Current Pharma Research
ISSN-2230-7842
CODEN-CPRUE6
www.jcpronline.in/

Research Article

Preliminary Phytochemical Screening and HPTLC Finger printing analysis of Leaf Extracts of *Dendrophthoe Falcate (L)* ***and*** *Tridax procumbens* (L).

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Received 12 September 2017; received in revised form 24 September 2017; accepted 24 September2017

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ABSTRACT

Objective: The present study was aimed to evaluate physical constants and develop the HPTLC finger print profile of leaf extracts of *Dendrophthoe Falcate* (L) *andTridax procumbens* (L). Methods: HPTLC studies were carried out using CAMAG HPTLC system equipped with Linomat IV applicator Results: Extract showed the presence of alkaloids, flavonoids, terpenoids, tannins, saponins, glycosides, phenolic compounds. HPTLC finger printing analysis of leaf extracts of *Tridax procumbens (TP)* and *Dendrophthoe Falcate (DF) revealed* presence phytoconstituents. TPMW extract showed 9 peaks at 366 nm with Rf values 0.03-0.85, TPEW extract showed 10 peaks at 366 nm with Rf values 0.03-0.82, TPDM extract showed 9 peaks at 254 nm with Rf values 0.04-0.82, at 366 nm showed 12 peaks with Rf values 0.33- 0.83 & at 560 nm showed 9 peaks with Rf values 0.03- 0.67. DFDM extract showed 11 peaks at 366 nm with Rf values 0.06-0.87 DFMW extract showed 7 peaks with Rf values0.11 - 0.90. DFEW extract showed 6 peaks with Rf values 0.01-0.89 showed maximum concentration. The chromatogram shows presence of multiple peaks which indicate diverse composition of extract. Conclusions: It can be concluded that HPTLC fingerprint analysis of leaf extracts of *Tridax procumbens* and *Dendrophthoe Falcate* can be used as a diagnostic tool for the correct identification of the plant, phytoconstituents and it was appropriate method for standardization of the extract.

KEYWORDS

Dendrophthoe Falcate (L) *andTridax procumbens* (L), Phytochemical Screening, Physical constants, HPTLC Fingerprinting.

1. INTRODUCTION

Indian medicinal plants possess various pharmacological activities. Not all the medicinal plants used in Ayurvedic formulations have been investigated in detail. The World Health Organization (WHO) defines traditional medicine as the "diverse health practices, approaches, knowledge and beliefs incorporating plant, animal or mineral based medicines, spiritual therapies, manual techniques and exercises applied singularly or in combination to maintain well-being, as well as to treat, diagnose, or prevent illness". It is clear, however, that there is a need to validate the information through an organised infrastructure for it to be used as an effective therapeutic means, either in conjunction with existing therapies, or as a tool in novel drug discovery. Traditional medicine utilizes biological resources and the indigenous knowledge of traditional plant groups (Timmermans, K 2003). Medicinal plants often contain additional active principles other than the major active principles and physiologically inert substances like cellulose and starch. As the constituents derived from the medicinal plants proved the cure the medicinal plants proved to cure the human disorders they isolated and used for their pharmacological action. The constituents having particular therapeutic effect are identified and isolated (Ali M 1990).

Standardization of plant crude material is becoming today's necessity. Products of primary metabolism such as amino acids, carbohydrates and proteins are vital for the maintenance of life processes, while others like alkaloids, flavonoids, terpenoids, tannins, saponins, glycosides, phenolic compounds are products of secondary metabolism and have toxicological, pharmacological and ecological importance (Sharma RK, Bhagwan D, et al,1996). Standardization and quality control of herbal drugs is very complicated because herbal products contain a group of phytoconstituents and are very capable of variation. There is the variability within the same plant material or between the different parts of the same plant. The variability may be from grower to grower, crop to crop and also depends on the harvest and post-harvest handling. On the other hand herbal drugs have multiple phytoconstituents including active, inactive, unknown which are dietary rather than therapeutic (Gupta A.K, 2004). Identification and quality evaluation of crude herbal extracts is a fundamental requirement. It is an accepted fact that the qualitative analysis of crude herbal extracts constitutes an important and reliable part of quality control protocol. Various extraction methods and analytical methods as spectrophotometry, high performance thin layer chromatography (HPTLC), high performance liquid chromatography (HPLC), gas chromatography-mass spectrometry (GC–MS) and (FT-IR) are developed for the study about plant active compounds (Kirtikar KR, Basu BD, 2005).

Chromatographic fingerprinting techniques are most significant methods which can be used for the routine herbal drug analysis and for quality assurance. High-performance thin layer chromatography (HPTLC) based methods could be considered as a good alternative, as they are being explored as an important tool in routine drug analysis that can generate a fingerprint of each extract in large collections would be useful to detect stability of the same extract over time. In the present study the plant *Dendrophthoe falcate (L)* is branched hemiparasite. The whole parasitic plant is used in indigenous system of medicine as cooling, bitter tonic, astringent, aphrodisiac, narcotic, diuretic, pulmonary tuberculosis, asthma, menstrual disorders, swelling wounds, ulcers, renal and vesicle calculi and vilated conditions of kapha and pitta (Nadkarni KM

1993). *Dendrophthoe falcata* belonging to the family *Loranthaceae* is an angiospermic hemiparasitic plant was most frequently observed on many host plants comprises of 20 species and about 7 species are found in India. *Dendrophthoe falcata* bark juice/decoction used in menstrual troubles and asthma while its paste is applied on boils, setting dislocated bones and extracting pus. The decoction of whole plant is used to treat joint pains (Jagtap S.D et al, 2006) and leaf juice is used for relief from chest pain
(Sandhya B; Thomas S; et al 2006) Biological activities of *Dendrophthoe falcata* are antioxidant, anti-inflammatory (Satish patil, Sneha Anarthe et al, 2011), wound healing and anti-microbial (S.P.Pattanayak, 2008). *Tridax procumbens (L*). family Compositae extensively used in ayurvedic system of medicine for various ailments and is dispensed for "Bhringraj" by some of the practitioners of ayurveda which is well known medicine for liver disorders (D. A. Bhagwat, et al l2008). *Tridax* possesses significant anti-inflammatory, hepatoprotective, wound healing, antidiabetic activity and antimicrobial activity against both gram-positive and gram-negative bacteria (R.B. Mahato, R.P. Chaudhary, 2005). The present research deals with the phytochemical investigation of and development of HPTLC fingerprints of Leaf Extracts of *Dendrophthoe Falcate* (L) *andTridax procumbens* (L) which can be used for identification, authentication and characterization. HPTLC offers better resolution and estimation of active phytoconstituents can be done with accuracy in a shorter time.

2. MATERIALS AND METHODS

2.1. Collection and Identification of Plant Material

The *Tridax procumbens* L. (Asteraceae) *plant and Dendrophthoe falcata (L.f)Etting.*(Loranthaceae) samples were collected at flowering stage from local region during September – November were collected from Kapurhol Satara-pune NH4, kasurdi, Bhor, pune Maharashtra (India). Both specimens plant were identified and authenticated by Botanical Survey of India (BSI) Pune.

2.2. Preparation of Plant Extract

Extracts were prepared in a sequential manner using ethanol: water, methanol: water, dichloromethane: methanol (order of increasing polarity) as solvents from of shade dried and coarsely powdered plant material using the maceration and soxhlet apparatus. Powder was defatted with petroleum ether (60-80 °C) and then exhaustively extracted with different solvent. The methanol: water and ethanol: water extract was prepared by maceration by soaking 10 g of powdered plant materials in 100 ml of solvent in conical flask at room temperature for 24 hr. Conical flask was allowed to stand for 30 mins in a water bath (at 100°C) with occasional shaking followed by rotary shaker at 200 rpm for 24h Extract was filtered after 48 hr., through a sterilized Whatmann No. 1 filter paper. The extract concentrated using a rotary vacuum evaporator with the water bath set at 40°C. The dried extract thus, obtained was sterilized by overnight UV-irradiation.

The sterile extract was transferred into a sterile lyophilisation flask & frozen in a deep freezer. The extract was stored at -20^0C till bioevaluation.

2.3. Preliminary Phytochemical Screening

The phytochemical investigation of the different extracts of *Tridax procumbens(L*) and *Dendrophthoe falcata (L.f)* was carried out with standard protocol (W.H.O., Geneva; 1989). The extracts were finally weighed. The phytochemical tests were performed on the liquid and dried extracts using standard methods (Sharma. M.K, Sharma S. 2010).The results are presented in Table 1.

2.4. Evaluation of Physical Constants

The extracts were finally weighed. The phytochemical tests were performed on the liquid and dried extracts using standard methods and the physical constants were evaluated. Foreign Matter, Moisture Content, Total Ash Value, Water Soluble Ash Value, Acid Insoluble Ash Value , Soluble, Extractive Value in Water, Chloroform, Methanol and Ethanol were carried out (Khandelwal KR. 2007),The results are stated in Table 2.

2.5. HPTLC Profile (High Performance Thin Layer Chromatography)

HPTLC is a sophisticated and automated form of TLC. HPTLC is the fastest of all chromatographic methods. HPTLC is an important analytical tool in the separation, identification and estimation of various classes of natural Phytoconstituents. HPTLC studies were carried out following the method (Harborne JB. 1998), (Wagner H, Baldt S.1996).

2.6. Sample Preparation and Application

5 mg/ml concentration of extracts were prepared in respective solvents of chromatographic grade and then filtered by whatmann filter paper No. 1. Prepared samples of different extracts were applied on TLC aluminium sheets silica gel 60 F 254 (Merck) 10µl each with band length of 5 mm using Linomat 5 sample applicator attached to CAMAG HPTLC system, which was programmed through WIN CATS software.

2.7. Development of Chromatogram

The chromatograms were developed in CAMAG twin trough chamber saturated with solvent n-hexane:Toluene: ethyl acetate (2: 4: 1)for 20 minutes up to thedistance of 80 mm.

2.8. Scanning of the Chromatogram

CAMAG HPTLC Densitometer (Scanner) was used as a scanner in absorbance mode at both 254 and 366 nm, using deuterium and tungsten lamp (slit dimension 6.0 X 0.45 macro). The scanned data was subjected for integration through the software winCATS Planar Chromatography Manager. The fingerprint so developed was used for the detection of phytocomponents present in the samples and the chromatograms and Rf value were noted. Bands were resolved and their colour was noted. Spots were visible without derivatization at 254nm, 366 nm wavelengths.

3. RESULTS AND DISCUSSION

3.1. Phytochemical Screening

The phytochemical test on ethanol: water, methanol: water & dichloromethane: methanol extracts of *Tridax procumbens (L*) and *Dendrophthoe falcata (L) leaf* powder showed the presence of various phytoconstituents like alkaloid, carbohydrate, glycoside, steroid, protein, tannin, terpenoids, flavonoids and phenolic compounds. (Table 1)

Table 1. Preliminary Photochemical Screening of Various Extracts of *Tridax procumbens (L).*and *Dendrophthoe falcata (L).*

Phyto-constituents	***Tridax procumbens* Extracts**			***Dendrophthoe falcata* Extracts**		
	Methanol: Water	Ethanol: water	Dichloromethane: Methanol	Methanol Water	Ethanol: Water	Dichloromethane: Methanol
Alkaloids	-	+++	++	+	-	+
Flavonoids	++	+++	+++	++	+	+++
Phenolic groups	+	+	-	-	-	-
Saponin Glycosides	++	+++	+	++	+	-
Tannins	+++	+++	+++	+++	+++	+++
Steroids	++	+++	++	++	++	++
Carbohydrates	+	+	+	+	+	+
Terpenoids	+	++	+	+	++	+
Cardiac Glycosides	-	-	-	+	+	+
Reducing sugar	+	-	++	+	-	+
Anthroquinone Glycosides	_	_	_	_	_	+

3.2. Evaluation of Physical Constants

The proximate analysis showed satisfactory result with respect to foreign matter, moisture content, Ash value and extractive values. The physical constants are given in Table.2

Table 2: Evaluation of Physical constants of powdered *Tridax procumbens (L)* and *Dendrophthoe falcata (L)*

Sr. No	Evaluation Parameter	Value (%)	
		Tridax procumbens(L)	***Dendrophthoe falcata.(L)***
1	Foreign Matter	1	1
2	Moisture Content	10.6	9.5
3	Total Ash Value	12.7	11.9
4	Water Soluble Ash Value	3.5	3.2
5	Acid Insoluble Ash Value	8.0	9.1

6	Water Soluble Extractive Value	4.89	3.87
7	Chloroform Soluble Extractive Value	0.4	0.5
8	Methanol Soluble Extractive Value	6.66	6.73
9	Ethanol Soluble Extractive Value	4	5.2

Table 3: Preliminary Photochemical Screening of Various Extracts of *Tridax procumbens (L)* and *Dendrophthoe falcata (L).*

Phyto-constituents	***Tridax procumbens* Extracts**			***Dendrophthoe falcata* Extracts**		
	Methanol : Water	Ethanol: water	Dichloromethane: Methanol	Methanol Water	Ethanol: Water	Dichloromethane: Methanol
Alkaloids	-	+++	++	+	-	+
Flavonoids	++	+++	+++	++	+	+++
Phenolic groups	+	+	-	-	-	-
Saponin Glycosides	++	+++	+	++	+	-
Tannins	+++	+++	+++	+++	+++	+++
Steroids	++	+++	++	++	++	++
Carbohydrates	+	+	+	+	+	+
Terpenoids	+	++	+	+	++	+
Cardiac Glycosides	-	-	-	+	+	+
Reducing sugar	+	-	++	+	-	+
Anthroquinone Glycosides	_	_	_	_	_	+

3.3. Evaluation of Physical Constants:

The proximate analysis showed satisfactory result with respect to foreign matter, moisture content, Ash value and extractive values. The physical constants are given in Table 2.

Table 4: Evaluation of Physical constants of powdered *Tridax procumbens (L)* and *Dendrophthoe falcata (L)*

Sr. No	Evaluation Parameter	Value (%)	
		Tridax procumbens(L)	***Dendrophthoe falcata.(L)***
1	Foreign Matter	1	1
2	Moisture Content	10.6	9.5

3	Total Ash Value	12.7	11.9
4	Water Soluble Ash Value	3.5	3.2
5	Acid Insoluble Ash Value	8.0	9.1
6	Water Soluble Extractive Value	4.89	3.87
7	Chloroform Soluble Extractive Value	0.4	0.5
8	Methanol Soluble Extractive Value	6.66	6.73
9	Ethanol Soluble Extractive Value	4	5.2

3.4. HPTLC studies

HPTLC studies of Tridax procumbens (L) extract.

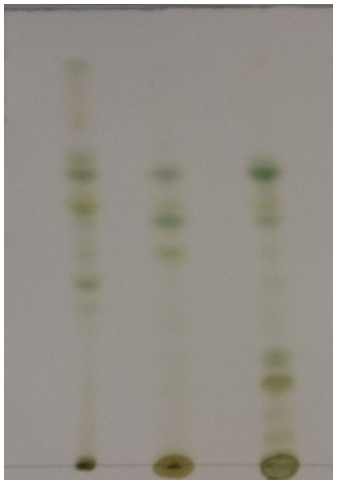

Fig.1. TLC plate showing different constituents of Methanol water, Ethanol: water Dichloromethane: Methanol extract of *Tridax procumbens* (leaf) (Track 1-3).

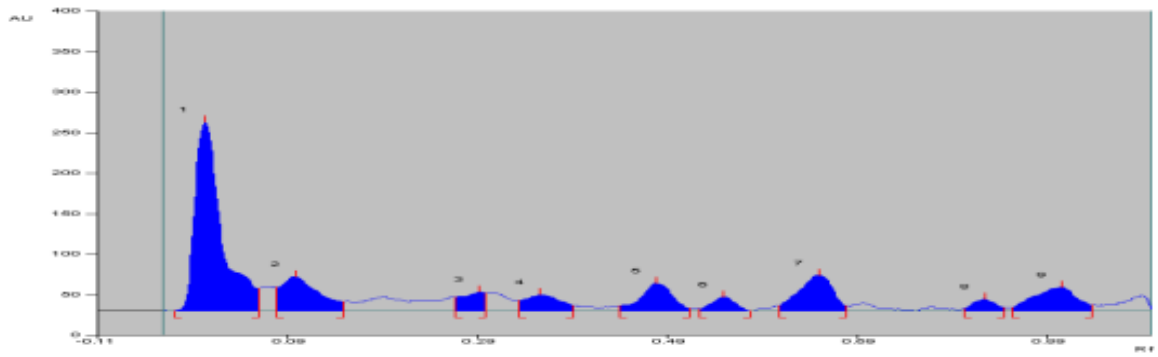

Fig. 2. Chromatogram of methanol water extract of *Tridax procumbens* (leaf) measured at 366nm.

Table 3. Rf values for methanol water extract of *Tridax procumbens* (leaf).

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	-0.03 Rf	0.2 AU	0.00 Rf	233.4 AU	50.87 %	0.06 Rf	27.4 AU	5374.6 AU	45.29 %
2	0.08 Rf	29.3 AU	0.10 Rf	42.5 AU	9.26 %	0.15 Rf	11.7 AU	1399.1 AU	11.79 %
3	0.27 Rf	16.9 AU	0.29 Rf	23.6 AU	5.15 %	0.30 Rf	21.8 AU	503.0 AU	4.24 %
4	0.33 Rf	12.7 AU	0.36 Rf	20.0 AU	4.35 %	0.39 Rf	6.8 AU	621.0 AU	5.23 %
5	0.44 Rf	6.1 AU	0.48 Rf	34.1 AU	7.43 %	0.51 Rf	3.1 AU	929.6 AU	7.83 %
6	0.52 Rf	2.7 AU	0.55 Rf	17.2 AU	3.76 %	0.58 Rf	0.0 AU	347.0 AU	2.92 %
7	0.61 Rf	6.4 AU	0.65 Rf	43.9 AU	9.57 %	0.68 Rf	6.7 AU	1306.1 AU	11.01 %
8	0.80 Rf	2.6 AU	0.82 Rf	14.8 AU	3.22 %	0.84 Rf	4.3 AU	287.9 AU	2.43 %
9	0.85 Rf	4.1 AU	0.90 Rf	29.4 AU	6.40 %	0.94 Rf	7.3 AU	1098.4 AU	9.26 %

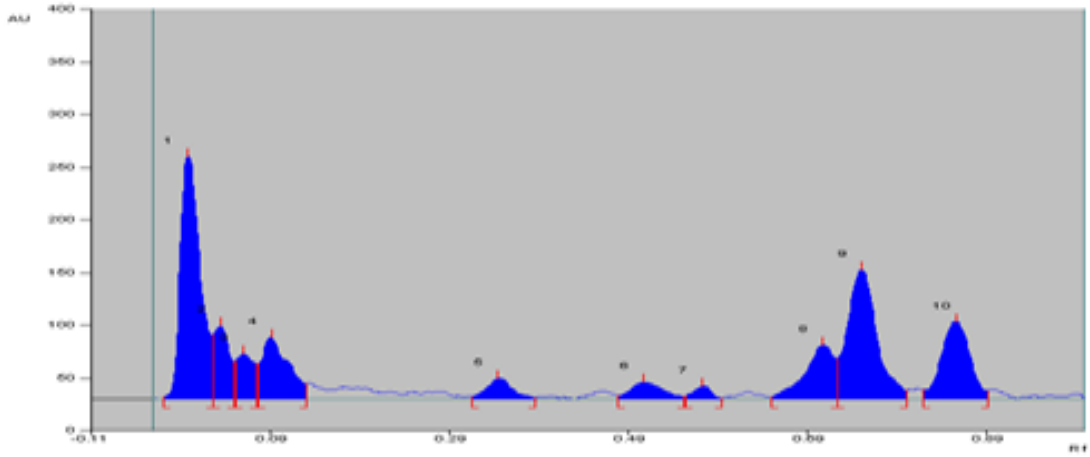

Fig. 3. Chromatogram of Ethanol: water extract of *Tridax procumbens* (leaf) measured at 366nm.

Table 4. Rf values for Ethanol: water **extract** of *Tridax procumbens* (leaf).

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	-0.03 Rf	2.0 AU	-0.00 Rf	230.4 AU	33.08 %	0.03 Rf	59.8 AU	4269.2 AU	28.12 %
2	0.03 Rf	60.7 AU	0.03 Rf	69.6 AU	9.99 %	0.05 Rf	35.1 AU	958.2 AU	6.31 %
3	0.05 Rf	35.5 AU	0.06 Rf	43.1 AU	6.18 %	0.07 Rf	33.1 AU	726.0 AU	4.78 %
4	0.08 Rf	33.3 AU	0.09 Rf	58.7 AU	8.42 %	0.13 Rf	14.4 AU	1436.2 AU	9.46 %
5	0.31 Rf	2.0 AU	0.34 Rf	19.3 AU	2.77 %	0.38 Rf	1.1 AU	466.4 AU	3.07 %
6	0.48 Rf	3.3 AU	0.51 Rf	16.0 AU	2.30 %	0.55 Rf	2.3 AU	477.6 AU	3.15 %
7	0.55 Rf	2.4 AU	0.57 Rf	12.1 AU	1.74 %	0.59 Rf	1.0 AU	204.1 AU	1.34 %
8	0.65 Rf	1.3 AU	0.71 Rf	51.3 AU	7.37 %	0.72 Rf	38.1 AU	1384.3 AU	9.12 %
9	0.72 Rf	38.5 AU	0.75 Rf	122.5 AU	17.59 %	0.80 Rf	8.1 AU	3314.2 AU	21.83 %
10	0.82 Rf	8.8 AU	0.86 Rf	73.5 AU	10.56 %	0.89 Rf	7.6 AU	1944.4 AU	12.81 %

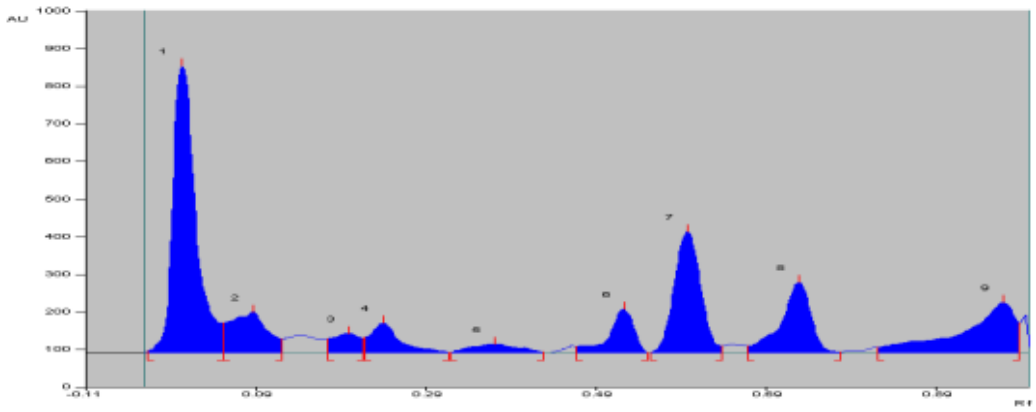

Fig. 4. Chromatogram of Dichloromethane: Methanol extract of *Tridax procumbens* (leaf) measured at 254 nm.

Table 5. Rf values for Dichloromethane: Methanol extract of *Tridax procumbens* (leaf). at 254 nm.

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	-0.04 Rf	5.1 AU	0.00 Rf	762.4 AU	42.84 %	0.05 Rf	78.7 AU	16961.6 AU	34.79 %
2	0.05 Rf	79.1 AU	0.09 Rf	108.5 AU	6.10 %	0.12 Rf	37.3 AU	3912.4 AU	8.03 %
3	0.17 Rf	37.5 AU	0.20 Rf	51.5 AU	2.89 %	0.22 Rf	39.4 AU	1377.5 AU	2.83 %
4	0.22 Rf	39.7 AU	0.24 Rf	78.3 AU	4.40 %	0.32 Rf	0.4 AU	2255.3 AU	4.63 %
5	0.32 Rf	0.7 AU	0.37 Rf	22.4 AU	1.26 %	0.43 Rf	1.6 AU	1060.3 AU	2.17 %
6	0.47 Rf	16.8 AU	0.52 Rf	115.8 AU	6.50 %	0.55 Rf	0.1 AU	2785.8 AU	5.71 %
7	0.55 Rf	0.5 AU	0.60 Rf	321.1 AU	18.05 %	0.64 Rf	18.2 AU	8184.9 AU	16.79 %
8	0.67 Rf	17.7 AU	0.73 Rf	186.3 AU	10.47 %	0.78 Rf	2.3 AU	5375.7 AU	11.03 %
9	0.82 Rf	13.7 AU	0.97 Rf	133.3 AU	7.49 %	0.99 Rf	78.6 AU	6838.9 AU	14.03 %

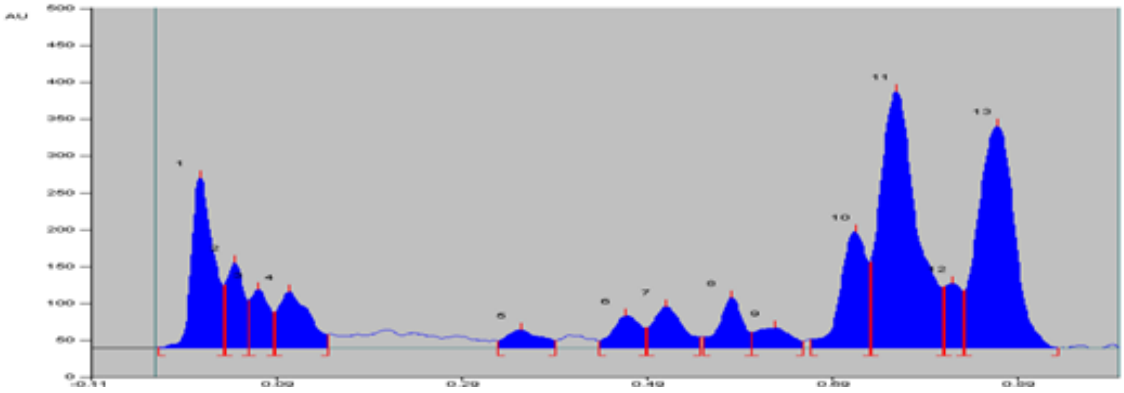

Fig. 5. Chromatogram of Dichloromethane: Methanol extract of *Tridax procumbens* (leaf) measured at 366nm.

Table 6. Rf values for Dichloromethane: Methanol extract of *Tridax procumbens* (leaf).

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	-0.04 Rf	0.9 AU	0.01 Rf	230.9 AU	14.29 %	0.03 Rf	34.7 AU	4419.5 AU	10.70 %
2	0.03 Rf	85.4 AU	0.04 Rf	115.6 AU	7.16 %	0.06 Rf	53.6 AU	1773.2 AU	4.29 %
3	0.06 Rf	64.1 AU	0.07 Rf	79.4 AU	4.92 %	0.09 Rf	48.6 AU	1326.9 AU	3.21 %
4	0.09 Rf	48.7 AU	0.10 Rf	76.1 AU	4.71 %	0.14 Rf	18.6 AU	2089.6 AU	5.06 %
5	0.33 Rf	9.4 AU	0.35 Rf	24.6 AU	1.52 %	0.39 Rf	10.3 AU	771.0 AU	1.87 %
6	0.44 Rf	9.4 AU	0.47 Rf	44.0 AU	2.72 %	0.49 Rf	27.1 AU	1083.5 AU	2.62 %
7	0.49 Rf	27.3 AU	0.51 Rf	56.0 AU	3.47 %	0.55 Rf	14.7 AU	1492.8 AU	3.62 %
8	0.55 Rf	15.1 AU	0.58 Rf	68.6 AU	4.24 %	0.60 Rf	21.1 AU	1393.0 AU	3.37 %
9	0.60 Rf	21.2 AU	0.63 Rf	27.5 AU	1.70 %	0.66 Rf	8.7 AU	850.6 AU	2.06 %
10	0.67 Rf	11.6 AU	0.71 Rf	157.4 AU	9.75 %	0.73 Rf	15.1 AU	3633.1 AU	8.80 %
11	0.73 Rf	116.4 AU	0.76 Rf	347.1 AU	21.49 %	0.81 Rf	32.7 AU	11437.8 AU	27.70 %
12	0.81 Rf	82.9 AU	0.82 Rf	87.6 AU	5.42 %	0.83 Rf	77.5 AU	1341.3 AU	3.25 %
13	0.83 Rf	78.0 AU	0.87 Rf	300.4 AU	18.60 %	0.93 Rf	0.0 AU	9674.4 AU	23.43 %

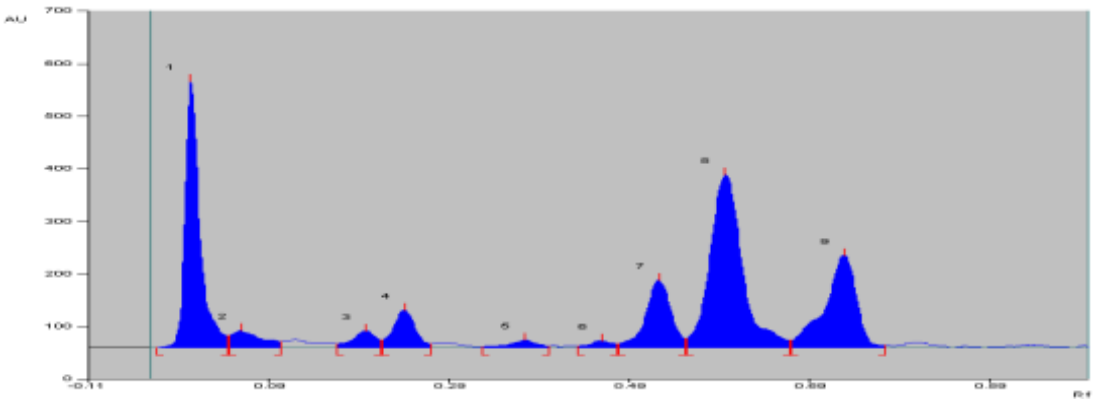

Fig. 6. Chromatogram of Dichloromethane: Methanol extract of *Tridax procumbens* (leaf) measured at 560 nm.

Table 7. Rf values for Dichloromethane: Methanol extract of *Tridax procumbens* (leaf). at 560 nm.

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	-0.03 Rf	0.4 AU	0.00 Rf	506.7 AU	38.92 %	0.04 Rf	22.5 AU	6696.8 AU	23.90 %
2	0.05 Rf	23.0 AU	0.06 Rf	32.8 AU	2.52 %	0.10 Rf	12.0 AU	893.7 AU	3.19 %
3	0.17 Rf	7.6 AU	0.20 Rf	31.6 AU	2.42 %	0.21 Rf	14.6 AU	681.8 AU	2.43 %
4	0.22 Rf	14.7 AU	0.24 Rf	71.1 AU	5.46 %	0.27 Rf	7.1 AU	1427.9 AU	5.10 %
5	0.33 Rf	1.4 AU	0.37 Rf	14.1 AU	1.08 %	0.40 Rf	2.4 AU	357.6 AU	1.28 %
6	0.43 Rf	2.8 AU	0.46 Rf	13.4 AU	1.03 %	0.48 Rf	7.6 AU	269.8 AU	0.96 %
7	0.48 Rf	7.7 AU	0.52 Rf	128.1 AU	9.84 %	0.55 Rf	17.5 AU	2947.9 AU	10.52 %
8	0.55 Rf	17.6 AU	0.60 Rf	328.6 AU	25.24 %	0.67 Rf	13.6 AU	9685.0 AU	34.56 %
9	0.67 Rf	13.6 AU	0.73 Rf	175.5 AU	13.48 %	0.77 Rf	3.9 AU	5059.5 AU	18.06 %

3.5. HPTLC studies of Dendrophthoe falcata (L) extract.

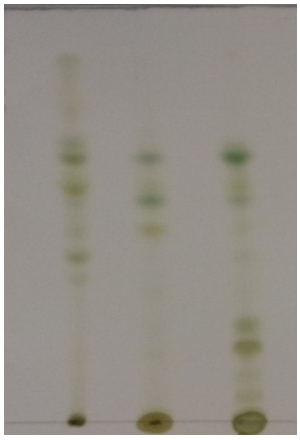

Fig. 7. TLC plate showing different constituents of Dichloromethane: Methanol, Methanolic and Ethanolic extract of *Dendrophthoe falcata* (leaf) (Track 1-3)

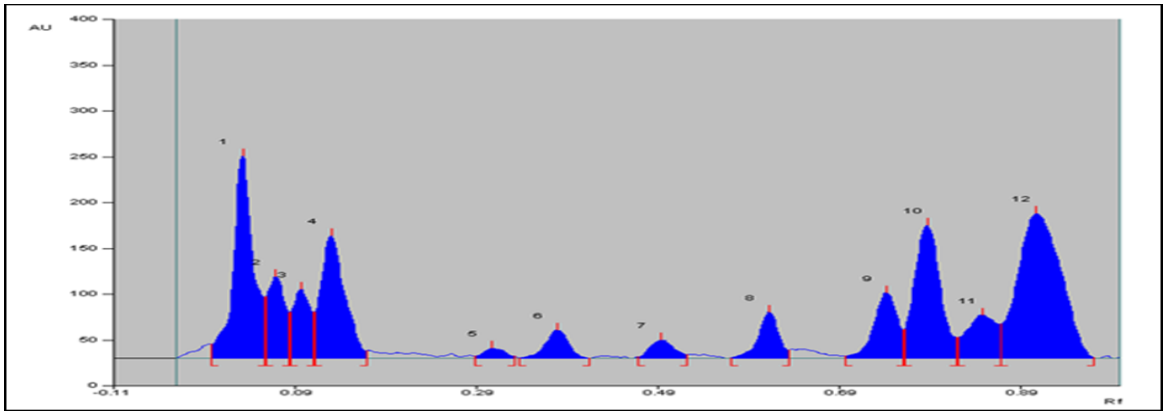

Fig. 8. Chromatogram of Dichloromethane: Methanol extract of *Dendrophthoe falcata* (leaf) measured at 366nm.

Table 8. Rf values for Dichloromethane: Methanol extract of *Dendrophthoe falcata* (leaf).

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	-0.00 Rf	15.7 AU	0.03 Rf	221.0 AU	20.95 %	0.06 Rf	56.8 AU	3962.6 AU	16.37 %
2	0.06 Rf	67.3 AU	0.07 Rf	89.3 AU	8.46 %	0.08 Rf	49.7 AU	1443.8 AU	5.96 %
3	0.08 Rf	50.6 AU	0.10 Rf	75.6 AU	7.17 %	0.11 Rf	50.3 AU	1189.4 AU	4.91 %
4	0.11 Rf	50.8 AU	0.13 Rf	134.2 AU	12.72 %	0.17 Rf	8.2 AU	2913.5 AU	12.03 %
5	0.29 Rf	2.2 AU	0.31 Rf	11.2 AU	1.06 %	0.33 Rf	2.1 AU	217.0 AU	0.90 %
6	0.34 Rf	0.3 AU	0.38 Rf	30.6 AU	2.90 %	0.41 Rf	0.0 AU	642.1 AU	2.65 %
7	0.47 Rf	1.6 AU	0.49 Rf	20.0 AU	1.89 %	0.52 Rf	4.1 AU	441.3 AU	1.82 %
8	0.57 Rf	0.4 AU	0.61 Rf	50.7 AU	4.80 %	0.63 Rf	8.9 AU	913.5 AU	3.77 %
9	0.70 Rf	2.2 AU	0.74 Rf	71.7 AU	6.79 %	0.76 Rf	30.9 AU	1534.5 AU	6.34 %
10	0.76 Rf	31.5 AU	0.79 Rf	145.3 AU	13.77 %	0.82 Rf	22.8 AU	3432.5 AU	14.18 %
11	0.82 Rf	22.8 AU	0.85 Rf	47.5 AU	4.50 %	0.87 Rf	37.0 AU	1288.1 AU	5.32 %
12	0.87 Rf	37.1 AU	0.91 Rf	158.2 AU	14.99 %	0.97 Rf	0.5 AU	6232.6 AU	25.74 %

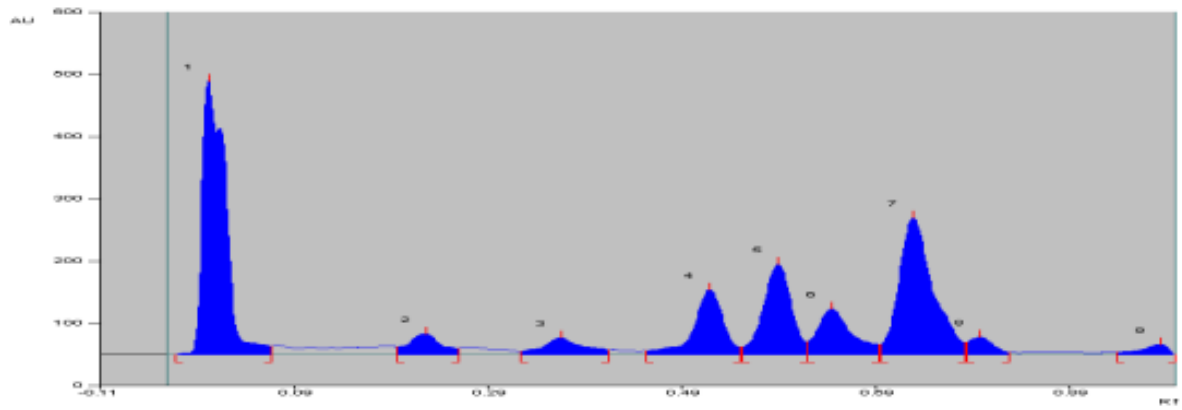

Fig. 9. Chromatogram of Dichloromethane: Methanol extract of *Dendrophthoe falcata* (leaf) measured at 560 nm.

Table 9. Rf values for Dichloromethane: Methanol extract of *Dendrophthoe falcata* (leaf).

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	-0.03 Rf	0.1 AU	0.00 Rf	439.4 AU	40.47 %	0.07 Rf	13.7 AU	8508.8 AU	32.97 %
2	0.20 Rf	12.6 AU	0.23 Rf	33.5 AU	3.09 %	0.26 Rf	9.2 AU	922.1 AU	3.57 %
3	0.32 Rf	5.2 AU	0.37 Rf	27.1 AU	2.49 %	0.41 Rf	7.7 AU	942.6 AU	3.65 %
4	0.45 Rf	5.8 AU	0.52 Rf	104.3 AU	9.60 %	0.55 Rf	11.8 AU	2593.4 AU	10.05 %
5	0.55 Rf	11.9 AU	0.59 Rf	144.8 AU	13.34 %	0.62 Rf	20.0 AU	3441.2 AU	13.33 %
6	0.62 Rf	20.3 AU	0.64 Rf	73.7 AU	6.78 %	0.69 Rf	15.8 AU	2117.4 AU	8.20 %
7	0.69 Rf	16.2 AU	0.73 Rf	219.2 AU	20.19 %	0.78 Rf	19.0 AU	6324.0 AU	24.50 %
8	0.78 Rf	19.5 AU	0.80 Rf	28.1 AU	2.59 %	0.83 Rf	3.7 AU	584.2 AU	2.26 %
9	0.94 Rf	3.1 AU	0.98 Rf	15.8 AU	1.46 %	1.00 Rf	0.1 AU	373.9 AU	1.45 %

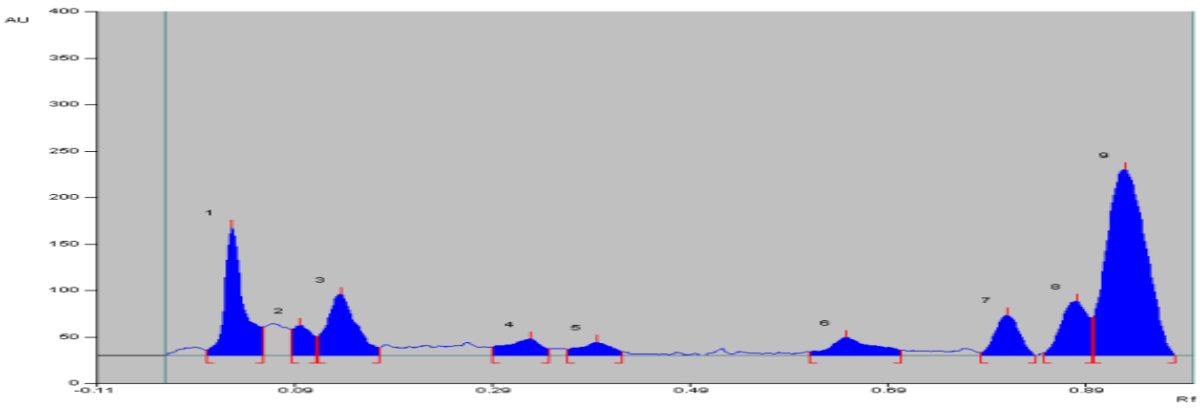

Fig.10. Chromatogram of Methanolic extract of *Dendrophthoe falcata* (leaf) measured at 366nm.

Table 10. Rf values for Methanolic extract of *Dendrophthoe falcata* (leaf).

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	5.7 AU	0.03 Rf	138.4 AU	23.42 %	0.06 Rf	30.6 AU	2144.0 AU	14.72 %
2	0.09 Rf	28.5 AU	0.09 Rf	32.4 AU	5.48 %	0.11 Rf	20.7 AU	528.6 AU	3.63 %
3	0.11 Rf	20.7 AU	0.14 Rf	66.0 AU	11.18 %	0.17 Rf	8.5 AU	1585.9 AU	10.89 %
4	0.29 Rf	9.3 AU	0.33 Rf	17.7 AU	2.99 %	0.35 Rf	7.4 AU	537.5 AU	3.69 %
5	0.37 Rf	6.8 AU	0.39 Rf	14.6 AU	2.46 %	0.42 Rf	3.7 AU	391.1 AU	2.69 %
6	0.61 Rf	3.9 AU	0.65 Rf	19.5 AU	3.30 %	0.70 Rf	5.8 AU	706.9 AU	4.85 %
7	0.78 Rf	3.2 AU	0.81 Rf	43.6 AU	7.38 %	0.84 Rf	0.1 AU	865.8 AU	5.94 %
8	0.85 Rf	2.9 AU	0.88 Rf	58.5 AU	9.90 %	0.90 Rf	40.7 AU	1307.2 AU	8.97 %
9	0.90 Rf	41.7 AU	0.93 Rf	200.1 AU	33.87 %	0.98 Rf	0.4 AU	6500.3 AU	44.62 %

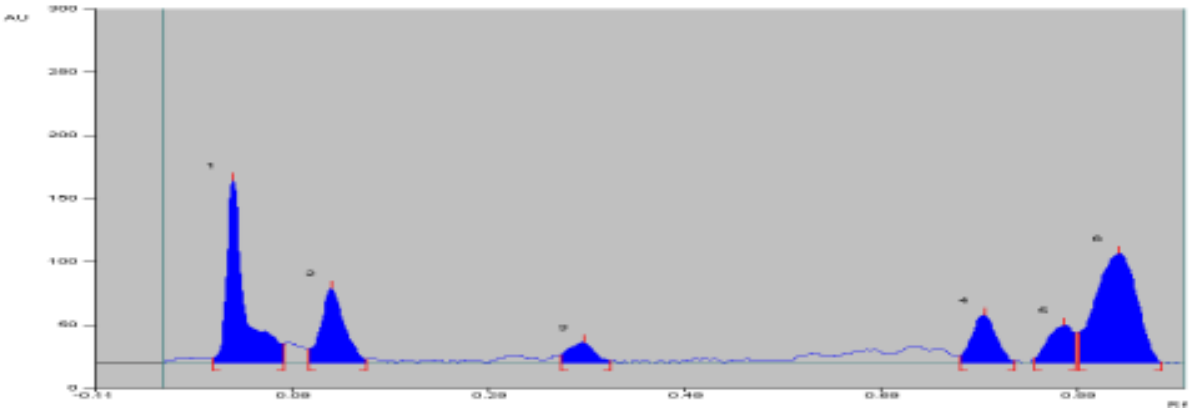

Fig. 11. Chromatogram of Ethanol water extract of *Dendrophthoe falcata* (leaf) measured at 366nm.

Table 11. Rf values for Ethanol water extract of *Dendrophthoe falcata* (leaf).

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	3.5 AU	0.03 Rf	144.6 AU	38.60 %	0.08 Rf	15.2 AU	2177.0 AU	26.84 %
2	0.11 Rf	11.0 AU	0.13 Rf	59.2 AU	15.79 %	0.17 Rf	2.9 AU	1151.8 AU	14.20 %
3	0.36 Rf	6.1 AU	0.39 Rf	16.5 AU	4.40 %	0.41 Rf	2.0 AU	361.4 AU	4.46 %
4	0.77 Rf	5.6 AU	0.79 Rf	37.9 AU	10.12 %	0.82 Rf	1.6 AU	776.1 AU	9.57 %
5	0.85 Rf	3.7 AU	0.88 Rf	29.7 AU	7.93 %	0.89 Rf	24.0 AU	639.8 AU	7.89 %
6	0.89 Rf	23.5 AU	0.93 Rf	86.7 AU	23.15 %	0.97 Rf	0.8 AU	3005.5 AU	37.05 %

The study revealed that *Tridax procumbens (L)* and *Dendrophthoe falcata (L)* showed best results in n-hexane: Toluene: ethyl acetate (2: 4: 1) solvent system for all extracts (fig.1,fig. 5). After scanning and visualizing the plates in absorbance mode at both 254nm, 366 nm The

HPTLC images shown that all sample constituents were clearly separated without any tailing and diffuseness.

The results from HPTLC finger print scanned (Fig. 2) at wavelength 366 nm for methanol water extract of *Tridax procumbens* (TPMW) leaf powder revealed the presence of nine polyvalent phytoconstituents (Table 3). The Rf values ranged from 0.03 to 0.85. The chromatogram as shown in Figure 2and out of 9 components, the component no.7 with *R*f values 0.61 showed maximum concentration**.** The bands revealed may be presence flavonoids, tannins steroids, terpenoids and saponins. The results from HPTLC finger print scanned (Fig. 3) at wavelength 366 nm for ethanol water extract of *Tridax procumbens* (TPEW)(leaf) powder showed presence of ten polyvalent phytoconstituents and Rf values ranged from 0.03 to 0.82 (Table 5). Component no. 9 & 10 having Rf values 0.72 and 0.82 showed maximum concentration 21.83%, 12.81%. The results from HPTLC finger print scanned (Fig. 4) at wavelength 254 nm for Dichloromethane: Methanol extract of *Tridax procumbens* (TPDM) leaf powder showed presence of 9 polyvalent Phytoconstituents and Rf values ranged from 0.05 to 0.82 (Table 5). Component no. 7, 8 & 9 having Rf values 0.55, 0.67 and 0.82 showed maximum concentration 16.79%, 11.03 % & 14.03.

The results from HPTLC finger print scanned (Fig. 5) at wavelength 366 nm for Dichloromethane: Methanol extract of *Tridax procumbens* leaf powder showed presence of 13 polyvalent Phytoconstituents and Rf values ranged from 0.03 to 0.83 (Table.6). Component no. 11 & 13 having Rf values 0.73 and 0.83 showed maximum concentration 27.70 %, 23.43 %. The results from HPTLC finger print scanned (Fig. 6) at wavelength 560 nm for Dichloromethane: Methanol extract of *Tridax procumbens* (leaf) powder showed presence of 9 polyvalent Phytoconstituents and Rf values ranged from 0.05 to 0.67 (Table 7). Component no. 7, 8 & 9 having Rf values 0.48, 0.55 and 0.67 showed maximum concentration 10.52%, 34.56 %& 18.06%. The colour bands significantly revealed may be presence flavonoids, tannins, steroids, and terpenoids.

The results from HPTLC finger print scanned (Fig.8) at wavelength 366 nm for Dichloromethane: Methanol extract of *Dendrophthoe falcata* (DFDM) leaf powder showed presence of 12 polyvalent phytoconstituents and Rf values ranged from 0.06 to 0.87 (Table 8). Component no. 4, 10 & 12 having Rf values 0.11, 0.76 and 0.87 showed maximum concentration 12.03%, 14.18% and 25.74.

The results from HPTLC finger print scanned (Fig. 9) at wavelength 560 nm for Dichloromethane: Methanol extract of *Dendrophthoe falcata* leaf powder showed presence of 9 polyvalent phytoconstituents and Rf values ranged from 0.20 to 0.94 (Table 9). Component no. 4,5& 7 having Rf values 0.45,0.55 and 0.69 showed maximum concentration 10.05 %, 13.33 % and 24.50 % The colour bands significantly revealed may be presence flavonoids, tannins and steroids.

The results from HPTLC finger print scanned (Fig. 10) at wavelength 366 nm for Methanolic extract of *Dendrophthoe falcata* (DFMW) leaf powder showed presence of 9 polyvalent phytoconstituents and Rf values ranged from 0.09 to 0.90 (Table 10). Component no. 3 & 9 having Rf values 0.11, and 0.90 showed maximum concentration 10.89 % and 44.62 % The colour bands significantly revealed may be presence flavonoids, Saponin, tannins and steroids.

The results from HPTLC finger print scanned (Fig.11) at wavelength 366 nm for Ethanol water extract of *Dendrophthoe falcata* (DFEW) leaf powder showed presence of 6 polyvalent phytoconstituents and Rf values ranged from 0.01 to 0.89 (Table 11). Component no. 2 & 6 having Rf values 0.11, and 0.89 showed maximum concentration 14.20 % and 37.05 %.The colour bands significantly revealed may be presence flavonoids, Saponin and steroids. The chromatogram shows presence of multiple peaks which indicate diverse composition of extract.
Phytochemicals are chemical compounds biosynthesized during the various metabolic processes. Various phytochemicals like alkaloid, carbohydrate, glycoside, steroid, protein, tannin, terpenoids, flavonoids and phenolic compounds possess a variety of pharmacological activities. These chemicals are often called secondary metabolites. These phytochemicals shows antimicrobial, antitubercular, anticancer activity etc. Phytochemical analysis revealed the presence of alkaloid, carbohydrate, glycoside, steroid, protein, tannin, terpenoids, flavonoids and phenolic compounds in leaf extract of *Dendrophthoe Falcate* (L) *and Tridax procumbens* (L). HPTLC fingerprint studies confirmed the results of phytochemical screening by the presence of various coloured bands at different wavelengths with specific solvent systems, symbolizing the presence of particular phytocompounds.

4. CONCLUSION

Herbal extracts composed of many phytochemicals It can be concluded that HPTLC fingerprint analysis of leaf extract of Dendrophthoe *Falcate* (L) *and Tridax procumbens* (L). can be used as a diagnostic tool for the identification of the plant and it was appropriate method for standardization of the extract. The present study shows presence of phytochemicals as secondary metabolites which may be responsible for pharmacological activity. Evaluation of all physical constant shown satisfactory results. Methanolic extractive value was found maximum. HPTLC chromatogram of Dichloromethane: Methanol extracts Dendrophthoe *Falcate* (L) *and Tridax procumbens* (L) results showed presence of many phytochemicals in. From the HPTLC studies of shows extracts contain not a single compound but a mixture of compounds and so it is revels that the pharmacological activity in extracts may be due to the cumulative effect of all the compounds in composite and it's detected by HPTLC.

5. ACKNOWLEDGEMENT

The authors are thankful to the head of the Institutions for providing the necessary facilities, thankful to Botanical Survey of India (BSI) Pune, INDIA for identification & authentication. The HPTLC instrumentation facility was provided by Poona College of pharmacy, pune (MH) gratefully acknowledged.

6. CONFLICT OF INTEREST

There is no conflict of interest.

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