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HPTLC FINGERPRINTING OF *CONVOLVULUS ARVENSIS* L.

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ABTRACT

Convolvulus arvensis L. is a long lived deep rooted weed belongs to the family convolvulaceae. It is commonly known as European bindweed**,** bindweed, creeping jenny, morning glory**,** and devil's guts. Most people assume weeds have no therapeutic value. However, the difference between weeds and herbs may merely be our understanding of them. Weeds are unwanted plants and are considered harmful, as they compete with crops for light, moisture and nutrients, and harbour insects and diseases harmful to crops. For farmers and agriculture specialists, weeds are unwanted plants, but for herbalists, all weeds are useful plants. Traditionally its was commonly used as ant-helmintic in animals, anticancer, Cytotoxic, laxative and stimulant action .The present study deals with HPTLC fingerprinting of methanol extract of *C. arvensis* carried out by using various types of solvent system for separation of as many as phytochemicals. Results revealed that the presense of several constituents in the extracts. The number of constituent in the extract and their retention factor (Rf) are summarized in Table 1-11 and chromatographic profile given in figure 1-5. HPTLC results indicate the number of constituents and further facilitate their quantitative estimation and qualitative separation of pharmacologically active chemical compounds. More phytochemical research work is required for isolation, purification and characterization of bioactive compounds.

KEYWORDS: Traditional medicine, herbal drugs, weeds , HPTLC fingerprinting , Rf value.

INTROUCTION

Herbal drugs play an important role in health care programs especially in developing countries. Ancient Indian literature incorporates a remarkably broad definition of medicinal

plants and considers all plant or parts of plants to be potential sources of medicinal substances. However, a key obstacle, which has hindered the acceptance of the alternative medicines in the developed countries, is the lack of documentation and stringent quality control. There is a need for documentation[18] of research work carried out on traditional medicines.[6] With this backdrop, it becomes extremely important to make an effort towards standardization of the plant material to be used as medicine. The process of standardization can be achieved by stepwise pharmacognostic studies, include morphological, anatomical study and biochemical characterization by qualitatively as well as quantitatively. These studies help in identification and authentication of the plant material. Correct identification and quality assurance of the starting materials is an essential prerequisite to ensure reproducible quality of herbal medicine which will contribute to its safety and efficacy.[24]

Convolvulus is a genus of about 250 species of flowering plants they are annual or perennial herbaceous vines, bines and (a few species of) woody shrubs, growing to 0.3-3 m tall. The leaves are spirally arranged, and the flowers trumpet-shaped, mostly white or pink, but blue, violet, purple or yellow in some species. Many of the species are problematic weeds, which can swamp other more valuable plants by climbing over them, but some are also deliberately grown for their attractive flowers. *Convolvulus* species are used as food plants by the larvae of some Lepidoptera species.[4] Previous preliminary studies have revealed that different members of the family of Convolvulaceae[11] possess Cytotoxic effects against a number of tumor cells.[3] *C.arvensis* is one of its species.[15] *C.arvensis* is a long lived deep rooted weed belongs to the family convolvulaceae. It is commonly known as European bindweed.[17]

bindweed, creeping jenny [23] , morning glory [2], and devil's guts. It has at least 84 common names.[11,19,20] Most people assume weeds have no therapeutic value. However, the difference between weeds and herbs may merely be our understanding of them. Weeds are unwanted plants and are considered harmful, as they compete with crops for light, moisture and nutrients, and harbour insects and diseases harmful to crops. For farmers and agriculture specialists, weeds are unwanted plants, but for herbalists, all weeds are useful plants. Such is the case of *convolvulus arvensis*; new research is showing it has great promise as a useful, safe and nontoxic chemotherapeutic agent.[14] It is which spreads by rhizome and seed . Field bindweed is found in a wide range of habitats: orchards, vineyards, roadsides, ditch banks, cropland [7], stream banks, and lakeshores[1,5] , suggests that trees and shading help control the weed, and habitats that are most like agricultural lands (little competition, repeated

disturbance and high light) are ideal for growth of field bindweed. It is a weakstemmed, prostrate plant that can twine and may form dense tangled mats.[9] Field bindweed.[23] Traditionally its was commonly used as ant-helmintic in animals. Lots of research has been done on its Anticancer, Cytotoxic, laxative and stimulant action All aerial parts of *C. arvensis* has been ethno medicinally used as a therapeutic agent for a variety of diseases.

MATERIAL AND METHODS

Material collection and sample processing

The plant material was collected from the local area and identified taxonomically with the help of standard floras.[8,12,13,16] The voucher specimens were deposited in the Department of Botany, Phulsing Naik Mahavidhyalaya, Pusad (MS). A small scale extraction was carried out view of preliminary analysis. The dried plants materials (1-5g) was extracted with methanol at room temperature the methanol was decanted after 24 hours and extraction repeated three times. The pooled extracts were filtered and then concentrated under vaccum using rotoryevaporator at 40 ^{o}C.

HPTLC fingerprinting

HPTLC study of methanol extract was carried out by the method of.[10,21] A number of solvent were tried individually as well as in combination for separation of different components of extract, but the satisfactory resolution was obtained in the solvent system chloroform: methanol (9:1 v/v) . HPTLC plates silica gel 60 F 254 (10cm ×10 cm) are used having calibration mode single level,statistics mode CV,evaluation mode peak area and application parameters are Spray gas - inert gas (100µl), dosage speed 150 nl/s and predosage volume 0.2 µl.

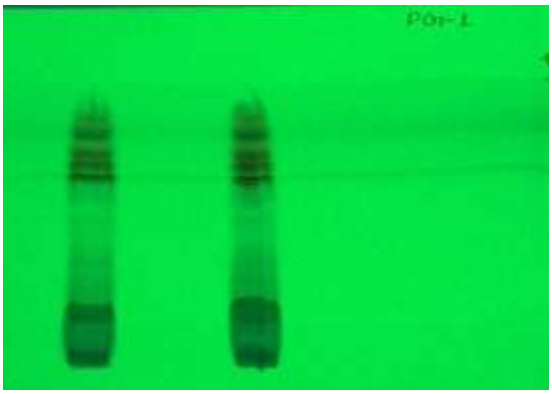
RESULTS AND DISCUSSION

HP T L C fingerprinting of methanol extract of had been carried out by using various types of solvent system for separation of as many as phytochemicals. Results revealed that the presense of several constituents in the extracts. The number of constituent in the extract and their retention factor (Rf) are summarized in Table 1-11 and chromatographic profile given in figure 1-5. HPTLC results indicate the number of constituents and further facilitate their quantitative estimation and qualitative separation of pharmacologically active chemical compounds. More phytochemical research work is required for isolation, purification and characterization of biologically compounds. Since the plant, *Convolvulus arvensis* is useful in

traditional medicine for the treatment of various ailments; it is need of time to standardize the plant for development of quality control parameters.

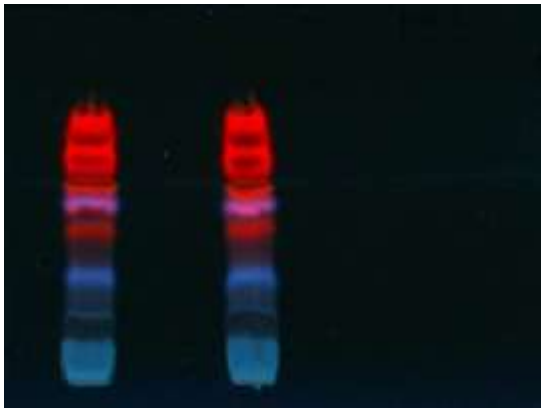
(a)

(b)



(c)

(d)



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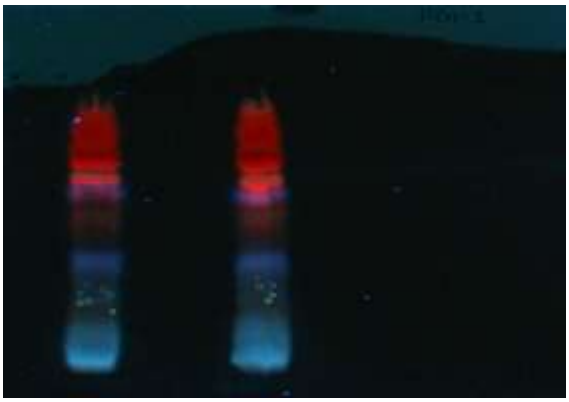


(f)

Figure (1): HPTLC of methanol extract :- (a) at white remission (exposure 54.51 ms), (b) at white remission (exposure 48.76 ms), (c) at 254 nm (exposure 120.98 ms), (d) at 254 nm (exposure 114.54 ms), (e) at 366 nm (exposure 448.27 ms), (f) at 366 nm (exposure 1309.39 ms).

(A) After derive AT 600

Table 1 : Sample I (Appl.volume 20.0 µl and Appl.position 20.0 mm)

	Start	Start	Max	Max	Max	End	End		Area %
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	
1	0.00	1.2	0.04	92.7	4.42	0.12	40.2	4636.3	6.92
2	0.17	42.2	0.21	55.5	2.65	0.23	50.5	1948.4	2.91
3	0.23	50.5	0.24	51.1	2.44	0.28	28.1	1613.2	2.41
4	0.30	28.3	0.31	31.7	1.51	0.34	21.1	766.3	1.14
5	0.35	20.9	0.39	31.1	1.48	0.43	21.5	1658.0	2.47
6	0.46	20.4	0.53	61.7	2.95	0.57	29.1	2877.5	4.29
7	0.57	29.1	0.61	75.1	3.58	0.63	68.8	2380.4	3.55
8	0.63	68.9	0.68	341.9	16.31	0.70	104.3	9537.7	14.23
9	0.70	105.1	0.73	362.7	17.30	0.74	346.4	6667.0	9.94
10	0.74	348.0	0.76	437.7	20.88	0.79	223.2	14543.7	21.69
11	0.79	223.7	0.82	357.5	17.05	0.87	194.7	15729.8	23.46
12	0.87	195.1	0.88	197.7	9.43	0.93	24.4	4685.2	6.99

Table 2 : Sample I (Appl.volume 25.0 µl and Appl.position 50.0 mm)

	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	-0.01	1.3	0.03	85.5	3.15	0.12	43.8	5279.5	5.62
2	0.16	45.9	0.21	64.6	2.38	0.24	54.8	3354.5	3.57
3	0.24	54.4	0.26	55.5	2.04	0.29	34.2	1682.4	1.79
4	0.29	34.3	0.30	45.4	1.67	0.32	37.1	892.9	0.95
5	0.32	37.3	0.33	43.8	1.61	0.34	32.2	648.0	0.69
6	0.34	32.3	0.37	41.3	1.52	0.44	30.4	2476.8	2.64
7	0.46	29.4	0.52	91.3	3.36	0.55	49.3	3776.5	4.02
8	0.55	49.4	0.59	111.3	4.10	0.61	102.6	3876.8	4.13
9	0.61	102.7	0.66	397.2	14.63	0.68	139.8	13148.3	14.00
10	0.69	140.7	0.71	465.5	17.14	0.72	456.2	8625.7	9.18
11	0.72	456.7	0.74	541.3	19.93	0.78	305.5	19510.3	20.77
12	0.78	306.5	0.81	479.2	17.64	0.86	282.1	22121.6	23.55
13	0.86	282.5	0.87	293.9	10.82	0.96	8.7	8538.0	9.09

Table 3 : Blank (Appl.volume 25.0 µl and Appl.position 80.0 mm)

	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	-0.03	0.4	0.11	74.4	49.84	0.12	73.0	4750.5	54.01
2	0.46	48.5	0.47	49.2	32.90	0.55	41.6	2870.8	32.64
3	0.71	25.0	0.72	25.8	17.26	0.78	18.6	1174.5	13.35

(B) Before derive AT 366

Table 4 : Sample I (Appl.volume 20.0 µl and Appl.position 20.0 mm)

	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	0.03	2.5	0.05	13.4	6.18	0.07	9.6	346.3	6.29
2	0.23	10.2	0.27	20.4	9.41	0.28	18.0	547.6	9.94
3	0.64	42.5	0.65	44.1	20.36	0.67	16.5	746.8	13.56
4	0.68	15.4	0.70	44.2	20.38	0.75	2.0	1489.3	27.04
5	0.75	0.2	0.79	30.6	14.13	0.82	0.1	724.3	13.15
6	0.83	0.9	0.85	18.1	8.36	0.88	12.7	475.7	8.64
7	0.88	13.0	0.91	26.8	12.38	0.92	24.5	691.1	12.55
8	0.97	14.9	0.98	19.1	8.80	1.04	3.0	486.1	8.83

Table 5 : Sample I (Appl.volume 25.0 µl and Appl.position 50.0 mm)

	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	0.03	12.7	0.06	19.5	5.45	0.07	18.2	423.8	3.17
2	0.26	19.9	0.28	32.8	9.15	0.29	29.3	568.2	4.25
3	0.32	25.2	0.34	39.0	10.89	0.35	37.6	621.2	4.64
4	0.56	57.7	0.61	69.6	19.44	0.65	30.5	4061.5	30.35
5	0.66	25.2	0.68	67.8	18.94	0.74	0.7	2231.1	16.67
6	0.74	0.1	0.77	48.8	13.64	0.81	2.2	1231.9	9.21
7	0.81	0.6	0.85	32.6	9.11	0.86	24.9	765.3	5.72
8	0.87	23.3	0.90	47.9	13.38	1.01	20.6	3479.5	26.00

(C) Before derive AT 254

Table 6 : Sample I (Appl.volume 20.0 µl and Appl.position 20.0 mm)

	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	-0.02	6.0	0.03	651.3	11.65	0.04	626.7	13671.3	6.22
2	0.04	627.2	0.06	654.0	11.70	0.11	453.3	27452.5	12.49
3	0.11	453.7	0.14	469.3	8.40	0.16	441.9	14272.6	6.50
4	0.16	442.9	0.18	565.3	10.11	0.29	256.0	39910.0	18.16
5	0.29	256.3	0.30	260.7	4.66	0.34	208.8	9081.7	4.13
6	0.34	208.9	0.38	277.3	4.96	0.47	131.1	18421.0	8.38
7	0.50	130.9	0.52	146.4	2.62	0.56	109.5	5931.8	2.70
8	0.56	109.7	0.63	238.5	4.27	0.64	234.4	11020.1	5.02
9	0.64	235.6	0.68	608.8	10.89	0.69	441.2	15648.7	7.12
10	0.69	444.2	0.72	557.2	9.97	0.74	444.9	17307.8	7.88
11	0.74	445.9	0.75	474.2	8.48	0.79	303.3	14775.7	6.72
12	0.79	303.4	0.82	404.3	7.23	0.86	276.0	18021.9	8.20
13	0.86	276.1	0.88	282.4	5.05	1.02	5.1	14207.9	6.47

Table 7 : Sample I (Appl.volume 25.0 µl and Appl.position 50.0 mm)

	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	-0.02	1.0	0.05	667.6	11.45	0.10	455.4	42271.4	16.04
2	0.11	455.6	0.14	510.8	8.76	0.15	507.5	14680.4	5.57
3	0.15	508.3	0.17	580.9	9.96	0.21	545.8	26170.2	9.93
4	0.21	545.8	0.22	546.6	9.37	0.29	292.4	23379.6	8.87
5	0.29	292.6	0.31	314.0	5.38	0.35	281.8	12446.1	4.72
6	0.35	281.9	0.38	310.4	5.32	0.47	165.7	21264.7	8.07
7	0.48	165.8	0.51	200.7	3.44	0.54	167.5	8173.0	3.10
8	0.54	167.8	0.66	658.2	11.28	0.68	515.8	34451.9	13.08
9	0.68	518.4	0.70	624.8	10.71	0.72	533.9	19757.8	7.50
10	0.73	534.1	0.74	556.2	9.54	0.78	377.5	18618.9	7.07
11	0.78	377.9	0.81	491.2	8.42	0.85	349.1	23474.1	8.91
12	0.85	349.4	0.87	371.7	6.37	1.01	1.7	18781.5	7.13

Table 8 : Blank (Appl.volume 25.0 µl and Appl.position 80.0 mm)

	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	-0.02	2.7	0.00	28.6	9.60	0.01	16.8	399.4	2.88
2	0.02	16.0	0.09	41.8	14.03	0.09	40.4	1704.6	12.28
3	0.13	38.9	0.14	40.6	13.63	0.17	18.5	951.2	6.85
4	0.61	21.3	0.68	95.9	32.22	0.71	32.0	3036.5	21.87
5	0.76	33.0	0.83	90.9	30.53	0.93	36.2	7793.3	56.13

(D) Before derive AT 600

Table 9 : Sample I (Appl.volume 20.0 µl and Appl.position 20.0 mm)

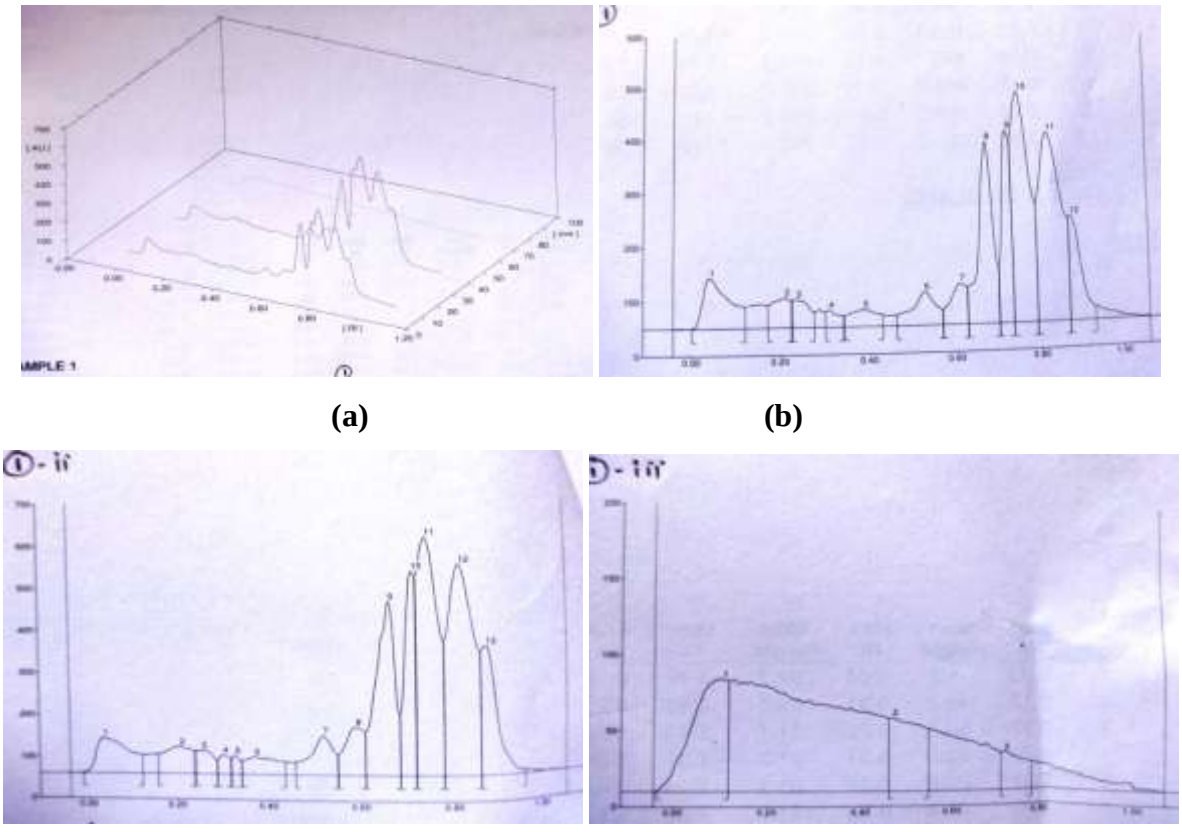
	Start	Start	Max	Max	Max	End	End		Area
Peak	Rf	Height	Rf	Height	%	Rf	Height	Area	%
1	-0.00	1.0	0.03	76.9	4.38	0.09	34.2	2963.1	4.89
2	0.11	33.8	0.13	36.3	2.07	0.17	24.8	1423.8	2.35
3	0.22	23.9	0.23	25.0	1.42	0.28	10.5	759.3	1.25
4	0.35	10.5	0.38	18.6	1.06	0.43	8.0	739.5	1.22
5	0.45	6.9	0.52	59.0	3.36	0.56	14.1	2171.1	3.58
6	0.57	14.1	0.60	62.1	3.53	0.62	49.9	1848.9	3.05
7	0.63	49.9	0.68	248.3	14.13	0.70	90.2	7554.1	12.46
8	0.70	93.3	0.72	405.0	23.05	0.73	373.6	7660.4	12.63
9	0.73	375.3	0.75	455.5	25.93	0.79	224.3	14680.7	24.21
10	0.79	225.9	0.82	370.1	21.07	0.97	1.7	20841.1	34.37

Table 10 : Sample I (Appl.volume 25.0 µl and Appl.position 50.0 mm)

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %
1	-0.02	0.1	0.02	72.5	3.01	0.10	33.9	3542.4	4.36
2	0.23	29.3	0.24	30.1	1.25	0.28	14.5	942.1	1.16
3	0.34	16.2	0.37	26.4	1.10	0.42	19.0	1265.1	1.56
4	0.46	16.8	0.52	87.3	3.63	0.55	36.1	3250.4	4.00
5	0.55	36.4	0.58	92.6	3.85	0.61	81.4	3307.4	4.07
6	0.61	81.9	0.66	308.7	12.83	0.68	125.1	10703.1	13.18
7	0.68	127.6	0.71	494.1	20.54	0.72	471.2	9956.7	12.26
8	0.72	472.4	0.74	541.4	22.51	0.78	301.4	18642.7	22.96
9	0.78	302.4	0.81	469.6	19.52	0.86	274.1	21979.2	27.07
10	0.86	274.7	0.87	282.8	11.76	0.96	3.2	7602.5	9.36

Table 11 : Blank (Appl.volume 25.0 µl and Appl.position 80.0 mm)

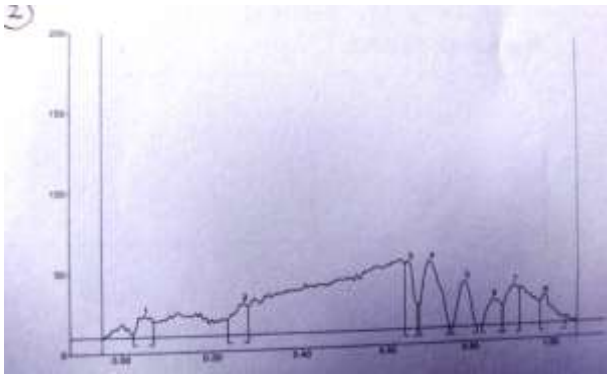
Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %
1	-0.03	1.7	0.09	47.0	100.00	0.10	46.2	2546.9	100.00



(a) **(b)**

(c) **(d)**

Figure 2 : After derive at 600 : (a) All tracks at wavelength Sc4 ,(b) Sample 1 (Volume 20 µl and Appl. Position 20.0 mm) ,(c) Sample 1 (Volume 25 µl and Appl. Position 50.0 mm), (d) Blank (Volume 25 µl and Appl. Position 80.0 mm)



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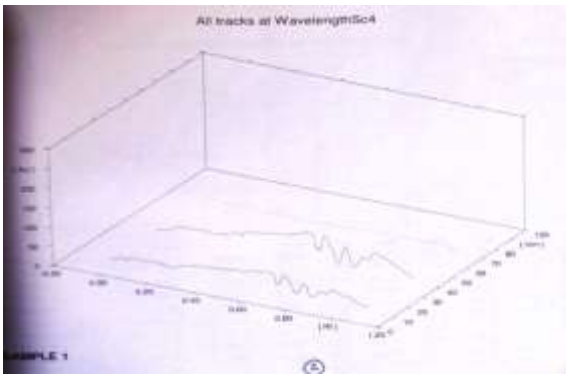

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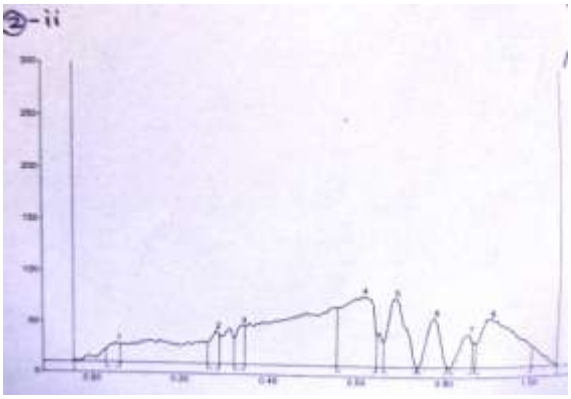

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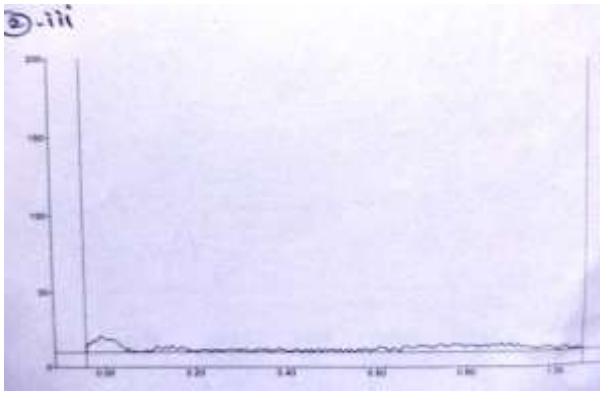
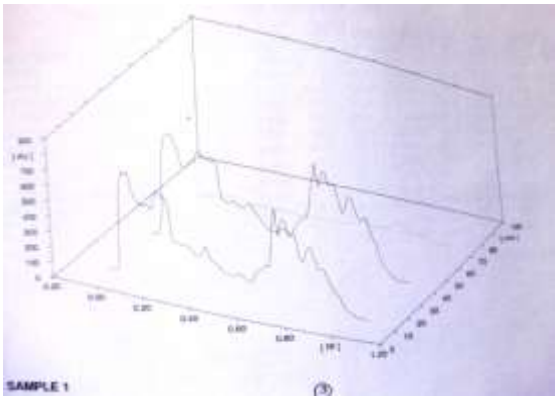


(h)

Figure 3 : Before derive at 366 : (e) All tracks at wavelength Sc4 ,(f) Sample 1 (Volume 20 µl and Appl. Position 20.0 mm) ,(g) Sample 1 (Volume 25 µl and Appl. Position 50.0 mm), (h) Blank (Volume 25 µl and Appl. Position 80.0 mm)



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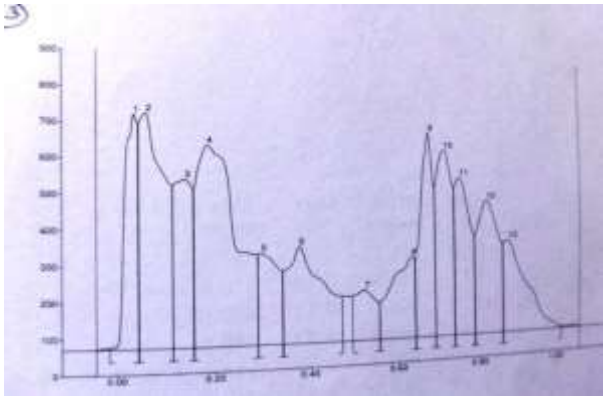
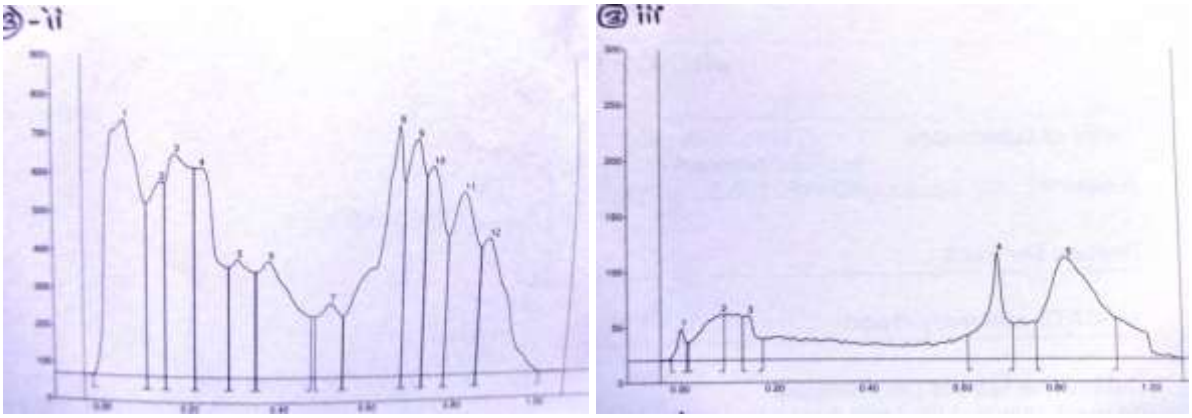
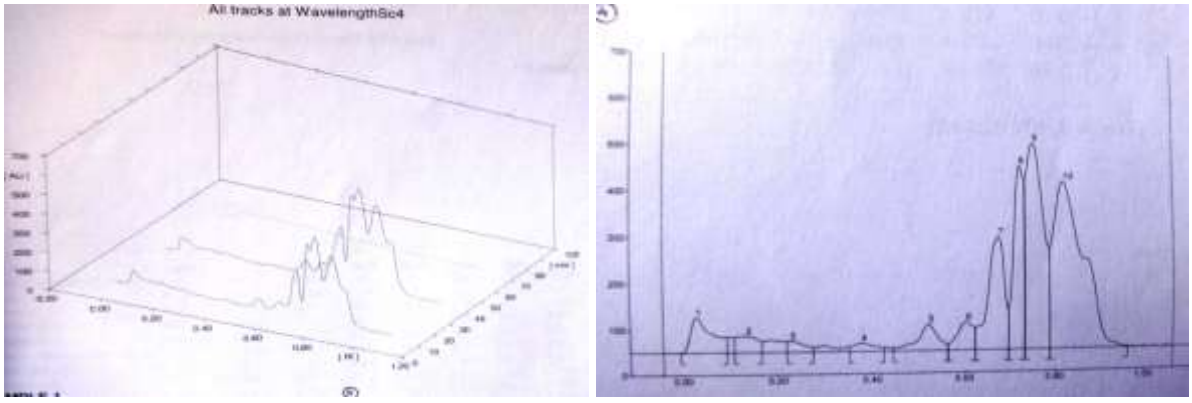


(j)

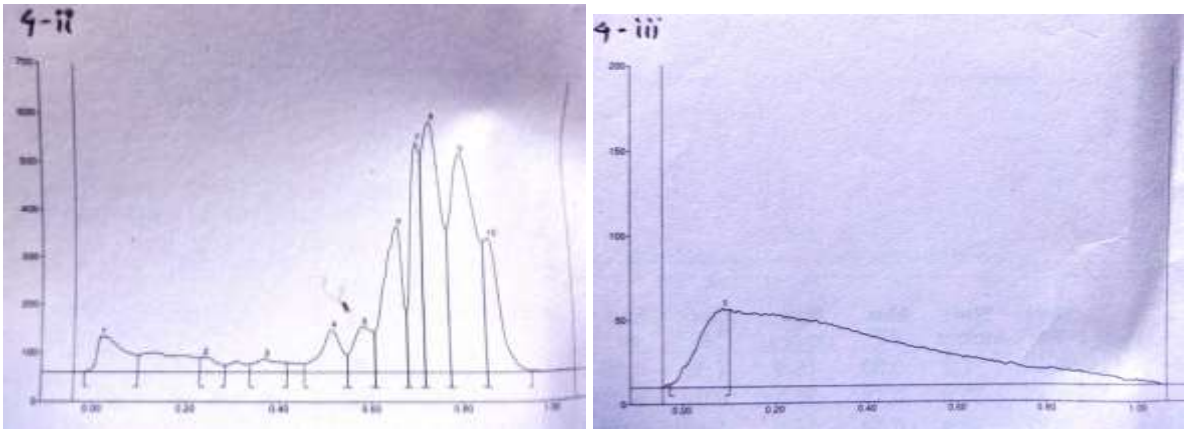


(k) **(l)**

Figure 4 : Before derive at 254 : (i) All tracks at wavelength Sc4 ,(j) Sample 1 (Volume 20 µl and Appl. Position 20.0 mm) ,(k) Sample 1 (Volume 25 µl and Appl. Position 50.0 mm), (l) Blank (Volume 25 µl and Appl. Position 80.0 mm).



(m) **(n)**



(o) **(p)**

Figure 5 : Before derive at 600 : (m) All tracks at wavelength Sc4 ,(n) Sample 1 (Volume 20 µl and Appl. Position 20.0 mm) ,(o) Sample 1 (Volume 25 µl and Appl. Position 50.0 mm), (p) Blank (Volume 25 µl and Appl. Position 80.0 mm)

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