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Heavy Metal Analysis of Various Parts of Ficus mollis (Vahl) by HPTLC

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ABSTRACT: Bark and leaves of Ficus mollis Vahl (Moraceae) are reported as folk medicinal plant parts. The samples of bark and leaves of ficus mollis (vahl) are subjected to HPTLC analysis. Heavy metal contents such as mercury, lead, cadmium, arsenic and minerals like Iron, copper, manganese, zinc, cobalt analysis using atomic absorption spectroscopy and pesticide residues by GCMS were carried out. The study revealed that heavy metals are within the permissible limit. The data evolved in the present work will aid in identifying these drugs in dry form and in standardization of the drug.
Key words:F.Mollis (Vahl), heavy metals, chloroform, ethyl acetate, bark, leaves, HPTLC profile, pesticide residue.

INTRODUCTION

Ficus mollis (Vahl) synonym: ficus tomentosa (Roxb) belongs to family moraceae is large tree with milky juice, distributed in rocky, hilly and dry lands[1,2,3]. The genus is remarkable for the large variation in the habits of its species. All species of Ficus yield latex containing caoutchouc[4]. It is widely used as folk medicine. The decoction of leaves of F.Mollis mixed with leaves of madhuca indica used for ear ache[5]. Alcoholic extracts of leaves and bark of ficus mollis (vahl) shown good anti bacterial activity[6].

ABOUT Ficus mollis

The common name of Ficus mollis is soft fig. It is a big tree found abundantly in south India and Sri Lanka. It is a tree growing up to 12m tall branchiate covered with yellow velvety hairs. Leaves are alternate or nearly opposite. They are elliptic ovate to fiddle shaped, 6-15cm long, 3-9cm wide with a 5cm long stalk.

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Figs occur in leaf axils, either in pairs or in clusters. They are stalkless, round up to 8mm across. Fig wall is same what fleshy, brownish, velvety[6].

MATERIALS AND METHODS

COLLECTION OF PLANT MATERIALS

The leaves and bark of Ficus mollis were collected from the local area of Tirumala hills, Chittoor district, Andhra Pradesh, India during February 2009. The plant was authenticated by Assistant Professor Dr. K. Madhava Chetty, Department of Botany, Herbarium keeper, Sri Venkateswara University, Tirupathi. The plant specimen access number is 870 (Eud-II/2006, 870) and preserved in the herbarium of the department.

PREPARATION OF EXTRACT OF TLC AND HPTLC

The leaves and bark of F. mollies were air dried coarsely powdered and stored in air tight container at 27°C 2gm of drug powder was extracted with chloroform and ethyl acetate and made up to 10 ml in a volumetric flask separately.

TLC / HPTLC PROFILE PROCEDURE [7, 8]

The TLC/HPTLC profile of chloroform extract of F. mollis was performed using aluminium plate precoated with silica gel 60F $_{254}$ (E-Merck) employing CAMAG Linomat IV sample applicator. The chromatogram was developed using toluene : ethyl acetate : formic acid (8:1:0.5) as the developing solvent. The plate was air

dried at room temperature and scanned using CAMAG TLC scanner, CATS V 4.06 software at UV λ 254 nm as the scanning wavelength. The plate was dipped in vanillin sulphuric acid and heated at 105°C till coloured sports appeared.

HEAVY METALS AND MINERAL ANALYSIS [9]

Heavy metal and mineral analysis was carried out by atomic absorption spectroscopy (Perkin Elmer-400), using argon as the carrier gas and flow rate was kept as 1 ml/2 min.

Accurately weighed 500mg of air dried powder was taken in round bottom flask. 5 ml of concentrated nitric acid was added and refluxed for half an hour in a hot plate at 60-80°C. It was then cooled, 5 ml of concentrated nitric acid was added and warmed on water bath. 2 ml of 30% hydrogen peroxide solution was added to the above mixture and warmed till clear solution was obtained. It was then cooled and filtered through Whatmann-42 filter paper, diluted with deionized water and made up to 100 ml in volumetric flask.

ANALYSIS OF PESTICIDE RESIDUE [10]

Pesticide analysis was carried out by gas chromatography-mass spectra (GCMS, Agilent), detector-mass selective detector, column specification – DB5MS, carrier gas-helium, flow rate 1ml/min column length-30m internal diameter -0.25mm column thickness-0.25mm.

PREPARATION OF SAMPLE

Accurately weighed 25gm of coarsely powdered air dried material was taken in a conical flask 65ml of acetonitrile and 35ml of deionized water was added. The mixture was shaken well and allowed it to stand for 2 hrs. with constant shaking. Filtered through Whatmann-41 filter paper and filtrate was collected in a separating funnel 3×65ml of petroleum ether was added (BP 60-80) and shaken vigorously and allowed to settle. The solvent layer was collected in the round bottom flask. Evaporated to dryness on the water bath. Then 1 ml of acetonitrile was added to the residue then injected this sample in the Gas chromatography-mass spectra (GC-MS).

Table 1: Heavy metal and mineral analysis of chloroform extract of leaves and bark of F. mollis

S.No.	Heavy metals	Leaf (PPM)	Bark (PPM)
1	Mercury	0.0692	0.0825
2	Lead	0.1321	0.0602
3	Arsenic	0.0021	0.0045
4	Cadmium	0.0021	0.0022
Mineral contents			
5	Iron	5.2154	2.3675
6	Copper	0.6202	0.2793
7	Manganese	0.4165	0.6254
8	Zinc	0.2152	0.3569
9	Nickel	0.1907	0.1602
10	Cobalt	0.0069	0.0125
11	Chromium	0.2152	0.2532

All values are mean of three determinations

RESULTS AND DISCUSSION

Heavy metal content namely lead, cadmium, mercury and arsenic of the drugs were found to be with in the permissible limits As per WHO and FDA. It was concluded that toxic elements was very low in leaves. Mineral elements such as Iron, copper, manganese, zinc, nickel, cobalt, chromium were observed to be present in considerable amount and may be directly or indirectly helpful in the management of many diseases. The iron, copper and nickel contents were found to be higher in leaf than bark, where as the manganese, zinc, cobalt, chromium contents were found be higher in bark than leaf of ficus mollis (Table No. 1).

Pesticide residue analysis revealed that organo chlorine pesticide residue such as o,p-DDD, p,p'-DDD, o,p-DDE, p,p'-DDE, o,p'-DDT, p,p'-DDT, endo sulfan, α-HCH, β-HCH, γ-HCH, δ-HCH were absent in both leaves and bark of Ficus mollis.

Fig. No. 1 showing heavy metal content in leaves and bark of F. mollis

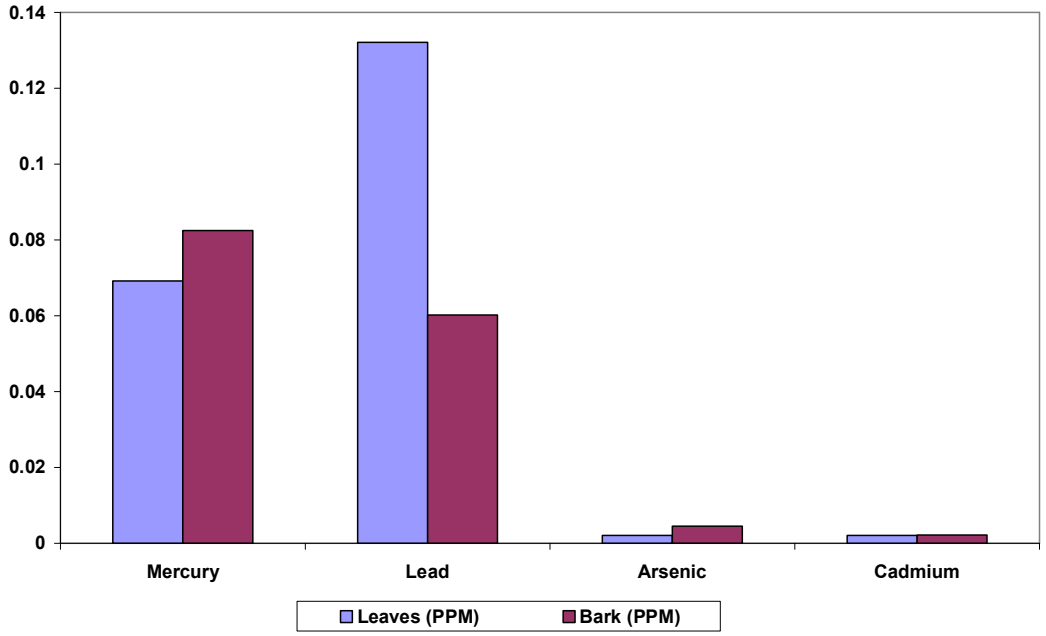
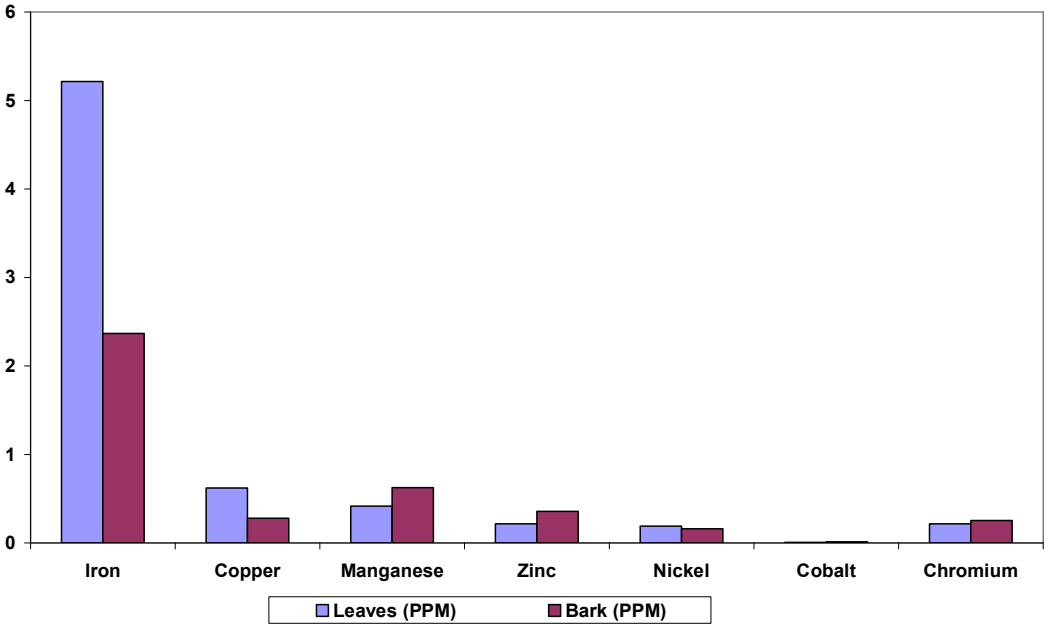
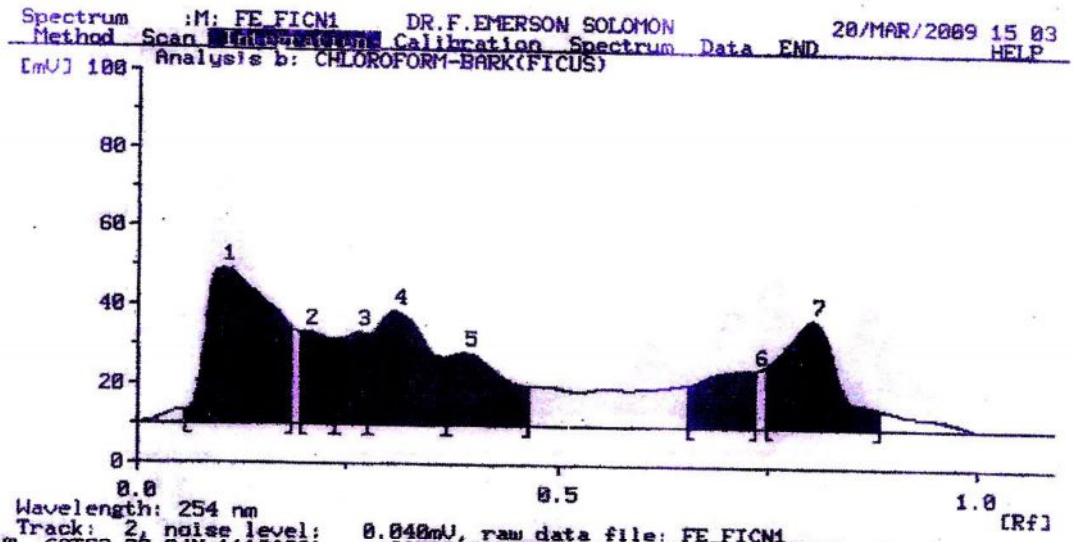


Fig. 2 showing mineral content in leaves and bark of F. mollis





Track 2. Analysis b: CHLOROFORM-BARK(FICUS)

Peak	start		max			end		area	
#	Rf	H	Rf	H	[%]	Rf	H	F	[%]
1	0.05	3.3	0.10	39.0	22.33	0.18	24.2	2646.1	29.27
2	0.19	23.4	0.19	23.4	13.42	0.23	21.5	675.5	7.47
3	0.23	21.5	0.26	23.2	13.31	0.27	22.8	672.7	7.44
4	0.27	22.8	0.30	28.8	16.47	0.36	17.1	1604.2	17.75
5	0.36	17.1	0.38	18.2	10.44	0.46	9.9	1022.2	11.31
6	0.65	10.8	0.73	14.6	8.38	0.73	14.6	787.3	8.71
7	0.75	15.9	0.80	27.3	15.65	0.88	5.1	1631.0	18.04
		Total height =		174.6			total area =	9038.9	

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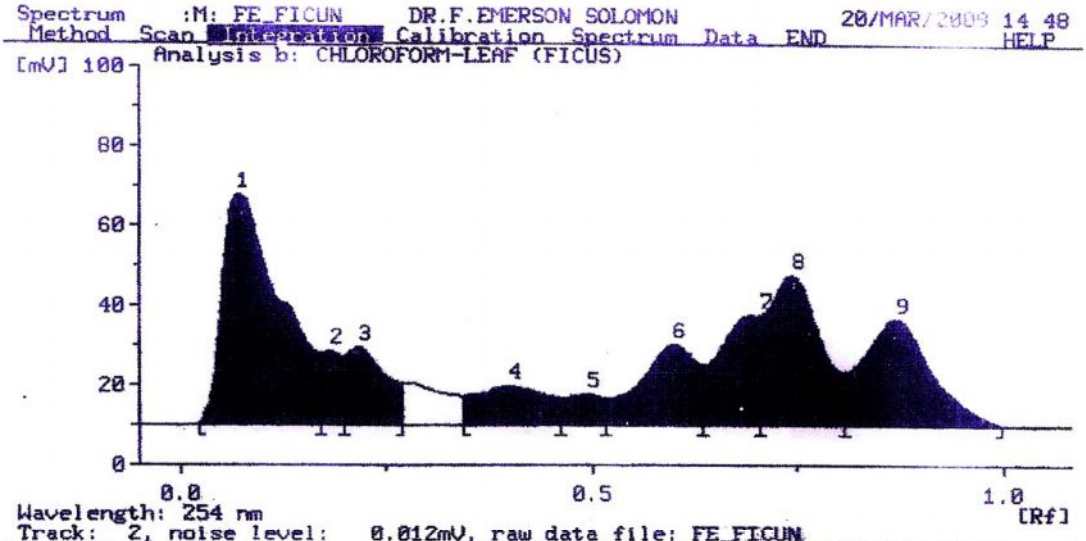

Track 2, Analysis b: CHLOROFORM-LEAF (FICUS)

Peak	start		max			end		area	
#	Rf	H	Rf	H	[%]	Rf	H	F	[%]
1	0.02	0.3	0.07	57.7	25.76	0.17	17.7	2923.0	29.16
2	0.17	17.7	0.18	18.3	8.15	0.19	17.0	302.7	3.02
3	0.19	17.0	0.21	19.4	8.65	0.27	10.4	683.8	6.82
4	0.34	7.2	0.40	9.7	4.32	0.46	6.6	606.7	6.05
5	0.46	6.6	0.49	7.5	3.37	0.52	6.6	241.1	2.40
6	0.52	6.6	0.60	20.0	8.92	0.64	14.9	1000.3	9.98
7	0.64	14.9	0.70	27.4	12.22	0.70	27.4	937.2	9.35
8	0.70	27.4	0.74	37.3	16.69	0.81	13.3	1736.0	17.32
9	0.81	13.3	0.87	26.7	11.92	1.00	0.0	1592.5	15.89
		Total height =		223.8			total area =	10023.2	

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