



GC-MS and HPLC Supported Phytochemical Analysis of *Tridax Procumbens* Linn Leaves

Gomathi Rajendran¹, Usharani Boopathy², Shobana Chandrasekar³, Rohini Durairaj⁴

¹Department of Biochemistry, Vels Institute of Science, Technology & Advanced Studies.

*Corresponding author's E-mail: drrohinidurairaj@gmail.com

Article History	Abstract
Received: 08 June 2023 Revised: 21 Sept 2023 Accepted: 10 Dec 2023	<p>Background: <i>Tridax procumbens</i> (<i>T. procumbens</i>) Linn. is a daisy family commonly known as “Ghamra” or “Coat Buttons”, the native of this plant is tropical America, Asia, Australia and throughout India, it is also distributed in roadside, waste grounds, riverbanks mainly during rainy season. It has medicinal properties due to presence of some bioactive chemical constituents. Objectives: Phytochemical analysis of <i>Tridax procumbens</i> (<i>T. procumbens</i>) Linn leaves were carried out in different extraction. Methods: Qualitative, Quantitative, HPLC, GC- MS & UV-FTIR. Results: The phytochemical qualitative analysis of <i>Tridax procumbens</i> (<i>T. procumbens</i>) Linn was performed with various extraction. The hydro-ethanolic extract contains a high concentration of phytochemical constituents than ethanol, hexane, petroleum ether & aqueous extracts. The phytochemical constituents found were tannin, saponin, flavonoids, steroids, terpenoids, triterpenoids, alkaloids, anthraquinone, polyphenol, glycoside, coumarins and emodin. The wide range of bioactive compounds were found to have antitoxicity, anticancer, antimicrobial, antifungal, antioxidant, antitumor, antimicrobial, antifouling, nematocidal, antiarthritic, hepatoprotective, hypocholesterolemic, 5-alpha reductase inhibitor, antihistaminic, anticoronary insectifuge, antieczemic, antiacne, anti-androgenic flavour, pesticide, lubricant, haemolytic 5-alpha reductase inhibitor, antipsychotic, potent antibacterial agent, antimalarial activities. Conclusion: Hydro-ethanolic extract showed several phytochemical constituents and bioactive compounds which can be used for therapeutic purposes.</p>
CC License CC-BY-NC-SA 4.0	Keywords: <i>Tridax procumbens</i> , Hydro-ethanolic extract, Phytochemical constituents, Bioactive compounds

1. Introduction

Tridax procumbens commonly known as “Coat Buttons” or “Tridax daisy” is a species of flowering plant in the family Asteraceae. The plant can be found in fields, meadows, croplands and roadsides in areas with tropical or semi-tropical climates. This plant being rich in numerous bioactive constituents of therapeutic values which is used as a source of remedy for treating various human diseases and it has diverse pharmacological properties. The plant is found to have antitoxicity, anticancer, hypocholesterolemic, nematocidal, antiarthritic, hepatoprotective, 5-alpha reductase inhibitor, antihistaminic, antimicrobial, antifungal, antioxidant, antitumor, antimicrobial, antifouling, anticoronary insectifuge, antieczemic, antiacne, anti-androgenic flavour, pesticide, lubricant, haemolytic 5-alpha reductase inhibitor, antipsychotic, potent antibacterial agent, antimalarial. The secondary metabolites are tannin, saponin, flavonoids, steroids, terpenoids, triterpenoids, alkaloids, anthraquinone, polyphenol, glycoside, coumarins and emodin. Furthermore, identification and purification of the active compounds responsible for the therapeutic activities may prove the plant of great pharmacological importance.

2. Materials And Methods

The plant was collected from local area of Chengalpattu district, Tamil Nadu, India. It was authenticated from botany department of University of Madras, Guindy. The leaves were separated from the plant and washed thoroughly with water. Further the cleaned leaves were shade dried, after complete drying the leaves were grinded in mechanical grinder to get fine powder that was stored in air tight containers. Then 10gms of powder was used for extraction. Cold extraction was performed

using the maceration method into different solvents such as ethanol, hydro-ethanol(ethanol&water-70:30), hexane, petroleum ether, aqueous for 24hours using the “intermittent shaking” method to obtain extracts (figure 1). The extracts were further filtered using Whatman filter no:1 paper and filtrate was used for phytochemical analysis (figure 2).

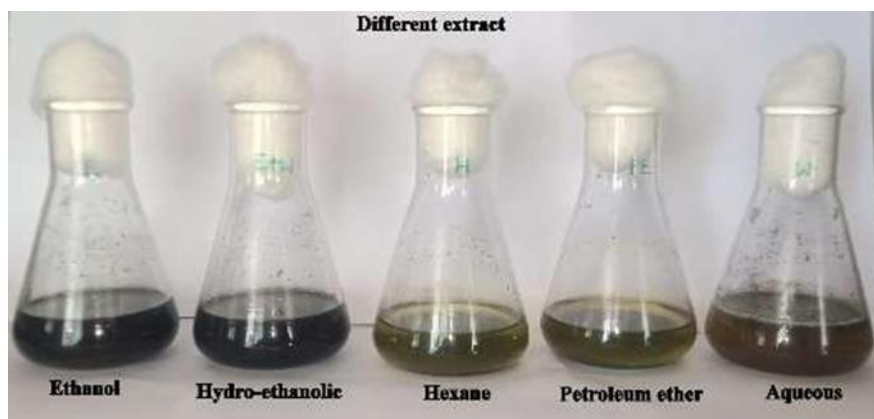


Figure: 1 Different extraction

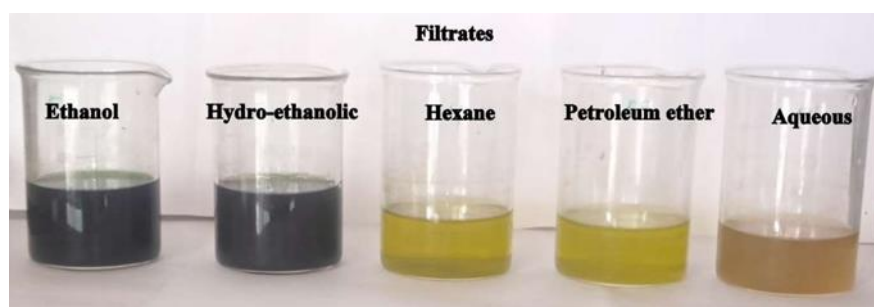


Figure: 2 Filtrates of different extraction

Determination of Extraction Yield

The extraction yield was calculated by the following equation: Extraction yield (%)

= $W1/W2 \times 100$; Where W1 is the weight of leaves extract and W2 is the dry weight of the sample taken (Table 1).

Table 1: Yield of various extracts

Extracts	Weight of (W1) leaves extract (g)	Weight of (W2) Sample taken (g)	Yield of Extract (s) (%)
Ethanol	1.216	10	12.16
Hydro-ethanolic	1.423	10	14.23
Hexane	0.675	10	6.75
Petroleum ether	0.846	10	8.46
Aqueous	1.034	10	10.34

Qualitative Preliminary Phytochemical Analysis

Preliminary phytochemical screening was carried out by using standard procedure followed by Sofowara (1993), Trease and Evans (1989) and Harborne (1973, 1984).

Test for Tannins

About 1ml of different extractions of leaves were boiled in 20 ml of water in different test tubes and then filtered. A few drops of 0.1% ferric chloride were added and observed for brownish green or a blue-black coloration.

Test for Saponin

About 2 ml of different extractions of leaves were taken in different test tubes and boiled in 20 ml of distilled water in water bath and filtered. 10ml of the filtrate was mixed with 5 ml of distilled water and shaken vigorously for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously, then observed for the formation of emulsion.

Test for Flavonoids

About 5 ml of dilute ammonia solution were added to different test tubes and a portion of the different extractions of leaves followed by addition of concentrated H₂SO₄. A yellow coloration observed in each extract indicated the presence of flavonoids. The yellow coloration disappeared after some time.

Test for Steroid

About 2 ml of acetic anhydride was added to 1ml of different extractions of leaves in different test tubes with 2 ml of H₂SO₄. The color changed from violet to blue or green in some samples indicating the presence of steroids.

Test for Terpenoids (Salkowski test)

About 5ml of different extractions of leaves were mixed in 2 ml of chloroform and 3 ml of concentrated H₂SO₄ carefully added to form a layer. A reddish-brown coloration of the inter face formed indicates positive results for presence of terpenoids.

Test For Triterpenoids

About 1ml of the different extractions of leaves was added to 1 ml of chloroform, 1 ml of acetic anhydride, followed by addition of 2 ml of concentrated H₂SO₄. Formation of reddish violet colour indicates the presence of triterpenoids.

Test For Alkaloids

Mayer's test: To 1 ml of the different extractions of leaves, a drop of Mayer's reagent was added by the side of the test tube. A creamy or white precipitate indicates the presence of alkaloids.

Test For Anthraquinones

About 5 ml of the different extractions of leaves was hydrolyzed with diluted H₂SO₄ extracted with benzene. Then 1 ml of dilute ammonia was added to it. Rose pink coloration indicates the presence of anthraquinones.

Test for Polyphenols

About 4 ml of ethanol is added to 1 ml of each extraction and the resulting solution is transferred in different test tube and warmed in a water bath for 15 mins. Three drops of freshly prepared ferric cyanide solution were added to the extract solutions. Formation of a blue green colour indicated the presence of polyphenols.

Test for Cardiac glycosides (Keller-Kiliani test)

About 5 ml of each extract was treated with 2 ml of glacial acetic acid containing one drop of ferric chloride solution. This was underlayered with 1 ml of concentrated sulfuric acid. A brown ring of the interface indicates deoxy sugar characteristics of cardenolides. A violet ring may appear below the brown ring, while in the acetic acid layer, a greenish ring may form just gradually throughout thin layer.

Test for Coumarins

About 3 ml of 10% NaOH was added to 2 ml of different extractions of leaves in different test tubes and formation of yellow colour indicates the presence of coumarins.

Test for Emodin

About 2 ml of NH₄OH and 3 ml of Benzene was added to the different extractions of leaves. Appearance of red colour indicates the presence of emodin.

Test for Anthocyanins

About 2 ml of different extractions of leaves was added to 2 ml of 2N HCl and ammonia. The appearance of pink-red turns blue-violet indicates the presence of anthocyanins.

Quantitative Analysis

Total Phenols Determination

The amount of total phenolic contents of leaves of *Tridax procumbens* were determined by the spectrophotometric method of Kim *et al.*, (2003) with slight modification. To 1g of leaves powder (dried powder) 10 ml of ethanol was added and filtered after 15 mins. 1 ml of Folin-Ciocalteu's phenol reagent was added to the mixture and shaken. After 5 min, 10 ml of 7% Na₂CO₃ solution was mixed in to the test sample solution was diluted to 25 ml distilled water and mixed thoroughly. Total phenol content was determined from extrapolation of the calibration curve which was made by preparing Gallic acid solution (10 to 100 µg/ ml). The total phenolic content was expressed as milligrams of Gallic acid (GAE) equivalents per gram dried sample.

Total Flavonoids Determination

The total flavonoids assay was conducted according to Katasani (2011). Total flavonoids content was determined by using Aluminium chloride colorimetric method. 1g of leaves powder (dried powder)

with 10 ml of water for 15 mins and filtered. Leaves extract (0.5 ml) was mixed with 1.5 ml of methanol, 0.1 ml of 10% aluminum chloride, 0.1 ml of 1M potassium acetate and 2.8 ml of distilled water. It remained at room temperature for 30 minutes. The absorbance of the reaction mixture was measured at 510 nm using UV-Visible spectrophotometer. The calibration curve was prepared by preparing quercetin solutions at concentrations 10 to 100 µg/ml in methanol. The total flavonoids content was expressed as milligrams of quercetin equivalents per gram of dried sample.

Determination of Steroids

The steroids were determined by the method of Attarde Daksha *et al.*, (2010). The sample was weighed to 1 gm, dissolved in chloroform to 10 ml and further diluted to 10 times (10,000 ppm). 3ml of diluted sample solution were mixed with 2ml of Liberman-Burchard reagent and 2 ml of chloroform. The tubes were covered with black carbon paper and kept in ice bucket in dark place for 15 min. Liberman-Burchard reagent react with the sterol to produced characteristic green colour, their absorbance was determined on spectrophotometer at 640 nm. The absorbances of standard cholesterol (20 to 80 µg/ml) were determined on spectrophotometer and standard graph was plotted. Steroid Content was expressed as milligrams of cholesterol equivalents per gram of dried sample.

Estimation of total saponins

Total saponins contents in dried powder sample were estimated by colorimetric methods (Hiai *et al.*, 1976). Take 2 gram of leaves sample into 100 ml conical flask and add 25ml methanol (99.9%). Flask was tightly sealed and kept in a shaker at 25°C and 120 rpm for 24hrs, followed by centrifuging at 3500 rpm for 20min. After centrifugation, methanol extracts were filtered using Whatman filter paper No. 1. The resulting methanolic extracts were evaporated to dryness under water bath. Dissolved the dried saponins content in 5-10 ml of distilled water and transferred the solution into a separate pre-weighed container and freeze dried. About 10 mg of saponin extract were dissolve in 5ml of 80% aqueous methanol and 0.05ml of leaves extract solution was taken and added 0.25ml of vanillin reagent (8%). Test tubes were placed in ice cold water bath and 2.5ml of 72% sulphuric acid was added slowly on the inner side of the wall. After mixing the content in tube, these were left as such for 3 min. The test tubes were warmed 60°C for 10 min using a water bath and then cooled them in ice cold water bath. Absorbance was measured at 544nm. Plot absorbance against mg of saponin taken (10 to 100 µg/ml). Saponin was used as a reference standard and the content of total saponins was expressed as saponin equivalents (mg/gm extract).

Determination of terpenoids

Terpenoids were determined by the method of Indumathi *et al.*, (2014). Briefly 0.8 g of leaves dried powder was taken in a test tube and 10 ml of methanol was poured in it. The mixture was shaken well and filtered to take 5 ml extract. Then 2 ml of chloroform were mixed in extract of leaves sample and 3 ml of sulphuric acid were added in leaves extract and read the absorbance at 538 nm. The absorbance of all standards was determined on spectrophotometer and standard Linalool (20 to 80 µg/ml) graph was plotted. Terpenoids content was expressed as milligrams of Linalool equivalents per gram of dried sample.

GC MS Analysis

GC MS analysis was carried out on Shimadzu 2010 plus comprising a AOC-20i auto sampler and gas chromatograph interfaced to a mass spectrometer instrument. Total GC running time is 51.25min. The relative percentage amount of each component was calculated by comparing its average peak area to the total areas. Software adopted to handle mass spectra and chromatograms was a Turbo Mass Ver 5.2.0 (Srinivasan *et al.*, 2013). Interpretation on GCMS was conducted using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST and WILEY 7 library. The name, molecular weight and structure of the components of the test materials were ascertained (Dr. Dukes, 2013).

Hplc Analysis

Flavonoids fractions were analyzed by using a HPLC method (Weerasak Samee *et al.*, 2007; Paranthaman *et al.*, 2012). The HPLC analysis of hydro-ethanolic leaves extract were carried out with Chromatographic system (Shimadzu Class-VPV6.14SP2, Japan) consist of autosampler with 20µl fixed loop and an UV-Visible detector. All chromatographic data were recorded and processed using autochro-software.

UV-Visible Spectroscopic and FTIR analysis

The hydro-ethanolic extract of leaves was examined under UV and visible spectrophotometer analysis. The extract was scanned in the wavelength ranging from 200-1100nm using Perkin Elmer Spectrophotometer and the characteristic peaks were detected. FTIR analysis was performed using Spectrophotometer system, which was used to detect the characteristic peaks in ranging from 400-4000 cm⁻¹ and their functional groups. The peak values of the UV and FTIR were recorded.

Ultraviolet-visible spectroscopy analysis

The UV-VIS profile of leaves extract was studied over the 200 to 1100nm wavelength due to the sharpness of the peaks and proper baseline.

FTIR analysis

The FTIR spectrum was used to identify functional groups of the active components present in leaves samples based on the peak's values in the region of IR radiation. When the extract was passed into the FTIR, the functional groups of the components were separated based on its peak's ratio.

3. Results and Discussion

Qualitative analysis of *Tridax procumbens* leaves extract

Qualitative preliminary phytochemical analysis the results confirm in the presence of tannin, saponin, flavonoids, steroids, terpenoids, triterpenoids, alkaloids, anthroquinone, polyphenol, glycoside while anthocyanins was absent in ethanol, hydro-ethanolic and aqueous extract. The hexane and petroleum ether extracts showed the presence of steroids, terpenoids, triterpenoids, anthroquinone and polyphenol. Among the various extracts, the hydro-ethanolic extract contains higher concentration of phytochemical constituents (table1).

Table 1: Qualitative preliminary phytochemical analysis of leaves extract

S. No	Phytochemicals	Extracts				
		Aqueous	Ethanol	Hexane	Hydro-ethanolic	Petroleum ether
1	Tannin	++	++	-	++	-
2	Saponin	++	++	-	++	-
3	Flavonoids	++	++	-	++	-
4	Steroids	+	++	+	++	+
5	Terpenoids	++	++	+	++	+
6	Triterpenoids	++	++	+	++	+
7	Alkaloids	+	+	-	+	-
8	Anthroquinone	+	+	-	+	-
9	Polyphenol	++	++	+	++	+
10	Glycoside	++	++	+	++	+
11	Coumarins	+	-	-	++	-
12	Emodins	+	-	-	+	-
13	Anthocyanins	-	-	-	-	-

(-) Absent, (+) Present and (++) High concentration

Quantitative analysis of *Tridax procumbens* leaves extract

Preliminary quantitative phytochemical analysis of *Tridax procumbens* revealed that the leaves possessed high amount of total phenolic content of 206.90±14.48 GAE equivalents per gram, total flavonoid content of 182.50±12.77 milligrams of quercetin equivalents per gram, total steroids content of 158.28±11.07 milligrams of Cholesterol equivalents per gram, saponin content of 110.54±7.73 milligrams of saponin equivalents per gram and total terpenoids content of 91.43±6.40 milligrams of linalool equivalents per gram (**table2**). The results obtained suggest that the hydro-ethanolic leaves extract of *Tridax procumbens* has good phytochemical contents that play a vital role in its medicinal applications.

Table 2: Quantitative analysis of phenol, flavonoids, terpenoids, steroids and saponin content in the hydro-ethanolic extract

Name of Extract	Total phenol (Milligrams of Gallic acid (GAE) equivalents per gram)	Terpenoids (Milligrams of Linalool equivalents per gram)	Steroids (Milligrams of Cholesterol equivalents per gram)	Flavonoid (Milligrams of quercetin equivalents per gram)	Saponin (Milligrams of saponin equivalents per gram)
Hydro-ethanolic	206.90 ± 14.48	91.43 ± 6.40	158.28 ± 11.07	182.50 ± 12.77	110.54 ± 7.73

GC MS Analysis of *Tridax procumbens* leaves extract

Seventeen compounds were identified in hydro-ethanolic extract by GC-MS analysis. The active principles with their retention time (RT), molecular formula, molecular weight (MW) and concentration (%) are presented in Table 1. The prevailing compounds are Spirio-10- (2,11-dioxabicyclo [4.4.1], Phenol, 2,4-bis(1,1-dimethylethyl), Phthalic acid, 2-chloropropyl ethyl ester, 1,2- benzenedicarboxylic acid, diethyl ester, 2-(allyloxycarbonyl)benzoic acid, phytol, acetate, 3,7,11,15-tetramethyl-2hexadecen-1-ol, cyclopentadecanone, 2-hexadecen-1-ol, 3,7,11,15-tetramethyl, tetracyclononane, n-hexadecanoic acid, hexadecanoic acid, ethyl ester, phytol, 9,12-octadecadienoic acid, 9,12,15-octadecatrienoic acid, octadecanoic acid, ethyl linoleolate found in this sample. However, isolation of individual phytochemical constituents and subjecting its biological activity (table 3&4).

Table 3: Identification of active compounds in leaves extract using GCMS

Peak	R. Time	Area %	Height %	Molecular Formula	Molecular Weight	Name of the compounds
1	8.617	0.02	0.06	C14H20O3	236	Spirio-10-(2,11-dioxabicyclo[4.4.1])
2	8.839	0.43	1.02	C14 H22 O	206	Phenol, 2,4-bis(1,1-dimethylethyl)
3	9.928	2.26	7.14	C13H16O4	236	Phthalic acid, 2-chloropropyl ethyl ester
4	10.129	94.85	86.14	C12 H14 O4	222	1,2-Benzenedicarboxylic acid, diethyl ester
5	11.037	0.05	0.14	C11H10O4	206	2-(Allyloxycarbonyl)benzoic acid
6	12.525	0.12	0.39	C22H42O2	338	Phytol, acetate
7	12.795	0.02	0.07	C20H40O	296	3,7,11,15-Tetramethyl-2-hexadecen- 1-ol
8	12.906	0.01	0.06	C16H30O	238	Cyclopentadecanone,
9	13.001	0.03	0.10	C20 H40 O	296	2-Hexadecen-1-ol, 3,7,11,15-tetramethyl-, [R-[R*,R*
10	13.689	0.06	0.19	C18H30	246	Tetracyclo[6.1.0.0(2,4).0(5,7)]nonane, 3,6,9-tr
11	13.887	0.73	1.47	C16H32O2	256	n-Hexadecanoic acid
12	14.158	0.14	0.28	C18 H36 O2	284	Hexadecanoic acid, ethyl ester
13	15.438	0.78	2.10	C20H40O	296	Phytol
14	15.712	0.14	0.28	C18 H32 O2	280	9,12-Octadecadienoic acid
15	15.808	0.22	0.31	C18H30O2	278	9,12,15-Octadecatrienoic acid
16	15.946	0.10	0.17	C18 H36 O2	284	Octadecanoic acid
17	16.050	0.04	0.09	C20 H36 O2	308	Ethyl linoleolate

Table 4: Biological activity compounds identified in plant extract using GCMS

R. Time	Name of the compounds	Biological activity**
8.617	Spirio-10-(2,11-dioxabicyclo[4.4.1])	Anti-toxicity, Anticancer

8.839	Phenol, 2,4-bis(1,1-dimethylethyl)	Antimicrobial, Antifungal, Antioxidant, Antitumor
9.928	Phthalic acid, 2-chloropropyl ethyl ester	Antioxidant, Antimicrobial
10.129	1,2-Benzenedicarboxylic acid	Antimicrobial, Antifouling Anti cancer, Anti-inflammatory
12.525	Phytol, acetate	Hypocholesterolemic, Nematicide, Anticoronary, Antiarthritic, Hepatoprotective, Anti -androgenic
12.795	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	Antimicrobial, Anti-inflammatory
13.001	2-Hexadecen-1-ol, 3,7,11,15-tetramethyl	Antimicrobial, Cancer-preventive, Anti-inflammatory, Analgesic, Fungicide Antioxidant, Hypocholesterolemic Nematicide, Anti-Androgenic Flavour, Pesticide, Lubricant, Haemolytic 5-
13.887	n-Hexadecanoic acid	Alphareductase Inhibitor, antipsychotic, Potent Antibacterial Agent, Antimalarial And Antifungal
14.158	Hexadecanoic acid, ethyl ester	Antibacterial and antifungal activity
15.438	Phytol	Antimicrobial, Anticancer, Diuretic, Anti-inflammatory Hypocholesterolemic, Nematicide Antiarthritic, Hepatoprotective Anti androgenic, Hypocholesterolemic Nematicide,
15.712	9,12-Octadecadienoic acid	5-Alpha reductase inhibitor, Antihistaminic, Anticoronary Insectifuge, Antieczemic and Antiacne Anti-inflammatory, Insectifuge Hypocholesterolemic, Cancer preventive, Nematicide, Hepatoprotective, Insectifuge, Antihistaminic, Antieczemic, Antiacne, 5-
15.808	9,12,15-Octadecatrienoic acid	Alpha reductase inhibitor, Antiandrogenic, Antiarthritic and Anticoronary
15.946	Octadecanoic acid	Lower LDL Cholesterol level, Antioxidant and anti-inflammatory

**Source: Dr. Duke's phytochemical and ethnobotanical databases [Online database].

HPLC analysis of *Tridax procumbens* leaves extract

Figure 3 shows the chromatograms of the hydro-ethanolic leaves extract containing flavonoid compounds obtained at wavelengths of 280nm. Table 5 show the retention times of peaks for each flavonoid compound for every wavelength, respectively. Flavonoids present in the extract were identified by comparing chromatographic peaks with the retention time (Rt) with previous literature and identified the flavonoid compound present in the leaves extract. HPLC analysis of the hydro-ethanolic extract of leaves revealed that the presence of Catechin, Quercetin, Catechol, Luteolin, Viterxin- rahmnose, Rutin and Quercetin-3-galactoside.

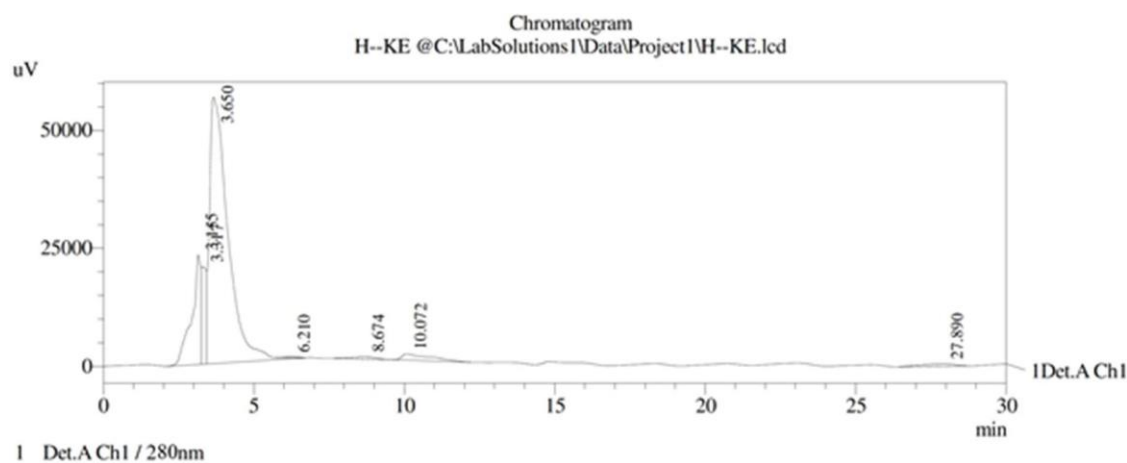


Figure 3: chromatogram of the leaves of *Tridax procumbens*

Table 5: HPLC profile of leaves extract

Peak	Ret. Time	Area	Height	Area	Peak	Ret. Time
1	3.155	496654	23295	15.112	22.576	Catechin
2	3.317	213916	20653	6.509	20.016	Quercetin
3	3.650	2404171	56581	73.152	54.835	Catechol
4	6.210	7714	242	0.235	0.235	Luteolin
5	8.674	28414	525	0.865	0.509	Viterxin- rahmnose
6	10.072	91561	1338	2.786	1.296	Rutin
7	27.890	44106	550	1.342	0.533	Quercetin- 3- galactoside
Total		3286535	103185	100.000	100.000	

UV-Visible Spectroscopic and FTIR analysis of *Tridax procumbens* leaves extract

The UV-VIS profile showed the peaks at 208.23, 416.24 and 658.22 with the absorption 3.970, 0.413 and 0.160 respectively (**figure 4 and table-6**). The UV-VIS spectroscopy revealed the characteristic peaks present in the leaves extract.

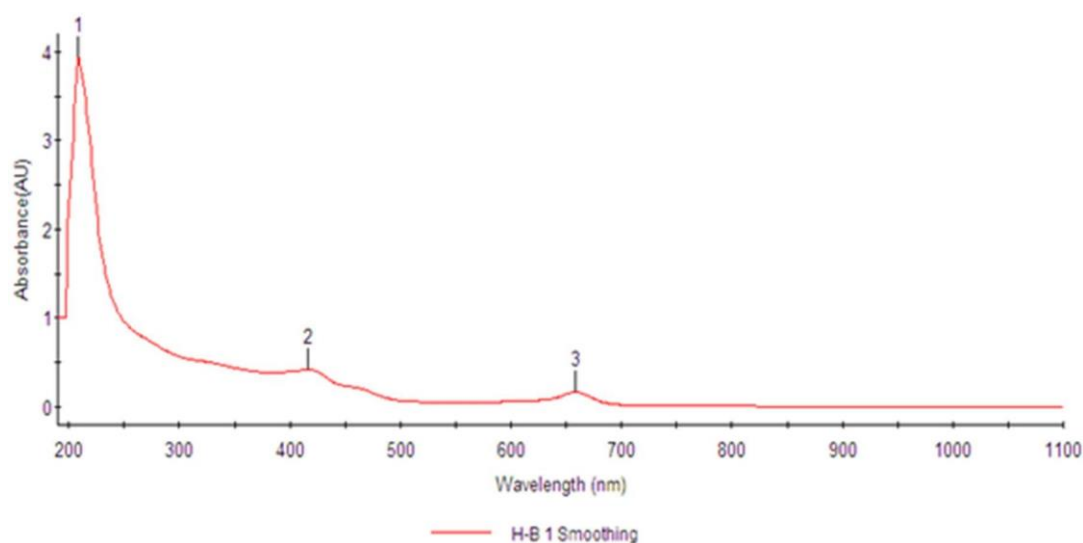


Figure 4: UV-Visible spectrum analysis of leaves extract

Table 6: UV-Visible spectrum analysis of Plant extract

Peaks	Wavelength (nm)	Absorbance (AU)
1	208.23	3.970
2	416.24	0.413
3	658.22	0.160

The results of FTIR peak values and functional groups were represented in figure 5 and table In the current investigation involving Plant extract, the results of FTIR analysis have confirmed the presence of alcohols, phenols, carboxylic acids, 1° amines, aromatics, aromatic amines, aliphatic amines and alkynes groups.

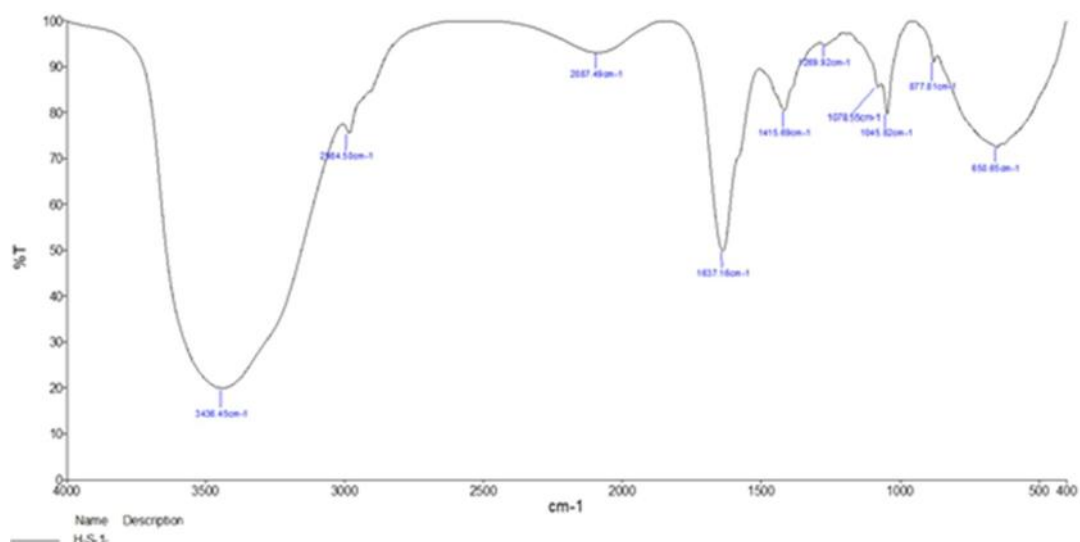


Figure 5: FTIR spectrum analysis of leaves extract

Table 7: FTIR spectrum analysis of leaves extract

Frequency cm-1	Bond	Functional group
3436.45	O-H stretch, H-bonded	Alcohols, Phenols
2984.50	O-H stretch	Carboxylic acids
1637.16	N-H bend	1° amines
1415.69	C-C stretch (in-ring)	Aromatics
1269.92	C-N stretch	Aromatic amines
1078.55, 1045.82	C-N stretch	Aliphatic amines
877.81	C-H "loop"	Aromatics
650.65	-C \equiv C-H: C-H bend	Alkynes

Ikewuchi Jude *et al.*, (2009) was reported six phytochemicals from the leaves of *Tridax procumbens* Linn. Ayyappa Das *et al.*, (2009) was calculated eight secondary metabolites from the aqueous and methanolic leaf extract of *Tridax procumbens* Linn. Dhanabalan *et al.*, (2008) also shows the presence of eight phytochemicals as Alkaloids, Tannin, Saponin, Steroid, Terpenoids, Flavonoids and Cardiac glycosides form the Methanolic extract of leaves of *T. procumbens* Linn. Dillard CJ *et al.*, (2000) was reported the biological activities of flavonoids are actions against free radical mediated cellular signalling, free radicals, inflammation, allergies, platelet aggregation, microbes, ulcers, viruses, tumours and hepatotoxins. Luteolin has antibacterial, anti-inflammatory, anti-mutagenic, antioxidant, anti-allergic, anti-andro-genic, anticancer, anti-diabetic, anti-estrogenic, antimicrobial, hypocholesterolemic, hypotensive, neuroprotective and radio-protective activities. Sutherland *et al.*, (2006) was reported the catechins are antimicrobial, anti-allergic, anti- carcinogenic, anti-proliferative, anti-diabetic, antihypertensive, anti-inflammatory, anti- mutagenic, anti-obesity, antioxidant, anti-platelet, anti-tumorigenic, antiulcer, chemo- preventive, hypocholesterolemic and neuroprotective agents. Dillard CJ *et al.*, (2000) & López-Lázaro *et al.*, (2009) was observed that luteolin has antibacterial, anticancer, anti-inflammatory, anti-mutagenic, antioxidant, anti-allergic, anti- androgenic, anti-diabetic, anti-estrogenic, antimicrobial, hypocholesterolemic, hypotensive, neuroprotective and radio-protective activities. Arnab Bera *et al.*, (2023) was reported that the methanolic extract of *Tridax procumbens* exhibits strong antioxidant activity and was found to correlate with the phenolic and flavonoid content positively and also antibacterial potential against *Staphylococcus aureus*

4. Conclusion

Tridax procumbens Linn. is found throughout India. The plant is widely distributed and found to have noble pharmacological activities. The study concluded that the leaves exhibited potential pharmacological properties due to the presence of phytochemicals such as tannin, saponin, flavonoids, steroids, terpenoids, triterpenoids, alkaloids, anthraquinone, polyphenol, glycoside, coumarins and emodin and bioactive compounds which can be used for therapeutic purposes.

Conflict of Interest

There are no conflicts of Interest

References:

1. Srinivasan K, Sivasubramanian S and Kumaravel S. Phytochemical profiling and GC- MS study of *Adhatoda vasica* leaves. *International Journal of Pharma and Bio Sciences*, 2013; 5(1):714-720.
2. Tapan seal Quantitative HPLC analysis of phenolic acids, flavonoids and ascorbic acid in four different solvent extracts of two wild edible leaves, *Sonchus arvensis* and *Oenanthe linearis* of North-Eastern region in India. *Journal of Applied Pharmaceutical Science*, 2016 Vol. 6 (02), pp. 157-166.
3. Baram Ahmed Hamahameen and Banaz Jamal. Determination of Flavonoids in the Leaves of Hawthorn (*Crataegus Azarolus*) of Iraqi Kurdistan Region by HPLC Analysis. *international Journal of Bioscience, Biochemistry and Bioinformatics*, 2013 Vol. 3, No. 1, 67-71.
4. Weerasak Samee, Suwanna Vorarat (2007) Simultaneous Determination of Gallic acid, Catechin, Rutin, Ellagic Acid and Quercetin in Flower Extracts of *Michelia alba*, *Caesalpinia pulcherrima* and *Nelumbo nucifera* by HPLC. *Thai Pharmaceutical and Health Science Journal* 2:131-137.
5. Paranthaman R, Praveen kumar P, Kumaravel S (2012) GC-MS Analysis of Phytochemicals and Simultaneous Determination of Flavonoids in *Amaranthus caudatus* (Sirukeerai) by RP-HPLC. *Journal of Analytical and Bioanalytical Techniques* 3:147.
6. Harborne J.B. (1973). *Phytochemical Methods; A guide to modern techniques of plant Analysis*. 2nd Edition, London New York.
7. Harborne J.B. (1984). *Phytochemical Methods; A guide to modern techniques of plant* 2nd Edition, London New York.
8. Sofowara A (1993). *Medicinal plants and Traditional medicine in Africa*. Spectrum Books Ltd, Ibadan, Nigeria. p. 289.
9. Trease GE and Evans WC. (1989) *Pharmacognosy*. 11th edn. Brailliar Tiridel Can. Macmillian publishers.
10. Attarde Daksha, Pawar Jaywant, Chaudhari Bhagyashree, Pal Subodh. (2010) Estimation of sterols content in edible oil and ghee samples. *International Journal of Pharmaceutical Sciences Review and Research*. 5(1): 135-139.
11. Indumathi C, Durgadevi G, Nithyavani S, Gayathri P K. Estimation of terpenoid content and its antimicrobial property in *Enicostemma litorale*. *International Journal of ChemTech Research*. 2014,6(9), pp 4264-4267.
12. Hiai, S., Oura, H. and Nakajima, T. 1976. Color reactions of some saponins and saponins with vanillin and sulphuric acid. *Planta Medica*. 29: 116-122.
13. Katasani D. Phytochemical screening, quantitative estimation of total phenolic, flavonoids and antimicrobial evaluation of *Trachyspermum ammi*. *J Atoms and Molecules*. 2011; 1:1-8.
14. Kim DO, Jeong SW, Lee CY. Antioxidant capacity of phenolic phytochemicals from various cultivars of plums. *Food Chem*. 2003; 81:321-326.
15. Dr. Duke's Phytochemical and Ethno botanical Databases, *Phytochemical and Ethnobotanical Databases*. www.ars-gov/cgi-bin/duke/. 2013.
16. Kushwaha P, Yadav SS, Singh V and Dwivedi LK: Phytochemical screening and GC- MS studies of the methanolic extract of *Tridax procumbens*. *Int J Pharm Sci & Res* 2019; 10(5): 2492-96. doi: 10.13040/IJPSR.0975-8232.10(5).2492-96.
17. C.I. Kewuchi Jude, C.I Kewuchi Catherine and M. Igboh Ngozi: Chemical profile of *Tridax procumbens* Linn, *Pakistan Journal of Nutrition* 8:548-550, (2009).
18. M.P. Ayyappa Das, R. Dhanabalan, A. Doss and M. Palaniswamy: Phytochemical screening and Antibacterial Activity of aqueous and Methanolic extract of two medicinal plants against Bovine Mastitis Bacterial Pathogens. *Ethnobotanical leaflets* 13:131-139 (2009).
19. R. Dhanabalan, A. Doss, M. Jagadeeswari, S. Balachandar, E. Kezia, V. Parivuguna, C.M. Reena Josephine, R. Vaidheki and K. Kalamani: In vitro Phytochemical Screening and Antibacterial Activity of Aqueous and Methanolic Leaf Extracts of *Tridax procumbens* against Bovine Mastitis Isolated *Staphylococcus aureus*, *Ethnobotanical Leaflets* 12: 1090-95, (2008).
20. Sawant RS, Godghate AG. Preliminary phytochemical analysis of leaves of *Tridax procumbens* Linn. *International Journal of Science, Environment and Technology*. 2013;2(3):388-94.
21. Dillard CJ, German JB. Phytochemicals: nutraceuticals and human health. *Journal of the Science of Food and Agriculture*. 2000 Sep 15;80(12):1744-56.
22. López-Lázaro, M. (2009) Distribution and Biological Activities of the Flavonoid Luteolin. *Mini-Reviews in Medicinal Chemistry*, 9, 31-59.

23. Sutherland, B.A., Rahman, R.M.A. and Appleton, I. (2006) Mechanisms of Action of Green Tea Catechins, with a Focus on Ischemia-Induced Neurodegeneration. *Journal of Nutritional Biochemistry*, 17, 291-306 <http://dx.doi.org/10.1016/j.jnutbio.2005.10-005>
24. Arnab Bera and Nilanjana Banerjee (2023). Estimation of Total Phenolic and Flavonoid content, Antibacterial and Antioxidant Potential of *Tridax procumbens* Linn. from Paschim Medinipur. *Biological Forum – An International Journal*, 15(5): 933-936.