ABSTRACT

The thesis deals with the isolation and structural studies of the various constituents from the following Indian medicinal plants:

(1) Whole plant of Limonia crenulata Roxb.
(2) Heartwood of Aegle marmelos Corr.
(3) Stem bark of Aegle marmelos Corr.

Anti-inflammatory activity of the ethanolic extract and a new compound isolated from the whole plant of Limonia crenulata and the analgesic activity of a new compound isolated from the heartwood of Aegle marmelos have also been given in this thesis. The thesis has been divided into five chapters.

CHAPTER - I

The first chapter is introductory. It deals with the importance and development of the chemistry of natural products with particular emphasis on the chemistry of steroids, terpenoids, anthraquinones, coumarins and esters. Some 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 from the whole plant of *Limonia crenulata*. A survey of the available literature on the chemistry of *Limonia crenulata* has also been incorporated in this chapter. Four compounds viz. A, B, C and D have been isolated and characterised by the author 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 four different sections - I, II, III and IV respectively. A brief description of the compounds - A, B, C and D is mentioned herein.

**Compound - A:** Molecular formula, $C_{29}H_{50}O$ ($M^+$ 414), m.p. 135-136° was found to be identical with β-sitosterol by m.p., chemical reactions, IR spectrum, mass spectral data and also by its m.m.p. and co-TLC with an authentic sample.

Hence the structure of the compound-A has been assigned as follows:

![Structure of Compound-A: β-sitosterol](image)

**Compound-A: β-sitosterol**

**Compound - B:** Molecular formula, $C_{30}H_{50}O$ ($M^+$ 426), m.p. 185-186° was found to be a triterpene by its specific colour tests. The IR spectrum of the compound-B was found to be
identical with α-amyrin which was further confirmed by chemical reactions, mass spectrum, m.m.p. and co-TLC with an authentic sample.

Hence the compound-B has been represented by the following structure.

\[
\text{Compound-B : α-amyrin}
\]

**Compound - C**: Molecular formula, \(C_{22}H_{22}O_{10}\), m.p. 157-158° was found to be an anthraquinone glycoside by its specific colour reactions. Acid hydrolysis of the compound-C with 7% \(H_2SO_4\) afforded an aglycone and a sugar. The sugar was identified as D-glucose by \(R_f\) value, osazone formation and co-PC with an authentic samples.

The aglycone, molecular formula, \(C_{16}H_{12}O_{5}\) (\(M^+ 284\)), m.p. 190-192° was responded colour reactions specific for an anthraquinone. The aglycone was found to contain two hydroxyl groups (by its diacetate formation) and one methoxyl group (by Ziesel's method). The aglycone gave 2-methyl anthracene on zinc dust distillation showing the presence of a methyl group at position-2 in the aglycone. The aglycone was identified as 3,8-dihydroxy-6-methoxy-2-methylanthraquinone by its various
specific colour reactions, and also by spectral studies. This aglycone have reported for the first time in nature.

The compound-C on hydrolysis with almond enzyme emulsin solution gave the aglycone and D-glucose showing the presence of β-linkage between the aglycone and D-glucose. The periodate oxidation of compound-C showed that compound-C is built up of one molecule of D-glucose and the aglycone. The sugar, D-glucose was found to be attached with C-3 hydroxyl group of the aglycone part by its chemical reactions. Thus the new compound-C was identified as 8-hydroxy-6-methoxy-2-methylantraquinone-3-O-β-D-glucopyranoside.

Hence the structure of the compound-C has been assigned as follows:

\[
\begin{align*}
\text{Compound-C : 8-hydroxy-6-methoxy-2-methylantraquinone-3-O-β-D-glucopyranoside}
\end{align*}
\]

Compound - D : Molecular formula, \( C_{27}H_{26}O_{12} \), m.p. 182-183\(^\circ\) was found to be a coumarin glycoside by its various specific colour reactions. Acid hydrolysis (7% \( H_2SO_4 \)) of the compound-D
yielded an aglycone and sugar which were identified as D-glucose and L-rhamnose (co-IPC).

The aglycone, molecular formula C_{13}H_{11}O_{3} (M^+ 162) gave specific reactions for a coumarin. The aglycone was identified as umbelliferone by its various chemical reactions, spectral studies and finally confirmed by its m.m.p. and co-TLC with an authentic sample.

Periodate oxidation of the compound-D showed that the sugars were present as a disaccharide unit. Partial acid hydrolysis of the compound-D L-rhamnose showed as the terminal sugar. The compound-D on takadiastase enzyme hydrolysis gave L-rhamnose (co-PC) and a product which on almond emulsin hydrolysis afforded D-glucose (co-PC) and umbelliferone (m.m.p. and co-TLC) indicating the presence of α-linkage between L-rhamnose and D-glucose, and β-linkage between D-glucose and the aglycone.

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

![Chemical structure of compound D](image)

*Umbelliferone-7-O-α-L-rhamnopyranosyl(1→6)-β-D-glucopyranoside*
CHAPTER - 3

The third chapter deals with the isolation, purification, crystallisation and structural studies of the four compounds viz. A, B, C and D isolated from the heartwood of Aegle marmelos. A survey of literature available on the chemistry of Aegle marmelos has also been incorporated in this chapter. The detailed investigation of these compounds have been discussed separately in four different sections - I, II, III and IV respectively. A brief description of these compounds is mentioned herein.

Compound - A: Molecular formula, $\text{C}_{29}\text{H}_{50}\text{O} (M^+ 414)$, m.p. 134-135° was found to be identical with $\beta$-sitosterol by its chemical reactions, spectral studies and also by m.m.p. and co-TLC with an authentic sample.

Hence the structure of the compound-A has been assigned as follows:

![Compound-A: $\beta$-sitosterol](image)

Compound - B: Molecular formula, $\text{C}_{30}\text{H}_{50}\text{O} (M^+ 426)$, m.p. 211-213° was found to be a triterpenoid by its specific chemical reaction. The compound-B was found to be identical with lupeol by its m.p., chemical reactions, spectral data and also by
m.m.p. and co-TLC with an authentic sample.

Hence the structure of the compound-B has been assigned as follows:

![Structure of Compound-B: Lupeol](image)

**Compound-B : Lupeol**

**Compound - C :** Molecular formula, \( C_{11}H_{12}O_4 \) (M⁺ 208), m.p. 210-211° was found to contain a methoxyl group (by Zeisel's method) and one hydroxyl group (by its acetylation study). Ozonolysis of the compound-C gave a product which was found to be identical with 2-hydroxyanisaldehyde, m.p. 40-42° (m.m.p. and co-TLC). The detailed study of its IR, \(^1H\)-NMR, mass and \(^{13}C\)-NMR spectral data confirmed the identity of the new compound-D as 2-hydroxy-4-methoxy phenyl-1-vinyl acetate.

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

![Structure of Compound-C](image)

**Compound-C :** 2-hydroxy-4-methoxyphenyl-1-vinyl acetate

**Compound - D :** Molecular formula, \( C_{17}H_{16}O_9 \), m.p. 213-220° gave specific reactions for coumarin glycoside. The acid
hydrolysis (7% \( \text{H}_2\text{SO}_4 \)) of the compound-1 afforded an aglycone and D-glucose (co-PC).

The aglycone, molecular formula, \( \text{C}_{11}\text{H}_6\text{O}_4 \) \( (\text{M}^+ 202) \) gave all the positive reactions to a coumarin. The aglycone was found to contain a hydroxyl group (by its acetylation study). The aglycone was confirmed as xanthotoxol by its m.p., various chemical reactions, methylation study, IR, \(^1\text{H}-\text{NMR} \) and mass spectral data.

The periodate oxidation and almond enzyme emulsion hydrolysis of the new compound-1 further confirmed its structure as xanthotoxol-8-O-\( \beta \)-D-glucopyranoside.

Hence the structure of the compound-1 has been assigned as follows:

\[ \text{Compound-1}: \text{Xanthotoxol-8-O-\( \beta \)-D-glucopyranoside} \]

**CHAPTER - 4**

This chapter deals with the isolation, purification and structural study of the various constituents isolated from the stem bark of *Aegle marmelos* Corr. Three compounds viz.
A, B and C were isolated by the author and their structural study have been done with the help of various chemical reactions and spectral data. The detailed investigation of these compounds have been discussed separately in three different sections- I, II and III respectively. A brief discription of all the three compounds- A, B and C is mentioned herein.

**Compound - A**: Molecular formula, C_{30}H_{50}O \ (M^+ \ 426), m.p. 197-198^\circ \text{C} was found to be identical with epi-lupeol on the basis of its m.p. chemical reactions and spectral data. Finally the structure of the compound-A was confirmed as epi-lupeol by its m.m.p. and co-TLC with an authentic sample.

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

\[ \text{Compound-A: epi-lupeol} \]

**Compound - B**: Molecular formula, C_{19}H_{24}O_{5} \ (M^+ \ 332), m.p. 122-123^\circ \text{C} was found to be a coumarin by its specific chemical reactions. The detailed study of its IR, $^1$H-NMR and mass spectral data as well as preparation of various derivatives of the compound-B showed its identity as marmin which was finally confirmed by its m.m.p. and co-TLC with an authentic sample.
Hence the structure of the compound-B has been assigned as follows:

\[
\begin{align*}
\text{CH}_2-\text{CH}=&\text{C}-\text{CH}_2-\text{CH}=&\text{C}-\text{CH}_3 \\
\text{CH}_3 & \quad \text{OH} & \quad \text{OH} & \quad \text{CH}_3
\end{align*}
\]

**Compound-B : Marmin**

**Compound - C :** Molecular formula, \(C_{14}H_{14}O_4\) (M\(^+\) 246), m.p. 192-193\(^\circ\) gave all the positive reactions for a coumarin. The detailed study of its acetylation, oxidation, mild oxidation, dehydration, IR, \(^1\)H-NMR and mass spectral data of the compound-C showed its identity as Marmesin. Finally the structure of the compound-C as marmesin was confirmed by its m.m.p. and co-TLC with an authentic sample.

Hence the structure of compound-C has been assigned as follows:

\[
\begin{align*}
\text{CH}_3-\text{C} & \quad \text{CH}_3 \\
\text{CH}_3 & \quad \text{OH}
\end{align*}
\]

**Compound-C : Marmesin**

**CHAPTER - 5**

This chapter deals with the pharmacological evaluations of the extract and compounds from the above selected plants. This chapter has been divided into two section - I and II respectively.
SECTION - I : This section deals with the introduction, development and various methods which are used for evaluation of anti-inflammatory activity.

The ethanolic extract of *Limonia crenulata* and a new compound, 8-hydroxy-6-methoxy-2-methylanthraquinone-3-O-β-D-glucopyranoside were successfully screened by the author for anti-inflammatory activity using the method as described by Winter, Risely and Nuss.

The ethanolic extract and the new compound were found to exhibit significant inhibition (68.19 and 65.91% respectively) in comparison with standard drug oxyphenbutazone which showed 72.73% inhibition in the same condition on rats.

SECTION - II : This section describes the introduction, description and various methods for evaluation of analgesic activity. The new compound, 2-hydroxy-4-methoxyphenyl-1-vinyl acetate, isolated from heartwood of *Aegle marmelos* was screened for analgesic activity. The rat tail flick procedure was adopted by the author for the screening of the analgesic activity on a hot wire analgesiometer using morphine-HCl as a standard drug. The results revealed that this new compound showed good analgesic effect.