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QUANTITAVE ESTIMATION OF THE ACTIVE CONSTITUENTS OF *PARKIA BIGLANDULOSA* BY USING HPTLC AND FTIR

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ABSTRACT

Parkia biglandulosa, a native of Malaya. The genus is named after Mungo Park. It is a large handsome, evergreen tree. Flowers are small, ball-shaped, and brown initially turning to white, in pendant flower heads on long stalks. Flowering is in March to April. Propagation is by seeds. The plant is grown in gardens as ornamental tree and also on roadsides as avenue tree. Fruit pulp is reported to be edible.

Pollen mixed with water makes a refreshing drink. The different parts of the plant shows anti ulcer activity, Antibacterial activity, antifungal activity, anti inflammatory activity etc. Saponin's from the seed bran of *Parkia biglandulosa* is used medicinally in India for its astringent and detergent properties and also as a fish poison. lectins from Parkia biglandulosa also have mitogenicity and anti-proliferative activity.

This present research paper mainly focused on the quantitative estimation of its constituents by using HPTLC and FTIR. After the extraction with petroleum ether and methanol the active constituents were found to be sterols.

Crude plant extracts are generally a mixture of active and non-active compounds. The quantitative estimation of all active constituents is very important for pharmacological point. The percentage compositions of the active constituents are as follows: Compesterol was found to be 0.026%, β-Sitosterol 0.074%, Lupeol 0.037%, Gallic acid 1.079%.

INTRODUCTION

Historically, plants have served humankind as sources of foods, medicines, oils, biocides, waxes, and other useful substances. However, it was not until the early 19th century that active compounds were isolated in a pure form from plants, and late in the 19th century that the chemical structures of natural plant compounds could be determined. A revolution in chemical technology has occurred in the last 50 years. New technologies have enabled the isolation, identification, and subsequent synthesis of biological compounds. Although some chemical compounds found in plants cannot be synthesized today because of technical or economic constraints, an increasing number of chemical compounds are being produced in the laboratory. Despite these capabilities, renewed interest has developed in using naturally produced chemicals from plants (botano-chemicals) as sources of new food proteins, medicines, biocides, and other materials

The objective of the present study was quantitative studies using HPTLC and FTIR.

MATERIALS AND METHODS

Extraction Procedure: The bark of *Parkia biglandulosa* was collected and dried under shed, then powdered to a course powder. The powder was passed through the 49 # sieve and exhaustively extracted with different solvents

mentioned below in a soxhlet apparatus. The extract is evaporated under reduced pressure until all the solvent had been removed to give extract sample with the yield of 10 % w/w. The chemical constituents of the extracts were analyzed and found to contain steroids and tannins, conformed by thin layer chromatography. The extracts were stored in a refrigerator for further analysis.

Sample I Preparation: Dried bark of *Parkia biglandulosa* was powdered, 5 gm was weighed and extracted with Soxhlet apparatus by using petroleum ether at 90 ^{0} C, and the extract was filtered.

Sample II Preperation: Dried bark of *Parkia biglandulosa* was defatted and powdered, exact 5 gm was weighed and extracted with Soxhlet apparatus by using methanol at 60^{0} C and the extract was filtered.

Sample I & Sample II were estimated by using FTIR (JASCO -5300) along with the standards of Campesterol, Lupeol, Gallic aciḍ,β-Sitosterol.

RESULTS AND DISCUSSIONS

Crude plant extracts are generally a mixture of active and non-active compounds. The quantitative estimation of all active constituents is very important for pharmacological point. The percentage compositions of the active constituents are as follows: Compesterol 0.026%, Lupeol 0.037%, Gallic acid 1.079%, β-Sitosterol 0.074%.

Description about the samples extracted with Petroleum ether.

Sample PB1: The compound isolated with petroleum ether was white amorphous substance and soluble in chloroform and benzene. It has the melting point range of 156-158 ^{0}C with the Rf value 0.5. The Liberman Burchard test shows green ring at the junction which shows the presence of sterols. The FTIR data was presented in Fig 1 and Table 1. The TLC and FTIR data confirms that the compound was Campesterol

Sample PBM_{22}: The compound isolated with methanolic extract of bark. It was pale yellow colour crystalline compound. Soluble in boiling water, ether, glycerol acetone and has the melting point range of 236- 239 ^{0}C. The compound had Rf value 0.79. The FTIR data was presented in Fig 2 and Table 2. The TLC and FTIR data of PBP_3 confirms that the compound was Gallic acid

Sample PBP_3: The compound isolated with petroleum ether was colour less glossy needle shaped crystals. Soluble in chloroform and benzene and has the melting point range of 213- 215 ^{0}C with the Rf value 0.18. The FTIR data was presented in Fig 3 and Table 3. The TLC and FTIR data of PBP_3 confirms that the compound was Lupeol.

Sample PBP_2: The compound isolated with petroleum ether was white amorphous substance and soluble in chloroform and benzene. It has the melting point range of 138-141 ^{0}C with the Rf value 0.37. The Liberman Burchard test shows green ring at the junction which shows the presence of sterols. The FTIR data was presented in Fig 4 and Table 4. The TLC and FTIR data of PBP_2 confirms that the compound was β-Sitosterol.

Fig 1
Sample code: PBP-1 (IR of Campesterol)

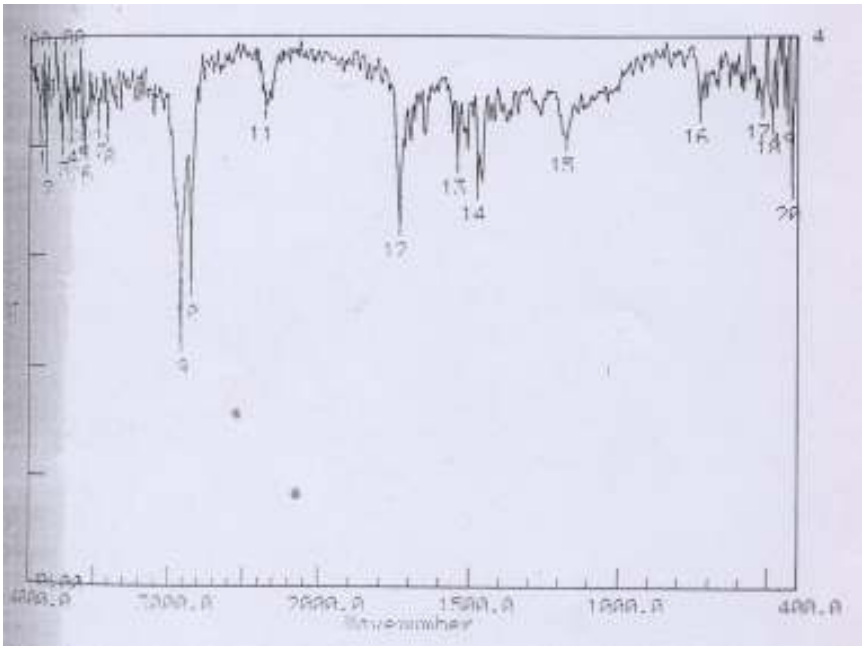
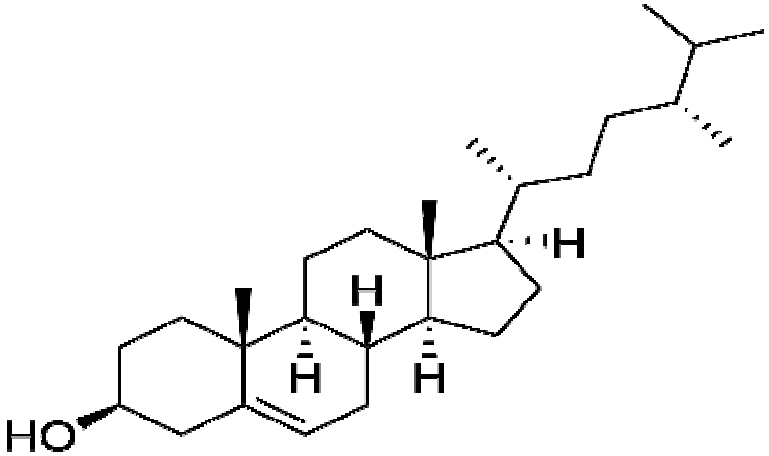


Table 1
(Campesterol)

S.No	Assignment	Peak
1	O-H	3497.97
2	C-H (alkane)	2916.63
3	C=C(Non conjugated)	1734.16
4	-CH_3 C-H bending	1473.75
5	-CH2- Twisting	1176.68
6	C-H bending (Alkenes)	779.16

Structure of Campesterol



Sample code: PBM-22 (IR of Gallic acid)

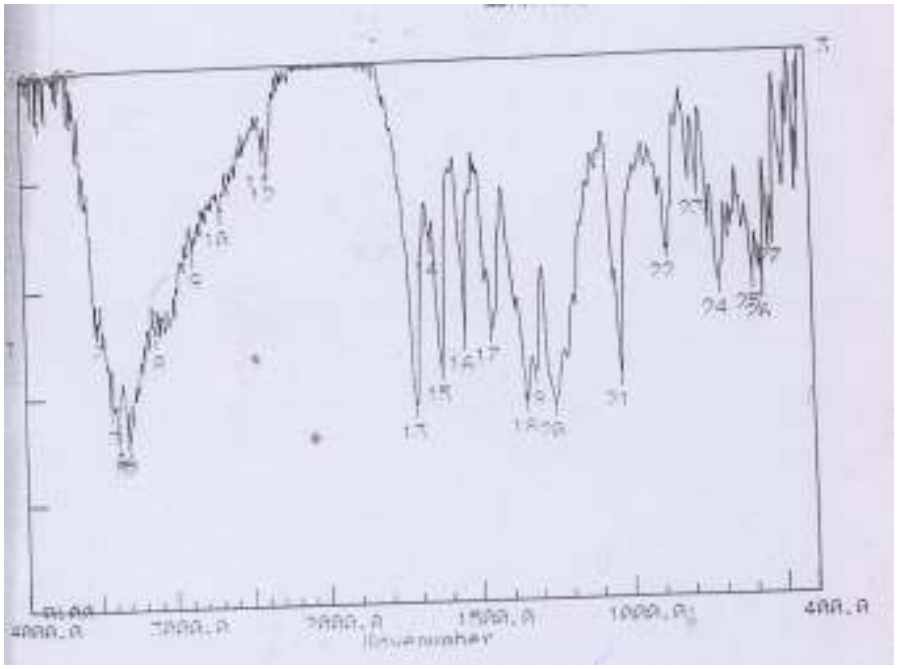
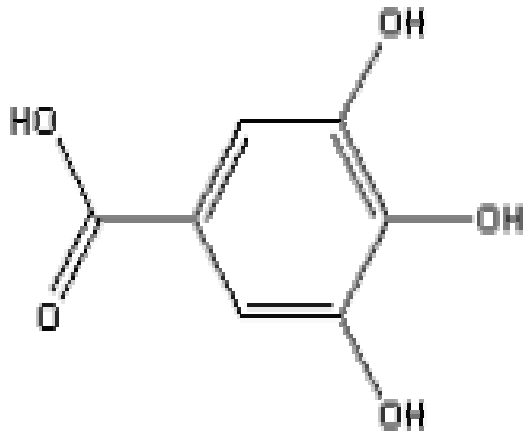


Table no 2
(Gallic acid)

S.No	Assignment	Peak
1	O-H	3408.52
2	Ar. C-H	3097.96
3	C=O	1705.23
4	C=C	1647.36
5	-C-O	1309.78
6	Ar. C-H bend	868.05

Structure of Gallic acid



Sample code : PBP-3 (IR of Lupeol)

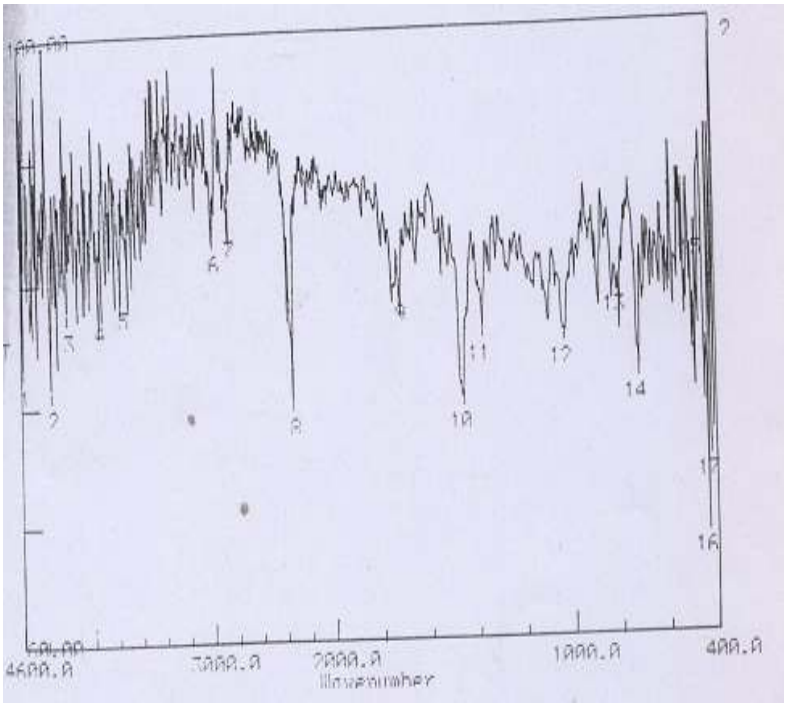
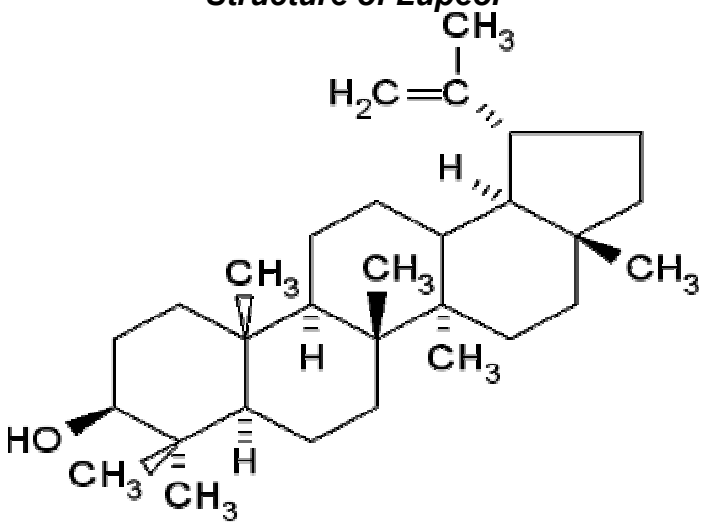


Table 3
(Lupeol)

S.No	Assignment	Peak
1	O-H	3244.17
2	C-H (alkane)	2976.43
3	C=C(Non conjugated)	1722.59
4	C-H bending (Methylene)	1456.39
5	-CH bending (Methyl)	1373.68
6	C-H bending (Alkenes)	1373.44
7	O-H bending	1032.01

Structure of Lupeol



Sample code : PBP-2 (IR of β-Sitosterol)

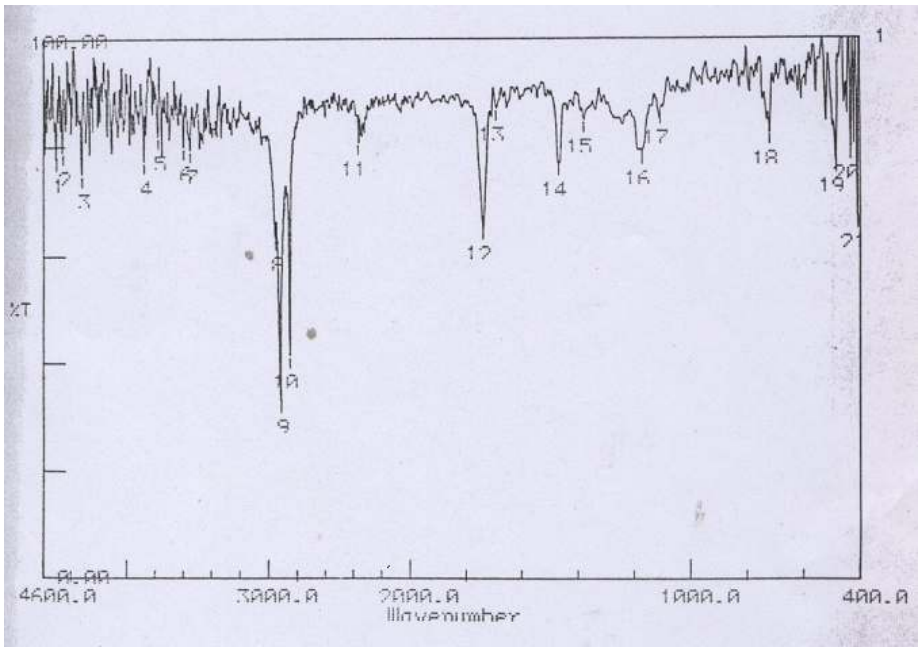
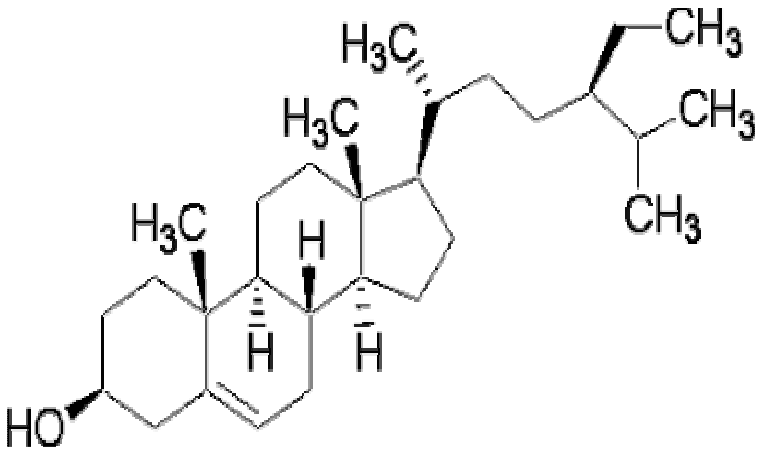


Table 4
(β-Sitosterol))

S.No	Assignment	Peak
1	O-H	3547.41
2	C-H (alkane)	2953.28
3	C=C(Non conjugated)	1739.95
4	C-H bending	1469.89
5	-CH2- Twisting	1174.75
6	C-C	1107.74
7	CH2-rocking	719.51

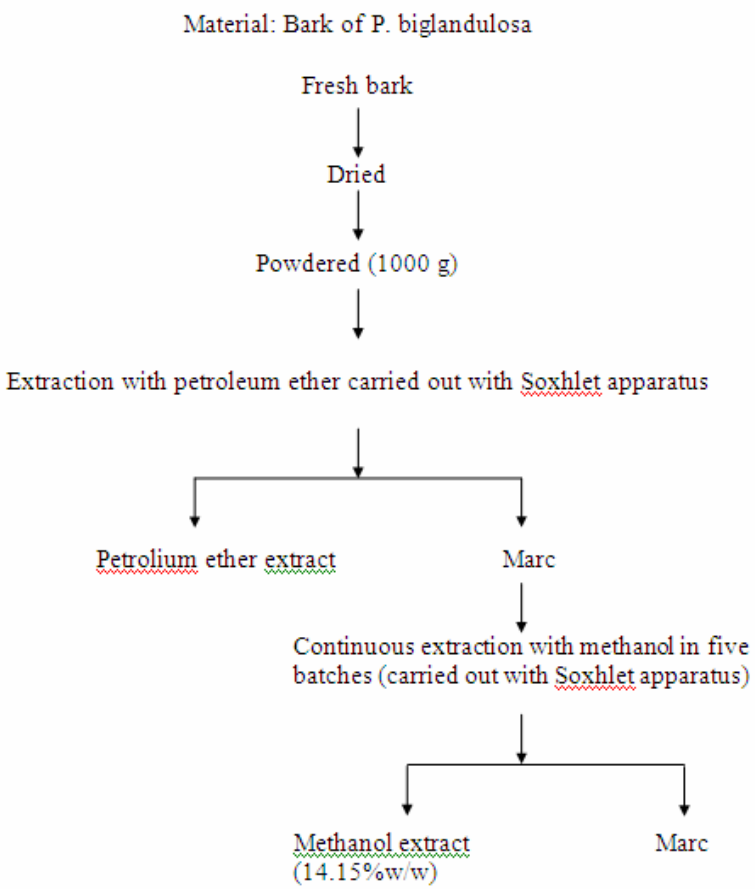
Structure of β-Sitosterol



CONCLUSION

Recent reports revealed the presence of steroids in different varieties of Parkia. The pharmacological activity of the fractions of the bark extract of *Parkia biglandulosa* has drawn our attention to study the chemical and biological activity of this plant. One of our efforts to discover the structures of biologically significant fractions of the extract lead to the isolation of four known compounds name as Campesterol, , Lupeol, Gallic acid, β-Sitosterol with spectral and chemical analysis.

Flow chart of Extraction Methodology



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