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HPTLC fingerprint profile of leaf extracts of *Euphorbia hirta* L.

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Abstract
HPTLC fingerprint method was used for determination of phytoconstituents present in leaf extract of *Euphorbia hirta* plant belongs to Euphorbiaceae family, all plant parts such as root, stem leaves, used as antioxidant, diarrhoea, dysentery, antitumor antibacterial. Attempts have been made to study the complete profile of leaf extract by using High performance thin layer chromatography. The densitometric analysis shows fingerprinting, RF value, peaks of densitogram and chemical variation, this technique is useful for drug identification, adulteration and also acts as biomarker in plant industry.

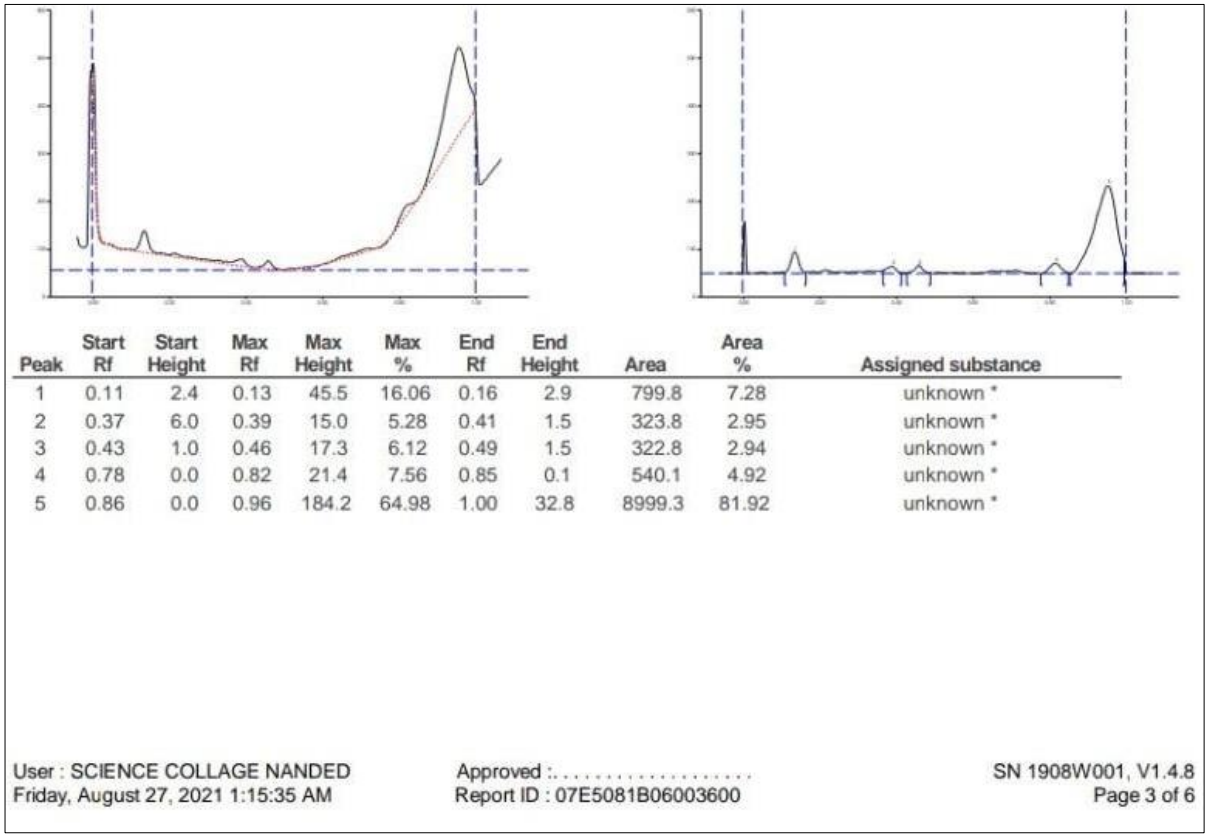
Keywords: euphorbiaceae, *Euphorbia hirta* L. HPTLC

Introduction
The peoples are aware about using chemical and synthetic medicines cosmetics and give more preference to use of herbal products. India recognize more than 2500 plants species which have medicinal value, However, large flora is waiting for their medicinal properties [1]. The use of medicinal plants as a source of medicine and human substances has been in vogue since antiquity India has rich heritage of use of plants as medicines and near about 805 medicines obtained from plants.

Raigad district of Kankan region is very well known for its huge Biodiversity of flora and fauna. The main range of sahyadri, spurs and valleys form important botanical pockets of high biodiversity. The north –east and east stretches of sahyadri supports luxuriant growth of vegetation in Maharashtra state. The area has forest situated on its surrounding mountains. Sahyadri hills has a huge reservoir of enormous natural resources including vegetation wealth and traditional knowledge of medicinal plants The last two decades Pharmaceutical industry has made massive investment in research throughout the world to discover new drugs. Plants have effectively passed the taste of commercial screening [2].

Morphology of plant
Euphorbia hirta L. *or* Birhat Dhugdhika is an annual herb with milky latex, medicinally important plant from the family Euphorbiaceae with 1600 species [3]. stem hispid with yellowish crisped hairs, leaves unequal, sided and cordate at base cyathia many, crowded in small axillary, sub sessile cyme.fruits globose. hispid. Seeds ovoid, trigonous transversely rugose reddish brown. It is also called asthma herb and pill bearing spurge [4, 5]. The stem sap is used in treatment of eyelid style and leaf poultices is used in swelling and boils [6]. Latex is applied on lower eyelids, like *surma* to cure sores, Root exudates exhibits nematicidal activity against juveniles of Meloidogyne incognita and also used for snake bite. The whole plant shows sedative, antispasmodic antifertility antifungal and antimalarial properties, antidiarrheal activity, antibacterial, anthelmintic, antifungal [7]. The powdered *E. hirta* showed a galactogenic activity in guinea pigs before puberty by increasing the development of the mammary glands and induction of secretion [8].

Further research is going on to find out more activities in constituents of *E. hirta*. There are many other traditional uses of *E. hirta* in Ayurveda which serves as the basis for further studies. In pharmacological study of *Euphorbia hirta* L may contain s afzelin, quercitrin, myricitrin, rutin, gallic acid, quercitin, euphorbin-A and ephorbin-B, euphorbin-C, euphorbin-D, β-amyrin, 24-methylenecycloartenol, β-sitosterol, heptacosane, n-nonacosane [10], shikmic acid, tinyatoxin, choline, camphol, and quercitol derivatives containing rhamnose, and chtolphenolic acid [11].



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0.11	2.4	0.13	45.5	16.06	0.16	2.9	799.8	7.28	unknown *
2	0.37	6.0	0.39	15.0	5.28	0.41	1.5	323.8	2.95	unknown *
3	0.43	1.0	0.46	17.3	6.12	0.49	1.5	322.8	2.94	unknown *
4	0.78	0.0	0.82	21.4	7.56	0.85	0.1	540.1	4.92	unknown *
5	0.86	0.0	0.96	184.2	64.98	1.00	32.8	8999.3	81.92	unknown *

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Fig 1: Chromatogram of *Euphorbia hirta* L leaf extract.

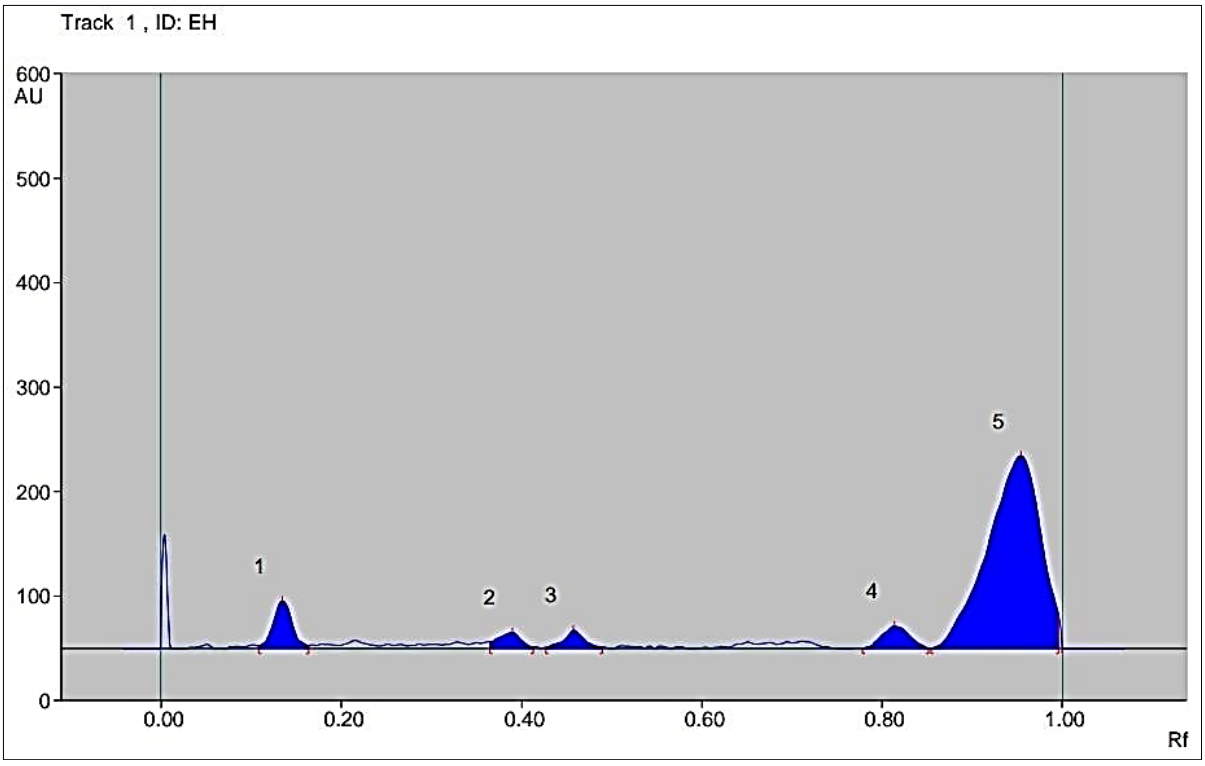

Fig 2: HPTLC Chromatogram of *Euphorbia hirta* L. leaf extract

Table 1: RF Value of leaf extract of *Euphorbia hirta* L Leaf at UV254nm.

Peak	Start Position	Start Height	Max position	Max height	Max %	End Position	End Height	Area	Area %
1	0.11Rf	2.24AU	0.13Rf	45.5AU	16.06%	0.16Rf	2.9AU	799.8AU	7.28%
2	0.37 Rf	6.0 AU	0.39Rf	15.0AU	5.28%	0.41Rf	1.5AU	323.8AU	2.95%
3	0.43 Rf	1.0AU	0.46Rf	17.3 AU	6.12%	0.49Rf	1.5AU	322.8AU	2.94%
4	0.78 Rf	0.0AU	0.82Rf	21.4AU	7.56%	0.85Rf	0.1AU	540.1AU	4.92%
5	0.86 Rf	0.0AU	0.96Rf	184.2AU	64.98%	1.00Rf	32.8AU	8999.3AU	81.92%

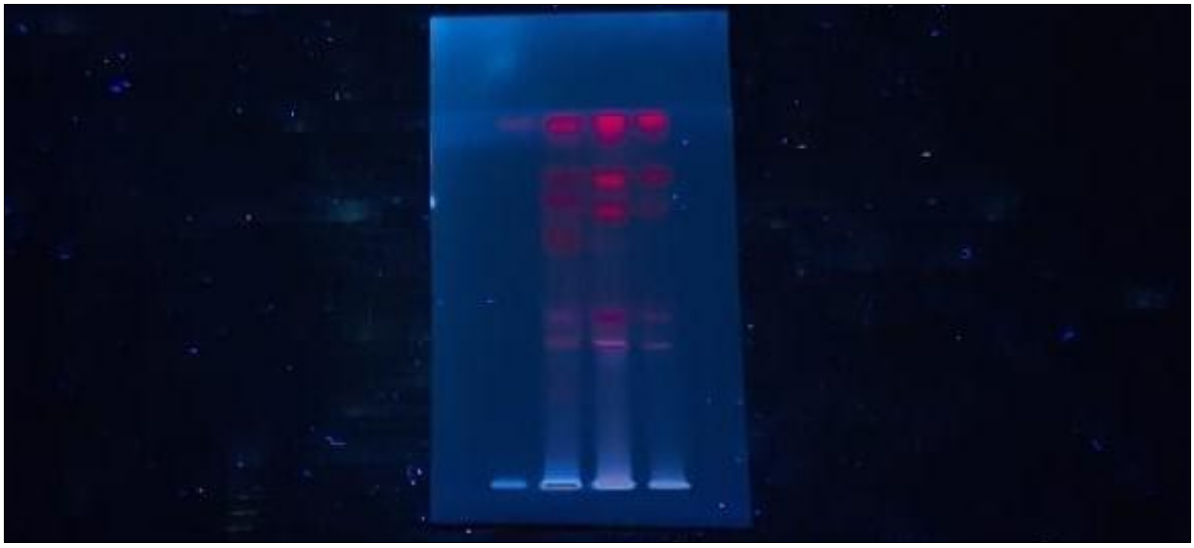

Fig 3: HPTLC profile of *Euphorbia hirta* L plant ethanol extract at 366 nm.

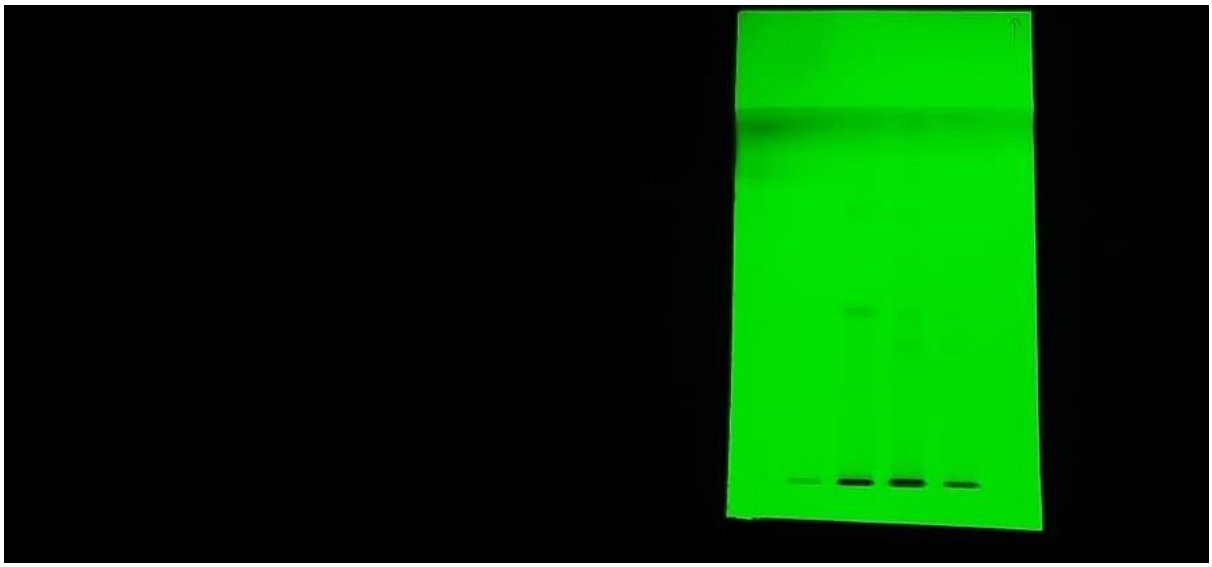

Fig 4: HPTLC profile of *Euphorbia hirta* L plant ethanol extract at 254 nm.

Material and Methods

Preliminary phytochemical analysis of leaf extracts of *Euphorbia hirta* L is done as per method described by Wagner [12], Harborne [13], and Eike Reich [14], HPTLC profiling was done by using CAMAG HPTLC System with WIN CATS software.

Collection of Plant material: The whole plant was collected from Sundarrao more arts, commerce and Science college Poladpur College campus area in the month of August 2020 and correctly identified with the flora of Kolhapur District (Dr S R Yadav) a herbarium was prepared and deposited in the Botany laboratory. The leaves were washed gently with running tap water to remove surface dust, pollutants and dried under the shade. The dried plant material was made of powder using a mixture grinder.

Extraction of Plant Material

About 10 gm. powder of *Euphorbia hirta* L was extracted separately using 70% ethanol in a Soxhlet Extractor (Borosil) for about six hours. After extraction the extracts were evaporated to dryness. The dried extracts were dissolved in 5 ml ethanol and filtered using Whatmann filter. The filtered extracts were later used for further phytochemical and HPTLC analysis [15] Wincats Planar Chromatography Manager. The sample of leaf extract of leaf extracts of *Euphorbia hirta* L were filtered through the whatman filter paper No.1 and injected analysis. The Following peaks were obtained in fig No 2.The leaf extract of *Euphorbia hirta* L showed peaks which may be. Afzelin, quercitrin, myricitrin, quercitin, euphorbin. etc.

Result and Discussion

The HPTLC analysis obtained high resolution and shows different peaks leaf extract of *Euphorbia hirta* L was runs along with the standard and perceived to validate the presence of phytochemical compounds from chromatogram after derivatization. The result from HPTLC fingerprint scanned at wavelength 366 nm for *Euphorbia hirta* L shows polyvalent phytoconstituents and corresponding ascending order of Rf value are from 0.11to 0.86 in which highest concentration of the phytoconstituents was found to be 64.98%. This is recorded in Table No.1 ethanol is used as a solvent Rf value and different wavelength were obtained in picture plate at UV254 nm. The graphical representation shows different peaks of polyvalent phytoconstituents. The Rf value starts from 0.11 to 0.86 in which highest concentration of phytoconstituents were found and maximum percentage starts

from 16.06 to 64, 98% and maximum height from 45.5AU to 184.2AU control. The peak retention in ethanol extracts and is found with Rf start with 0.11,0.37,0.43,0.78,0.86 and end with 0.13, 0.39, 0.46, 0.82, 0.96 and maximum percentage is 16.06%, 5.28%6.12%7.56%, 64.98% in Table no. 1. These studies have shown that it is more versatile than ordinary TLC methods as the spots are well resolved. The HPTLC method is simple, rapid, accurate, reproducible, selective and economic can be for quality and quantitative determination of plant material [16].

Conclusion

The study of HPTLC fingerprints profile of *Euphorbia hirta* L useful to determine the quality of crude drug and also useful for separation of secondary metabolites alkaloids, flavonoids glycosides, amino acids, tannins, saponins, etc.

Acknowledgement

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