ABSTRACT

The thesis deals with the isolation and structural studies of the various constituents from the following Indian medicinal plants:
(1) Stem bark of Diospyros discolor Willd.
(2) Seeds of Peganum harmala Linn.
(3) Stem bark of Aegle marmelos Corr.

Antimicrobial activity of the ethanolic extract isolated from the stem bark of Aegle marmelos has also been done and included in this thesis. The thesis has been divided into four chapters.

CHAPTER - I

The first chapter is introductory. It deals with the importance and development of phytochemistry with particular emphasis on the chemistry of anthraquinones, tannins and coumarins. Some of the important compounds isolated from various plant sources have also been described with their pharmacological uses in medicines.

CHAPTER - II

This chapter deals with the isolation, purification, crystallisation and structural study of the various constituents isolated by the authoress from the stem bark of Diospyros discolor. A survey of literature available on the chemistry of Diospyros discolor has also been incorporated in this chapter. Three
unreported compounds - A, B and C have been isolated from the stem bark of this plant by the authoress and their structural study have been done by the application of chemical and spectral methods. The detailed investigation of these compounds have been discussed separately in three different sections- I, II and III respectively. A brief description of the compounds - A, B and C is mentioned herein.

**Compound - A**: Molecular formula, C_{21}H_{20}O_{12}, m.p. 224-25\(^\circ\) (dec.), was found to be an anthraquinone glycoside by its various specific colour reactions and spectral studies. Acid hydrolysis of the compound-A with 7% H\(_2\)SO\(_4\) yielded an aglycone and a sugar which was identified as D-glucose by its Co-paper chromatography and osazone formation.

The aglycone, molecular formula, C_{15}H_{10}O_{7} (M\(^+\) at m/e 302) m.p. 308-10\(^\circ\) (dec.), was found to contain five hydroxyl groups (by the formation of its pentaacetate). The aglycone on zinc dust distillation gave 2-methyl-anthracene showing the presence of a methyl group at position-2 in the aglycone. On the basis of various specific colour reaction, ultra violet-visible spectra, methylation, acetylation, chromic acid oxidation and also by spectral studies, the aglycone was identified as 1, 3, 5, 6, 8-pentahydroxy-2-methyl-anthraquinone.

The compound-A could be hydrolysed with almond enzyme emulsin solution to give D-glucose (Co-paper chromatography) and an aglycone 1, 3, 5, 6, 8-pentahydroxy-2-methyl-anthraquinone (m.m.p.
and CO-thin layer chromatography) indicating the presence of 
β-glycosidic linkage between the aglycone and D-glucose.

The periodate oxidation of the compound-A showed that one 
molecule of the compound-A was built up of one molecule of 
D-glucose and an aglycone. Thus the compound-A was identified as 
1, 3, 5, 6-tetrahydroxy-2-methyl-anthraquinone-8-O-β-D-glucosepyranoside.

Hence the compound-A has been assigned by the following 
structure:

![Chemical structure of compound-A](image)

**STRUCTURE OF THE COMPOUND-A**

[1,3,5,6-TETRAHYDROXY-2-METHYL-ANTHRAQUINONE-8-O-β-D-GLUCOPYRANOSIDE]

**Compound-B**: Molecular formula, C_{22}H_{22}O_{12}, m.p. 255-56°C, was also 
found to be an anthraquinone glycoside by its various specific 
colour reactions and spectral studies. Acid hydrolysis of the 
compound-B with 7% H_{2}SO_{4} yielded an aglycone and a sugar which was
identified as D-glucose by its CO-paper chromatography and osazone formation.

The aglycone, molecular formula $C_{16}H_{12}O_7$ ($M^+$ at m/e 316), m.p. 150-53° (dec.), was found to contain four hydroxyl groups (by the formation of its tetraacetate) and one methoxyl group (by Zeisel's method of methoxyl group estimation). The aglycone on zinc dust distillation gave 2-methyl-anthracene confirming the presence of a methyl group at position-2 in the aglycone. The aglycone was identified as 1,3,5,8-tetrahydroxy-6-methoxy-2-methyl-anthraquinone on the basis of various specific colour reactions, ultra violet-visible spectra, methylation, acetylation, chromic acid oxidation, demethylation and also by its spectral studies.

The compound-B could be hydrolysed with almond enzyme emulsin solution to give D-glucose (CO-paper chromatography) and an aglycone 1,3,5,8-tetrahydroxy-6-methoxy-2-methyl-anthraquinone (m.m.p. and CO-thin layer chromatography) indicating the presence of β-glycosidic linkage between the aglycone and D-glucose. The periodate oxidation of the compound-B showed that one molecule of the compound-B was built up of one molecule of D-glucose and an aglycone. Thus the compound-B was characterized as 1, 3, 5-trihydroxy-6-methoxy-2-methyl-anthraquinone-8-0-β-D-glucopyranoside.

Hence the compound-B has been assigned by the following structure:
STRUCTURE OF THE COMPOUND-B

[1,3,5-TRIHYDROXY-6-METHOXY-2-METHYL-ANTHRAQUINONE-8-O-β-D-GLUCOPYRANOSIDE]

Compound - C: Molecular formula, C$_{28}$H$_{30}$O$_{18}$, m.p. 220-220°C (dec.), responded positive colour tests for a tannin glycoside. Acid hydrolysis of the compound-C with 7% H$_2$SO$_4$ yielded an aglycone and sugars which were identified as D-glucose and D-galactose (CO-paper chromatography).

The aglycone, molecular formula, C$_{10}$H$_{10}$O$_{8}$, m.p. 273-74°C was found to be identical with 3,3'-di-O-methyl ellagic acid on the basis of its m.p., various colour reactions, preparation of their various derivatives and spectral studies which was finally confirmed by its m.m.p. and CO-thin layer chromatography with an authentic sample.

Periodate oxidation of the compound-C showed that one molecule of the compound-C was built up of one molecule of each sugar, D-glucose, D-galactose and an aglycone. Partial hydrolysis
of the compound-C with 7% H$_2$SO$_4$ showed that D-galactose was the terminal sugar. Permethylation of the compound-C (Hakomori's method) followed by acid hydrolysis afforded 2,3,4,6-tetra-O-methyl-D-galactose ($R_G$ value and CO-paper chromatography); 2,3,6-tri-O-methyl-D-glucose ($R_G$ value and CO-paper chromatography) and 3,3',4'-tri-O-methyl ellagic acid (m.m.p. and CO-thin layer chromatography) confirming the presence of both the sugars at position-4 in the aglycone with 1$\rightarrow$4 intersugar linkage. Enzymatic hydrolysis of the compound-C showed $\beta$-linkage between D-galactose and D-glucose, and D-glucose and an aglycone. Thus the compound-C was assigned as 3,3'-di-O-methyl-ellagic acid-4-O-$\beta$-D-galactopyranosyl-(1$\rightarrow$4)-$\beta$-D-glucopyranoside.

Hence the compound-C has been assigned by the following structure:

![Structure of the compound-C](image)

**STRUCTURE OF THE COMPOUND-C**

[3,3'-DI-O-METHYL-ELLAGIC ACID-4-O-$\beta$-D-GALACTOPYRANOSYL-(1$\rightarrow$4)-$\beta$-D-GLUCOPYRANOSIDE]
CHAPTER III

The third chapter deals with the isolation, purification, crystallisation and structural studies of the two compound isolated from seeds of *Peganum harmala*. A survey of literature available on the chemistry of *Peganum harmala* has also been incorporated in this chapter. Two unreported compounds - A and B have been isolated from the seeds of this plant by the authoress and their structural studies have been done by the application of chemical and spectral methods. The detailed investigation of these compounds have been discussed separately in two different sections- I and II respectively.

**Compound - A**: Molecular formula, $C_{16}H_{12}O_4 \ (M^+ \text{ at m/e } 268)$, m.p. 269-70°C, gave negative Molisch's test showing the absence of glycosidic nature of the compound-A. The compound-A was found to be an anthraquinone by its specific colour reactions and spectral studies.

The compound-A was found to contain one hydroxyl group (by the formation or its monoacetate) and one methoxyl group (by Zeisel's method of methoxyl group estimation). The compound-A on zinc dust distillation gave 2 methyl-anthracene showing the presence of a methyl group at position-2 in the compound-A. The compound-A was identified as 8-hydroxy-7-methoxy-2-methyl-anthraquinone on the basis of its various specific colour reactions, acetylation, methylation, demethylation, chromic acid oxidation and also by spectral studies.
Hence the compound-A has been assigned by the following structure:

\[ \text{STRUCTURE OF THE COMPOUND-A} \]

[8-HYDROXY-7-METHOXY-2-METHYL-ANTHRAQUINONE]

**Compound - B** : Molecular formula, \( \text{C}_{16}\text{H}_{12}\text{O}_{5} \) (\( \text{M}^+ \) at m/e 284), m.p. 190-92\(^{0}\), gave negative Molisch's test showing the absence of glycosidic nature of the compound-B. The compound-B was found to be an anthraquinone by its specific colour reactions and spectral studies.

The compound-B was found to contain two hydroxyl groups (by the formation of its diacetate) and one methoxyl group (by Zeisel's method of methoxyl group estimation). The compound-B on zinc dust distillation gave 2-methyl-anthracene showing the presence of a methyl group at position-2 in the compound-B. The compound-B was characterized as 3,6-dihydroxy-8-methoxy-2-methyl-anthraquinone on the basis of its various specific colour reactions, acetylation, methylation, demethylation, chromic acid oxidation and also by spectral studies.
Hence the compound-B has been assigned by the following structure:

![Structure of the Compound-B](image)

**STRUCTURE OF THE COMPOUND-B**

[3,6-DIHYDROXY-8-METHOXY-2 METHYL-ANTHRAQUINONE]

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**CHAPTER - IV**

This chapter has been divided into two parts - A and B respectively.

**PART - A**

This part deals with the isolation, purification, crystallisation and structural study of only one known compound isolated by authoress from the stem bark of *Aegle marmelos*. A survey of literature available on the chemistry of *Aegle marmelos* has also been incorporated in this part.

The structural study of the compound isolated in the part-A has been described in section-I of this part. A brief description of this compound is mentioned herein.
SECTION - I:

STUDY OF THE COMPOUND:

The detailed study of infra-red spectrum, $^1$H-NMR spectrum, mass spectrum, melting point and preparation of various derivatives of the compound confirmed its identity as marmin.

Hence the structure of this compound has been represented by the following structure:

![Structure of the Compound](image)

**STRUCTURE OF THE COMPOUND (MARMIN)**

PART - B:

This part deals with the importance and development of the antimicrobial activity along with some recent ideas about the use of drugs as antimicrobial agents. The experimental technique has also been included in this part.

This part has been further divided into two sections - II and III to describe the antibacterial and antifungal activity of the ethanolic extract isolated from the stem bark of *Aegle marmelos*.
For the antibacterial and antifungal activities, the following bacteria and fungi were selected during the study:

**NAME OF BACTERIA**
(a) *Vibrio cholerae*
(b) *Salmonella typhimurium*
(c) *Klebsiella pneumonia*

**NAME OF FUNGI**
(a) *Candida albicans*
(b) *Aspergillus fumigatus*
(c) *Trichophyton mentagrophytes*

**SECTION - II**

It deals with the antibacterial activity of the ethanolic extract isolated from the stem bark of *Aegle marmelos* which was tested on the above said bacteria. This extract was found to be effective against all the bacteria tested and can be used as antibacterial agents for therapeutic purpose against the disease caused by the above bacteria.
SECTION - III

It deals with the antifungal activity of the ethanolic extract isolated from stem bark of Aegle marmelos which was tested on the above said fungi. This extract was found to be effective against all the fungi tested. This extract showing such activity that could be employed as surface applicants as preventive measure for the dermal diseases caused by the above fungi.

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