TFC	0.071	1.000				
TAC	0.863	0.371	1.000			
TTC	-0.646	0.467	-0.346	1.000		
FRAP	0.707	-0.404	0.409	0.993	1.000	
Phosphomolybdenum	0.863	0.371	1.000	-0.346	0.409	1.000

TABLE 9: PEARSON CO-RELATION OF STEM METHANOL EXTRACT OF *OLDENLANDIA PUMILA*

	TPC	TFC	TAC	TTC	FRAP	Phosphomolybdenum
TPC	1.000					
TFC	0.308	1.000				
TAC	0.996	0.259	1.000			
TTC	-0.996	-0.259	-1.000	1.000		
FRAP	0.607	0.864	0.559	-0.559	1.000	
Phosphomolybdenum	0.996	0.259	1.000	1.000	0.559	1.000

TABLE 10: PEARSON CO-RELATION OF LEAVES ACETONE EXTRACT OF *OLDENLANDIA PUMILA*

	TPC	TFC	TAC	TTC	FRAP	Phosphomolybdenum
TPC	1.000					
TFC	0.416	1.000				
TAC	0.931	0.622	1.000			
TTC	0.966	0.559	0.993	1.000		
FRAP	0.661	-0.106	0.456	0.519	1.000	
Phosphomolybdenum	0.903	0.663	0.997	0.981	0.414	1.000

TABLE 11: PEARSON CO-RELATION OF STEM ACETONE EXTRACT OF *OLDENLANDIA PUMILA*

	TPC	TFC	TAC	TTC	FRAP	Phosphomolybdenum
TPC	1.000					
TFC	-0.326	1.000				
TAC	-0.183	0.674	1.000			
TTC	0.487	0.371	0.860	1.000		
FRAP	0.509	0.945	-0.492	0.187	1.000	
Phosphomolybdenum	-0.182	-0.674	1.000	0.859	0.493	1.000

TABLE 12: PEARSON CO-RELATION OF LEAVES AQUEOUS EXTRACT OF *OLDENLANDIA PUMILA*

	TPC	TFC	TAC	TTC	FRAP	Phosphomolybdenum
TPC	1.000					
TFC	-0.326	1.000				
TAC	1.000	-0.326	1.000			
TTC	-0.108	0.745	-0.108	1.000		
FRAP	0.509	0.945	0.509	0.567	1.000	
Phosphomolybdenum	0.539	-0.927	0.539	0.538	0.999	1.000

TABLE 13: PEARSON CO-RELATION OF STEM AQUEOUS EXTRACT OF *OLDENLANDIA PUMILA*

	TPC	TFC	TAC	TTC	FRAP	Phosphomolybdenum
TPC	1.000					
TFC	-0.881	1.000				
TAC	0.708	0.936	1.000			
TTC	0.260	-0.538	0.732	1.000		
FRAP	0.043	0.233	-0.431	0.860	1.000	
Phosphomolybdenum	0.376	0.099	0.097	0.547	-0.835	1.000

CONCLUSION: The phytochemical screening showed that *Oldenlandia pumila* extract contains a wide range of phytochemicals such as alkaloids, phenols, flavonoids, tannins, steroids, and carbohydrates. The two significant phytoconstituents for the plant's capacity to

scavenge among all the bioactive substances are flavonoids and phenols. The plant extract had greater phenolic content than flavonoid content. The high content of phenolic compounds shows significant linear association with antioxidant activity indicate its strong redox potential. The current study reveals that it has potential for pharmaceutics and has several medical applications. The current study indicates that *Oldenlandia pumila* has amazing medicinal qualities and promises for pharmaceutics; moreover, it has yet to receive the attention it deserves and will need to be investigated more to look into its many pharmacological aspects. The available data showed that the *Oldenlandia pumila* needs to be attended attentively for its hidden potential.

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