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Pharmacognostic Evaluation, Estimation of Phenolic, Flavonoid Composition and Antioxidant Activity of *Aspidopterys indica* (Willd.)W.Theob: An Endemic Plant to Peninsular India

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Abstract

Aspidopterys indica (Willd.) W.Theob is endemic plant to Peninsular India belonging to family Malpighiaceae commonly known as Chuttakulaa-tigga. The study aims to explore Pharmacognostic, Phytochemical evaluation, Total Phenolic, Flavonoid Content and Antioxidant Activities. The Pharmacognostic study comprises of Ultrasonic extraction, microscopy of Leaf and stem, fluorescence investigation, ash values, extractive values, Loss on drying, foaming index, crude fiber content, total phenolic and flavonoid content, 1,1-diphenyl-2-picrylhydrazyl (DPPH) scavenging activity and Nitric oxide scavenging activities were assessed. The leaf shows presence of paracytic stomata, T shaped trichomes, Stem shows presence of vessels and pith. The physicochemical characters assessed as Total Ash value 10%, Water insoluble ash 0.5%, acid insoluble ash 8.5%, sulphated ash 4.2%, alcoholic extractive value 16.8%, water soluble extractive value 17.6%, chloroform extractive value 0.8%, Loss on drying 3.36%, Foaming index is <100, crude fiber content is 2.4%, phytoconstituents like tannins, flavonoids, phytosterols, alkaloids and carbohydrates are available in methanol, aqueous, chloroform extracts. Total phenolic content turned greatest in methanolic extract (90.69±2.86 mg GAE/g), Aqueous extract showed maximum Total flavonoid content (109.89±0.33mg of QE/g). Methanolic extract indicated most intense 1,1-diphenyl-2-picrylhydrazyl (DPPH) scavenging action IC_{50}(134.97±8.37) whereas aqueous extract is potent to exclude nitrile radical IC_{50}108.00±4.18). Total phenolic content - DPPH scavenging assay are positively correlated. The microscopical, physicochemical parameters are valuable to authenticate the identification and purity of the drug. *A.indica* is a perspective source of antioxidants.

Key words:

DPPH scavenging assay, Microscopical characters, Nitric oxide radical scavenging activity, Ultrasonic extraction

Introduction:

Ecological changes are causing the arrival of a wide range of free radicals; even then, the plants are managing to endure. Antioxidant agent are the particles adequately stable to donate an electron to calm down free radicals and accordingly lessening its ability to harm. (Lobo, Patil, Phatak, & Chandra, 2010) Natural characteristic phytoconstituents have the ability to scavenge

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them (Saeed, Khan, & Shabbir, 2012). These free radicals are responsible for causing and develop several disorders, including cancer, neurodegeneration, heart diseases, diabetes. The prevalence of antioxidants in medicinal plants expedites opposition for some degenerative diseases (Gulcin, 2011) In light of developing necessity in this area, there is a requirement for the researcher to investigate new novel entities of antioxidant defenses to balance free radicals.

Aspidopterys indica(Willd.)W.Theob **(**Malpighiaceae) is an endemic plant to Peninsular India, Telangana (Pullaiah, 2015) disseminated in Eastern Himalayas, Orissa, Deccan, and the Western Ghats, Meghalaya (Khare C.P, 2007, p. 70). Synonyms: *Aspidopterys roxbhurgiana, Triopterys indica.* Vernacular names are Telugu- Chuttakulaa-tigga. It is a slender climbing shrub with round and cylindrical branches. Leaves are ovate 7-10 x 4-6 cm; the apex is acuminate, the entire margin, the base is cordate, Fruits are 16 mm in length, as shown in Figure 1. Petioles are 8-15mm long rusty-tomentose, Flowers are Yellow or white, acuminate tip, texture glabrous, Pedicles are 16mm in length. Petals are clawed, 5 partite lobes, 3 lobed ovaries, Styles are 3, and stamens are 10 in number (Thamanna, Narayana Rao, & Madhava chetty, 1994, p. 22 ; T. Pullaiah, 2015; Reddy & Sudhakar Reddy, 2016, p. 112) Aerial parts show Hypotensive action (Khare, 2017). The 50% Ethanolic extract of Aerial parts, fractionated portion showed hypotensive activity on experimental animals. The LD_{50} of extract calculated was 750 mg/kg i.p in experimental mice (Dhawan, Dubey, Mehrotra, & Rastogi, 1980).The phytochemical composition and detailed information of this plant are still Hazy. Here is an endeavor made to explore phytochemical, Microscopical, antioxidant activities of plant extracts. The current study is intended to probe the phytochemicals and biological activities corresponding to *A.indica,* which enables the further up to date innovation of newer restorative medicinal plants.

Fig.1 Photomicrograph of Aerial parts of *Aspidopterys indica*

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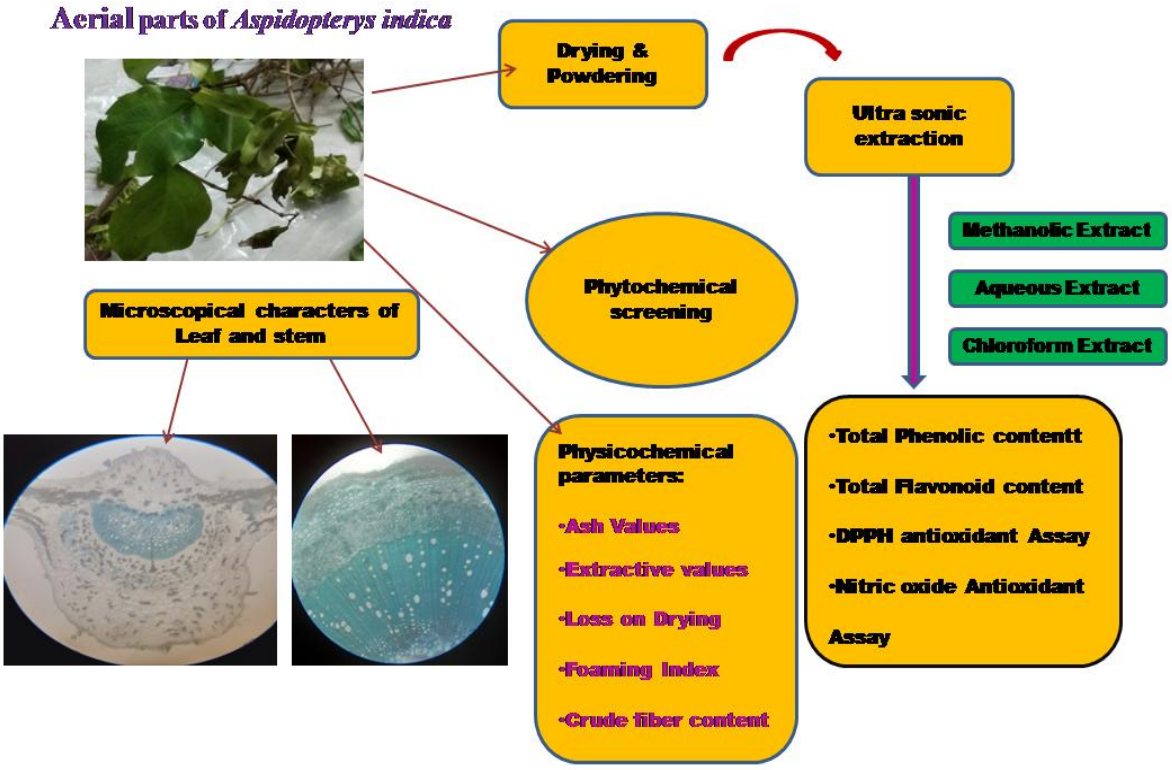

Fig.1a Design of study

Methodology

Plant collection and Authentication

Aspidopterys indica, Family Malphigiaceae was gathered from Kinnerasani Wild Life sanctuary, Bhadradri Kothagudem District Telangana, and confirmed by Botanist P. V. Prasanna, Botanical Research of India, Hyderabad and Voucher example no. BSI/DRC/2019-2020/Tech./838 was kept in BSI herbarium for additional reference.

Preparation of plant samples for Microscopic examination

The Dried plant samples were boiled slightly in water to render the tissues soft. The hydrated, fresh samples were taken; with the help of a neat sharp blade, the samples were cut into thin sections and then immediately transferred to watch glass containing water. Add several drops of clearing solution (85-92%) of Lactic acid solution to the sections and stained with 0.1%(w/v) Toluidine blue O (Tak Yeung, Stastolla, Sumner, & Huang, 2015, pp. 14–70). Plant tissue samples were added with a fixative solution, i.e., FAA (80ml of 70% v/v Ethyl alcohol, 10 ml 40% Formaldehyde, 5ml Glacial Acetic acid) and the samples were slightly dehydrated with ethanol and are ready for examination (Cutler, Botha, & Stevenson, 2007, pp. 57–172)

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Extraction

The aerial parts of *A.indica* were collected, washed, dried under shade, powdered coarsely, and extracted with solvents like methanol, water, and chloroform independently on ultrasonicator at 40 kHz for 45 mins at a temperature of 40ºC. The supernatants were separated, filtered, and dried under vacuum, and the extract concentrates were kept in a desiccator.

Physicochemical parameters

Physicochemical parameters such as Ash values and extractive values were performed according to the official method prescribed in WHO guidelines on quality control methods for medicinal plants (Quality control methods for medicinal plants.,1998). Fluorescence analysis of powdered plant was carried out by standard methods (Kabra, Sharma, Singla, Kabra, & Baghel, 2019). Each concentrate was treated with different reagents and tried for the presence of essential and optional metabolites like Carbohydrates, Fats, Proteins, Gums, alkaloids, Glycosides, Tannins, Flavonoids, Phenols, by common standard procedures (Kokate, 1994, pp. 107–113).

Quantification of Total Phenolic Content by Folin-Ciocalteu calorimetric method

The standard stock solution was prepared by adding 0.001g Gallic acid to 10 ml Methanol. 50mg of dried Methanolic, Aqueous, Chloroform extracts were taken and diluted with 50 ml of ensuing solvents resulting in 1mg/ml stock solutions. Subsequent dilutions of 25µg/ml, 50µg/ml, 75 µg/ml, 100 µg/ml, 125 µg/ml, 150 µg/ml were made. To each 1ml of dilution (standard/extract) 5ml 5% FCR (Folin-ciocalteu reagent), final volume of 10 ml modified with 7% Na_2Co_3 (P^H 10). The resulting blue colored mixture was brooded in an Incubator for 30 min at 40ºC. Absorbance was estimated at 760 nm against the blank. The absorbance of different concentrations was taken threefold. The standard calibration plot of Gallic acid was used to determine the content of phenols in the extracts and expressed as mg Gallic acid equivalents(GAE) per gram of extract. (Phuyal, Jha, Raturi, & Rajbhandary, 2020).The total phenolic content in the samples was resolved utilizing equation:

$C=c\ V/M$

C= Total Phenolic content mg GAE/g dry extract

C= Concentration of Gallic acid obtained from standard Calibration curve in mg/ml

V= Volume of the extract in ml

M= Mass of the extract in g

Quantification Total Flavonoid Content by Aluminum calorimetric assay

The Total Flavonoid content was determined by Aluminum Calorimetric assay. 4000µg of Quercetin was dissolved in 1ml of 95% ethanol as a stock. 1mg/ml extract stock solutions were prepared. 1ml of each Subsequent concentrate dilution was added to the volumetric flask containing 3ml of 95% ethanol, 0.2 ml of 10% $AlCl_3$, 0.2ml of 0.1mM Potassium acetate solution, and the final volume of 10 ml was made with distilled water. Absorbances were taken

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threefold for all the dilutions at 415nm (Kaneria, Kanani, & Chanda, 2012). Based on the Linear Calibration curve, flavonoid content was estimated as Quercetin equivalent mg QE/g.

Antioxidant activity

DPPH Scavenging Assay

DPPH scavenging assay as described in (Shimada, Fujikawa, Yahara, & Nakamura, 1992)About 1ml DPPH (0.004%w/v) solution was added to 3ml of extract solutions of different concentration (25µg/ml, 50µg/ml, 75 µg/ml, 100 µg/ml, 125 µg/ml, 150 µg/ml) blended properly and permitted to stay at room temperature for around 30 minutes. Methanol was taken as blank, and about 1ml of DPPH in 3ml Methanol was taken as Negative control. The absorbances of resulting solutions were measured at 517 nm three-fold. IC_{50} is 50% inhibition or concentration of sample required to inhibit 50% free radicals of DPPH (Johari & Khong, 2019. The Free radical scavenging activity of extracts is concentration-dependent. Ascorbic acid is used as Positive control.

$$\% \text{ Scavenging activity} = \frac{A_0 - A_s}{A_s} \times 100$$

A_0 shows the absorbance of Negative control reaction mixture
A_s shows the absorbance of Extract and DPPH reaction mixture

A graph was plotted against % scavenging and various concentrations of extracts and Ascorbic acid. By utilizing the trend line and Regression equation, for each curve in the graph, IC_{50} was determined. All the outcomes were determined three-fold and communicated in Mean ± SD.

Nitric oxide scavenging assay

The assay was performed on different concentrations of the extract prepared by serial dilution of the stock solutions (10mg/ml). Gallic acid was used as a positive control. The reaction mixture (2ml of 10mM Sodium Nitroprusside solution + 1ml of Standard/ Extract (25-150µg/ml) was incubated at 25ºC for 3hrs, to which 2ml of Griess reagent was added and kept at room temperature for 30 mins. The same reaction mixture without a sample is Negative control. The absorbances were taken threefold at 546nm against the blank (Alam, Bristi, & Rafiquzzaman, 2013).

$$\% \text{ NO Scavenging} = \frac{A_{\text{Negative control}} - A_{\text{sample}}}{A_{\text{sample}}} \times 100$$

$A_{\text{Negative control}}$ = Absorbance of Negative control

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A_{Sample} = Absorbance of Gallic acid or Sample extract.

Data analysis

The outcomes were examined in sets of three and experimental data is analyzed statistically by one-way ANOVA followed by Post hoc test Tukey($P<0.05$) conveyed as mean ± SD with aid of Graph Pad Prism 9 software. Pearson's correlation coefficient is used to correlate total phenolic content and DPPH antioxidant assay.

Results

Phytochemical Evaluation

Phytochemical screening is indicated in Table 1, the presence of Tannins, Phytosterols, Flavonoids, Alkaloids,Carbohydrates, Proteins.

Table 1: Phytochemical investigation for Methanolic, Aqueous and chloroform extracts *A. indica*

Phyto constituents	AIME	AIAE	AICE
Alkaloids	+Ve	+Ve	-Ve
Glycosides	-Ve	-Ve	-Ve
Tannins	+Ve	+Ve	+Ve
Saponins	-Ve	-Ve	-Ve
Flavonoids	+Ve	+Ve	+Ve
Phytosterols	+Ve	+Ve	+Ve
Carbohydrates	+Ve	+Ve	-Ve
Volatile Oils	-Ve	-Ve	-Ve
Proteins	-Ve	-Ve	+Ve

Physicochemical parameters

Fluorescence analysis indicated in Table 2 and Ash values, Extractive values, crude fiber content, loss on drying, foaming index of various extracts were given in Table.3

Table 2: Fluorescence analysis of *A. indica*

S.NO	Reagent	Visible light	UV Fluorescence	
			254 nm	365 nm
1	Powder as such	Dark Green	Green	Pale Green
2	Powder + 1N Aqueous NaOH	Pale green	Green	Blue
3	Powder + 1N Alcoholic NaOH	Green	Brown	Blue
4	Powder + 1N HCL	Pale Green	Yellow	Purple
5	Powder + conc H2SO4	Brown	Pale Green	Purple

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6	Powder + conc HNO3	Green	Dark Green	Pale Green
7	Powder + Acetic acid	Green	Yellow	Fluorescent green
8	Powder + Picric acid	Green	Fluorescent Purple	Green
9	Powder + Ferric chloride	Brown	Pale Green	Purple
10	Powder + Chloroform	Green	Pale green	Brown
11	Powder + Acetonitrile	Brown	Dark green	Purple
12	Powder + Petroleum ether	Pale green	Yellow	Brown
13	Powder+ Water	Green	Green	Brown
14	Powder+ Diethyl ether	Green	Pale green	Purple

Table 3: Physicochemical Parameters of Aerial parts of *A. indica*

S.NO	Parameters Determined	Values in (%) W/W
1	Total Ash	10 %
2	Water insoluble ash	0.50 %
3	Acid Insoluble Ash	8.50 %
4	Sulphated ash	4.20 %
5	Alcoholic Extractive value	16.8
6	Water soluble Extractive value	17.6
7	Chloroform Extractive value	0.8
8	Loss on Drying	3.36
9	Foaming Index	< 100
10	Crude Fibre content	2.4

Microscopical evaluation

Microscopy of Leaf: Transverse section of leaf showed dorsiventral pattern and Figure 2 illustrates a transverse segment of a leaf stained with toulidine blue and safranin in Figure 3. The lamina is comprised of the upper epidermis, mesophyll and lower epidermis. Paracytic stomata and T Shaped covering trichomes are present on both surfaces of the leaf. The upper epidermis was single-layered with more or less rectangular cells. The mesophyll was differentiated into palisade and spongy parenchyma. On the adaxial surface below the upper epidermis, single-layered elongated, compact palisade-like cells are present, which are absent above the lower epidermis. The spongy parenchyma layer consists of loosely arranged cells with intercellular spaces. The epidermal layers of lamina were proceeded in the midrib region. The strips of collenchyma are present just below the upper epidermis and above the lower epidermis. A well-developed arc-shaped vascular bundle was seen in the center of the midrib region. The vascular bundle was surrounded by lignified sclerenchymatous endodermis layer. Phloem pole (non-lignified) is facing towards the abaxial surface and Xylem pole (lignified) towards the adaxial

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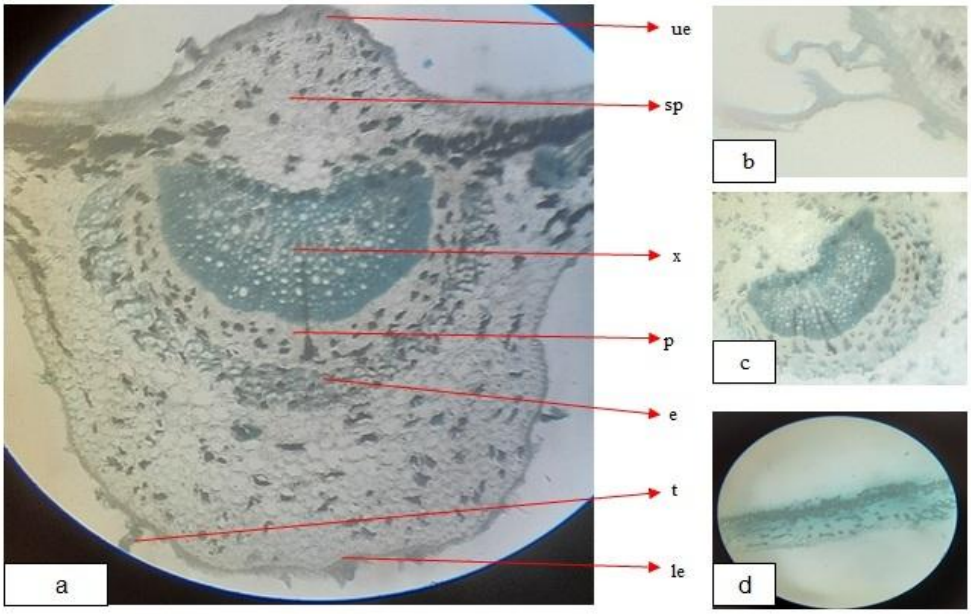
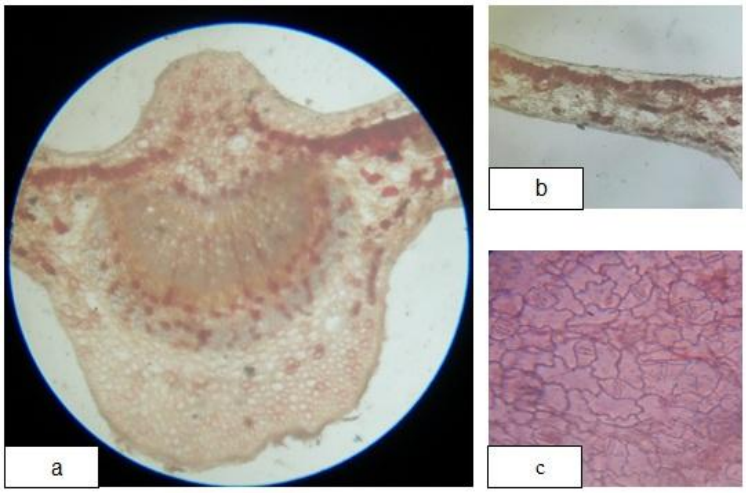
surface.

Fig. 2 Transverse section of Leaf



a. ue) Upper Epidermis **sp**) Spongy Parenchyma **x)** Xylem **p)** Phloem **e)** Endodermis **t)** Trichomes **le)** Lower epidermis **b**. T shaped Trichomes **c**. Midrib **d**. Palisade

Fig. 3



a.Transverse section of Leaf stained with Safranin Leaf lamina c. Paracytic Stomata

Microscopy of stem:

Microscopy of Stems (Figure 4) shows single-layered epidermis and multilayered hypodermis Well developed stomata and trichomes are present. Cortex is narrow, phloem and xylem are

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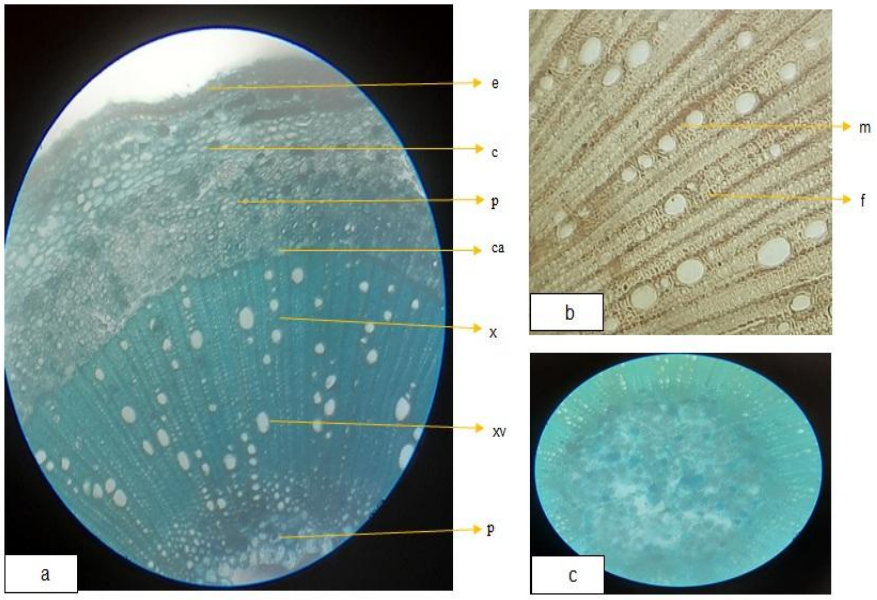
separated by cambium composed of elongated fusiform cells, phloem is non lignified, and xylem is lignified. The secondary xylem occupies the major part of the stem in comparison with the phloem. Vessels may be present as solitary or in groups arranged in radial chains, surrounded by thin-walled fibers and scattered parenchyma. The outermost phloem layers are almost embedded into the bark. Medullary rays are single-layered runs radially from phloem to xylem and then merge into the pith. The central region of the stem is composed of ground tissue or pith composed of thin-walled Parenchymatous cells.

Fig. 4 Transverse Section of Leaf stem



a. e) Epidermis **c)** Cortex **p)** Phloem **ca)** Cambium **x)** Xylem) **xv)** Xylem vessels **p)** Pith **b. m)** Medullary rays **f)** Fibres **c.** Pith

Total Phenolic content and Flavonoid content

The calibration curve is constructed using the absorbances of Gallic acid and Quercetin at different concentrations, and the total phenolic content and flavonoid content of *A.indica* are assessed using the regression equation as shown in Figures 5 and 6. From the results tabulated in Table 4,5 the total phenolic content turned maximum in methanolic extract range starting from 5.65 mg GAE/g to 90.69 mg GAE/g, minimal content was turned in chloroform extract starting from 3.47 mg GAE/g to 66.90 mg GAE/g. In aqueous extract it was recorded from 18.43 mg GAE/g to 83.66 mg GAE/g. In equal tendency, the higher total flavonoid content has been recorded in Aqueous extracts (109.89±0.33mg QE/g). The flavonoid content in methanolic extract was found to be equivalent to 95.65±0.48mg QE/g, 80.54±0.372mg QE/g in chloroform extract.

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Table 4: Total phenolic content of *A. indica*

Concentration µg/ml	Total phenolic content of Gallic acid mg/g	Total phenolic content of AIME mg GAE/g	Total phenolic content of AIAE mg GAE/g	Total phenolic content of AICE mg GAE/g
25	28.75±0.36	5.65±0.212	18.43±1.26	3.47±1.388
50	49.03±0.96	27.45±0.68	31.44±1.73	22.31±0.64
75	80.74±0.56	64.86±2.52	42.82±0.65	33.61±0.36
100	108.15±1.42	76.30±1.18	50.37±0.699	42.59±2.12
125	128.28±0.65	81.81±0.94	62.73±0.94	47.50±0.24
150	140.88±0.76	90.69±2.86	83.66±1.84	66.90±0.32

Values are expressed in Mean±S.D, n=3, significant at P< 0.05
AIME- *A. indica* Methanolic extract, AIAE- *A. indica* Aqueous Extract, AICE- *A.indica* chloroform Extract GAE/g – Gallic acid equivalents per gram.

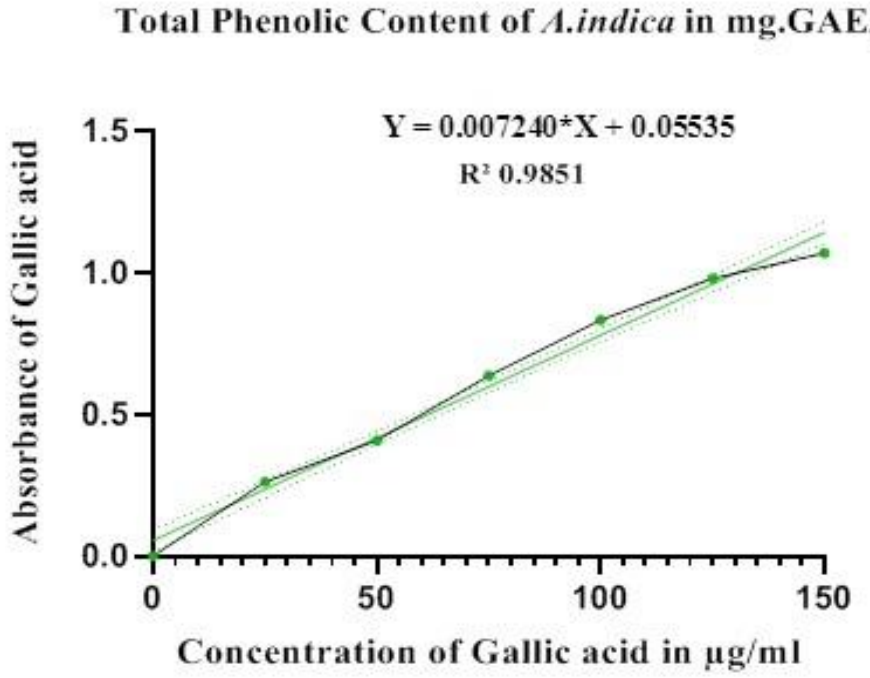

Fig.5 Plot of absorbances of Gallic acid at various concentrations

Table 5: Total Flavonoid content of *A. indica*

Concentration µg/ml	Total Flavonoid content of Quercetin mg /g	Total Flavonoid content of AIME mg QE/g	Total Flavonoid content of AIAE mg QE/g	Total Flavonoid content of AICE mg QE/g
0	0	0	0	0

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25	31.29±0.322	15.97±0.48	7.81±0.201	15.81±0.73
50	49.52±0.42	34.62±0.24	34.73±1.02	26.40±0.18
75	72.63±0.46	43.12±0.61	52.31±0.37	42.58±0.58
100	96.40±0.186	78.44±1.22	82.37±0.79	64.95±0.56
125	129.30±0.40	92.26±1.61	95.54±1.49	67.69±0.56
150	150.86±0.46	95.65±0.48	109.89±0.33	80.54±0.372

Data represented as Mean ± SD of n=3, significant at P< 0.05
QE/g Quercetin equivalents per gram

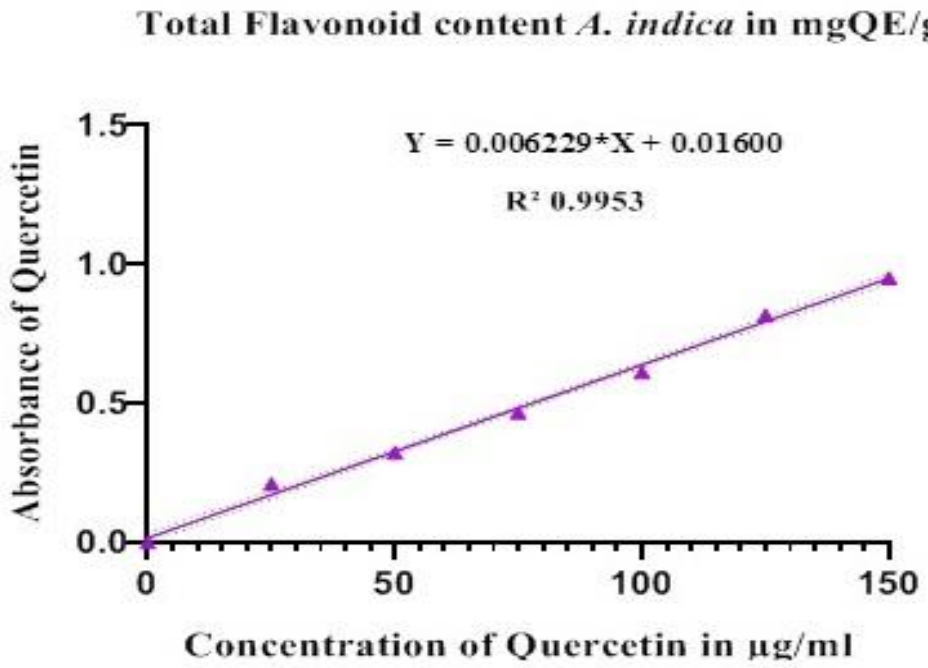

Fig.6 Plot of absorbances of quercetin at various concentrations

DPPH scavenging assay

The DPPH radical is a stable organic free radical with an absorption band at 515-528 nm, and in this manner, it is a helpful reagent for exploring the free radical scavenging activities of different compounds (Sharma and Singh, 2012). Antioxidant molecules can extinguish DPPH radicals (by giving Hydrogen atoms or electron donation) and convert them into colorless compound (Nikavar and Esbati, 2012). IC_{50} values (Table 6) were determined as 134.97±8.37, 143.48±1.31, 156.35±8.54 for extracts. Among all extracts, Methanolic extract indicated intense scavenging action. The other extracts Aqueous and chloroform, showed moderate activity. Graph of percent DPPH radical scavenging of various *A.indica* extracts illustrated in Figure 7. The results as shown in Figure 8 manifest that there is a significant difference between methanolic and chloroform extracts (P<0.05)

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Table 6: Mean % DPPH scavenging of Ascorbic acid and *A.indica* extracts at various concentrations

Concentration µg/ml	% Scavenging of Ascorbic acid	% Scavenging of AIME	% Scavenging of AIAE	% Scavenging of AICE
0	0	0	0	0
25	22.77±3.05	3.56±1.29	5.29±0.183	2.576±0.35
50	31.39±1.39	18.71±1.07	21.27±1.78	11.546±0.21
75	61.89±4.06	31.5±1.78	25.77±1.92	13.36±0.21
100	74.67±3.69	35.67±3.04	31.69±1.58	25.75±2.39
125	81.62±1.62	45.15±2.95	41.27±1.67	42.083±2.61
150	96.23±4.64	56.22±3.19	53.33±0.26	45.61±4.19
IC_{50}	65.86±3.963	134.97±8.37	143.48±1.31	156.35±8.54

Data represented as Mean ± SD of n=3, significant at P< 0.05

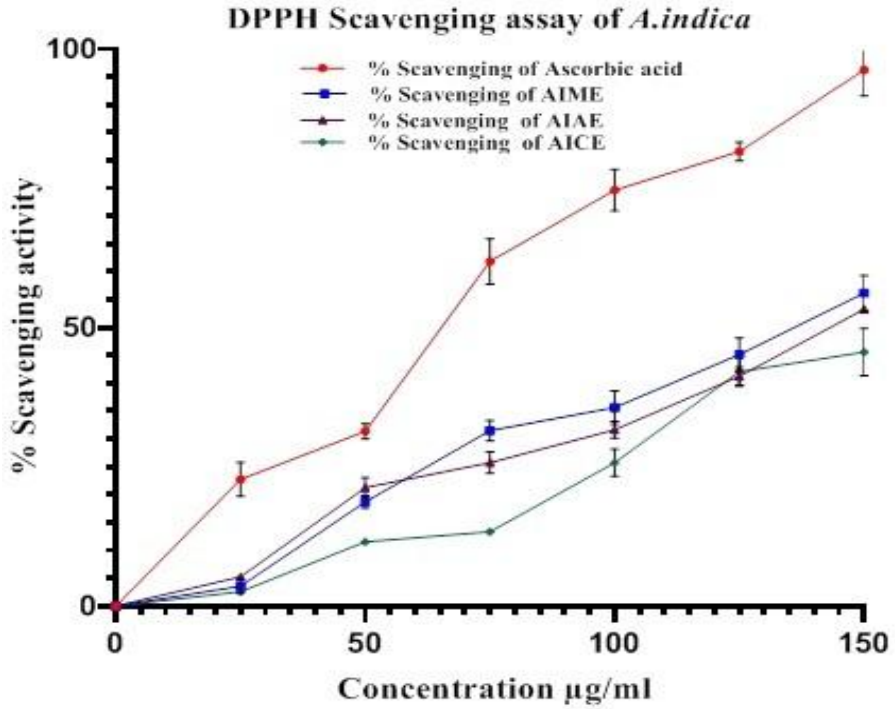

Figure.7: Plot of % DPPH radical scavenging of various extracts of *A.indica* and Ascorbic acid as positive control

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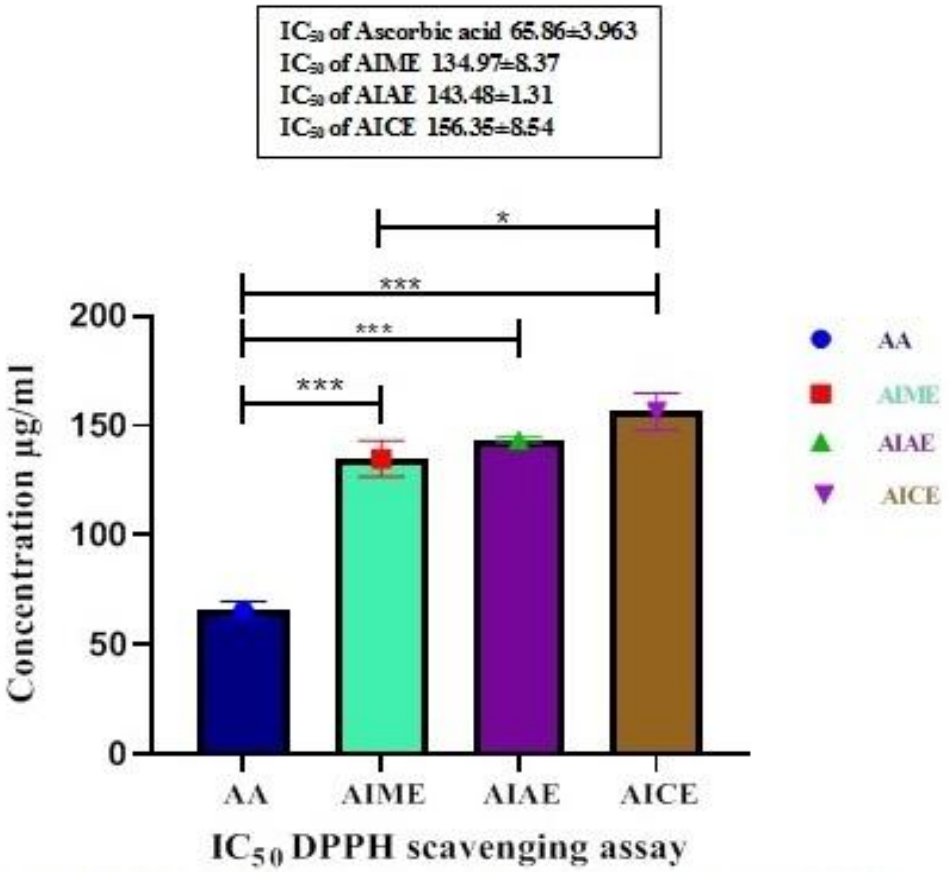

Figure.8 : IC_{50} of DPPH scavenging activity of various extracts of *A.indica* and ascorbic acid, results analyzed by One-way ANOVA followed by Tukey. The average values were significant at P<0.05

Nitric oxide radical scavenging Activity

The Plants manifest antioxidant activity through combat with oxygen to scavenge the nitrile radical which has been produced from SNP at physiological p^H in an aqueous environment (Boora and Chirisa, 2014). The Nitric oxide scavenging activity was given in Figure 9, the aqueous extract shows more intensity to eliminate nitrile radical contrasted to other extracts. IC_{50} values (Table 7) for the extracts were determined as 127.4±3.67, 108.00±4.18, 143.23±9.77. Figure 10 displays the findings of the experiment that there is a significance between all the extracts (P<0.05).

Table 7: Mean % Nitric oxide scavenging of Gallic acid and *A. indica* extracts at various concentrations

Concentration µg/ml	% Scavenging of Gallic Acid	% Scavenging of AIME	% Scavenging of AIAE	% Scavenging of AICE

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25	21.73±1.77	10.54±2.02	1.89±0.19	6.83±1.79
50	39.28±2.81	8.386±2.09	23.03±3.11	15.38±3.06
75	60.51±3.8	21.67±3.25	32.34±1.30	26.11±3.81
100	74.85±5.29	29.66±8.11	42.41±3.00	32.55±2.07
125	86.43±4.77	49.41±3.23	63.22±2.96	44.33±3.72
150	97.76±3.55	63.95±5.31	69.29±3.04	51.07±1.61
IC_{50}	62.81±0.37	127.4±3.67	108.00±4.18	143.23±9.77

Data represented as Mean ± SD of n=3, significant at P< 0.05

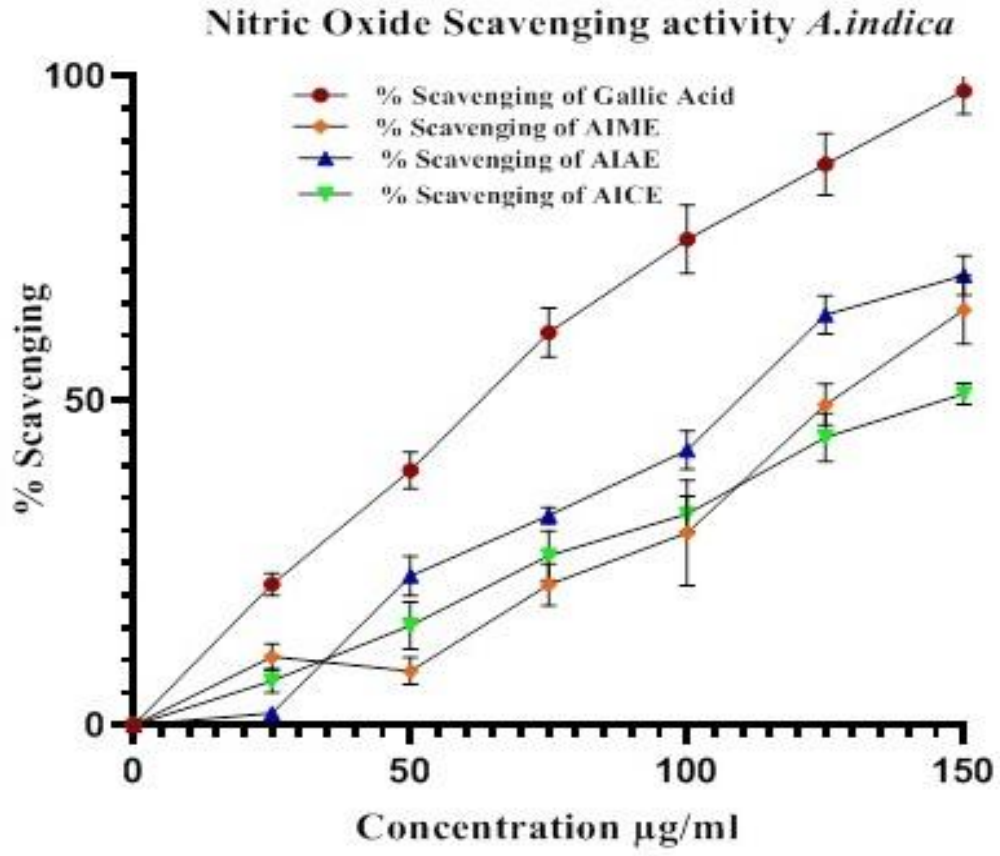

Figure.9: Plot of % Nitric Oxide radical scavenging of various extracts of *A.indica* and Gallic acid as positive control

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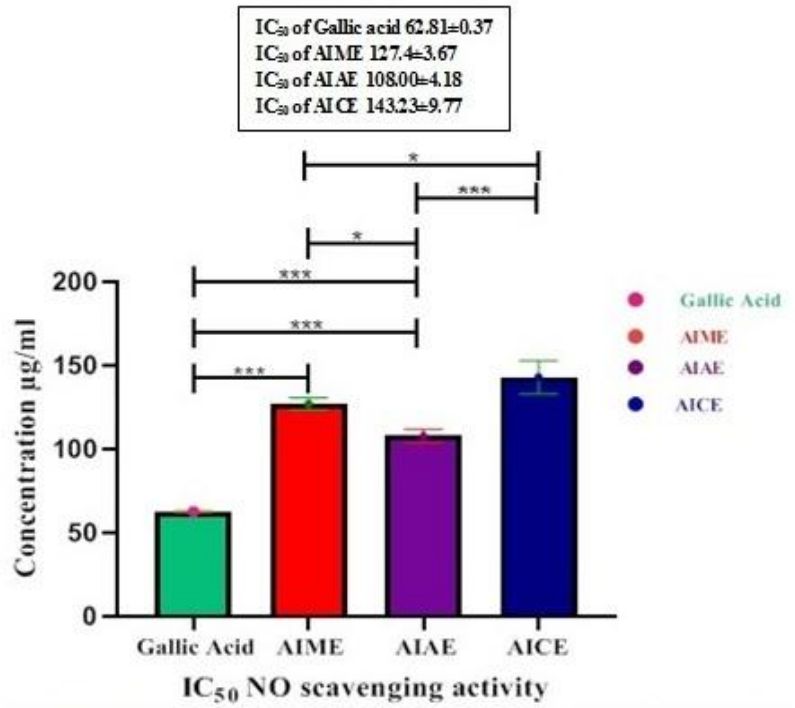

Figure.10 : IC_{50} of Nitric Oxide scavenging activity of various extracts of *A.indica* and Gallic acid, results analyzed by One-way ANOVA followed by Tukey. The average values were significant at P<0.05

Correlation

Correlation was figured using Pearson correlation coefficient, Graph pad prism 9. It was found that, Total phenolic content of (AIME, AIAE, AICE) significantly correlated with its DPPH scavenging assay R^2 0.9581, r 0.9788; R^2 0.9826, r 0.9913; R^2 0.9002, r 0.9488. It was recommended that antioxidant action in plants is possibly ascribed to phenolic compounds (Saha and Varma, 2016).

Discussion

Today complex current exploration devices for assessing plant drugs are accessible, yet microscopical studies are one of the most straightforward and least expensive techniques to begin for building up the right personality of source material. The microscopical characters are prerequisite analytical parameters. The extractive quantities are valuable to assess chemical constituents present in crude drugs and furthermore help in the assessment of constituents soluble in a particular solvent. The ash values give a thought of earthy or inorganic content (Kumar, Kumar, & Prakash, 2011). The phytochemical examination of *A. indica* extracts contains a combination of phytochemicals carbohydrates, phenolic compounds, flavonoids, phytosterols, tannins, alkaloids and proteins. In our work, we have picked for identification proof of Microscopical characters of stem and leaf of *A. indica*, phytochemical screening,

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physicochemical parameters, total phenolic content, total flavonoid content, DPPH antioxidant activity, Nitric oxide scavenging activity. The most noteworthy phenolic content was seen in methanolic extract and flavonoid content in Aqueous extract. The plants possessing a wide assortment of Antioxidant capacity may be associated with phenols, flavonoids and other phytocontent (Medini, Fellah, Ksouri, & Abdelly, 2014).

Conclusion

The present study the contributes the information of microscopical characters and physicochemical standards of *A. indica*. Since phenols and antioxidant activity have a positive relationship, the findings indicate that phytochemicals present in extract may function as scavengers. It has been suggested that polyphenols, flavonoids and other phytoconstituents in *A.indica* may contribute to antioxidant action.

Conflict of Interest: The current investigation claims that there is no conflict of interest

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Limitations and Future study

However, the safety profile and isolation of bioactive compounds are needed to be thoroughly investigated and screened for biological potential.

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