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Phtochemical Investigation And Characterization Of Bioactive Secondary Metabolites Rom Fruits Of *Terminalia Chebula*

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ABSTRACT

Phytochemical investigation and characterization of acetone extract of dried fruits of Terminaliachebula, well known medicinal plant (Family : Combretaceae) resulted in the isolation of three known bioactive secondary metabolitew, namely Stigmasterol (1), Chebulinic acid (2), Gallic acid (3) and Vanillin(4) . These compounds were identified by the comparision of spectroscopic data (^{1}H, ^{13}C NMR, IR and MASS) with reported data.

1.1 DEFINITION OF NATURAL PRODUCTS:

A Natural product is a chemical organic compound, found in the nature, produced naturally by a living organism. These compounds possess pharmacological and biological activity, to combat various diseases. Human societies used natural products, since millennia.

Natural products are the active components of many traditional medicines as well as modern medicines. They also have a great effect on culture of human's and they have been used throughout human history as condiments, pigments, and pharmaceuticals. They are lead compounds for drug discovery and the current importance of drugs from natural origin is undebatable.

1.2 HISTORY OF NATURAL PRODUCTS:

- Liquid CO2 is constrained into a supercritical state by controlling its temperature and weight.
- Supercritical CO2 has dissolvable power and concentrates dominatingly lipophilic and unstable mixes.

- Gaseous CO2 comes back to CO2 tank. After a full round, the new extraction begins with flowing CO2.

Counter current extraction:

This is a nonstop procedure in which the plant material moves against the dissolvable. It is reasonable system for generation of a lot of concentrates on a modern scale. A few sorts of extractors are accessible. In the screw extractor the plant material is transported by a screw through a tube and meets the dissolvable which is drawn the other way.

LITERATURE REVIEW

2.1 INTRODUCTION OF MANGROVES:

Mangroves develop in better water or beach front saline by and large they are little or bushes. The term is additionally utilized for tropical seaside vegetation including such species. They happen worldwide in the tropics and sub-tropics, more often than not between scopes 25 °S. As per the year 2000, the territory of mangroves was 53,200 square miles (137,800 km²), spreading over 118 nations and domains. They adjust well to cruel beach front conditions and in low oxygen (anoxic) states of waterlogged and complex salt filtration framework to deal with wave activity and salt water submersion.

The term is utilized as a part of three detects:

- By and large to allude to the entire plant and natural surrounding collection or mangal, for which the terms mangroves woods, mangroves bog and mangrove backwoods biome are additionally utilized.
- To allude to all bushes and trees in the mangrove overwhelm.
- To allud to the mangrove group of plants, the Combretaceae, or all the more particularly to mangrove trees of the variety Terminalia.

2.2 PLANT PROFILE:

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TERMINALIA CHEBULA

Terminaliachebula is a moderate tree used in traditional medicine. It is belonging to the family *Combretaceae*. It is commonly known as black myrobalan , Ink tree, (or) Chebulicmyrobalan and also known as king of medicine. It is extensively used in unani ,ayurveda , and homeopathy medicine.

Terminaliachebula is a popular traditional medicine not only in India but also in other countries of Asia and Africa. This is used in traditional due to the wide spectrum of pharmacological activities associated with the biologically active chemicals present in this plant.

Taxonomy:

Biological source: It is derived from the dried fruits of the *TERMINALIA CHEBULA* .Which belonging to the *family Combtretaceae.*

Systematic classification :

Kingdom : Plante

Clade : Angiosperms

Division : Mangnoliophyta

Class : Mangloliopsida

Subclass : Epigynae

Order : Myrtales

Family : Combretaceae

Genus : Terminalia

Species : Chebula

Vernacular names of T. chebula :Karakkaya , haritaki (India),Aralu(Srilanka),Zhang-Qin-Ge , Hezin (China),Harra, Harro in Tibet, Myrobalan (Grmany and France)

Common names: Buceraschebula(Retz.) Lyons; CombretumargyrophyllumK.Schum ;Myrobalanuschebula(Retz.) Gaertn.Terminaliazeylanica Van Heurck&Mull . Arg.; TerminaliatommentellaKurz ;Terminalia reticulate Roth.

2.3 DISTRIBUTION:

Terminaliachebula is found in throughout South East Asia .It is grown in all conditions. The *T.chebula* plant is spread in all Asian countries.

Countries:

Asian: India, Srilanka , Thailand , Pakistan, Malasia, Indonessia, Vietnam, Nepal, Cambodia.

West-Africa: Kenya, Madagascar, Maldives, Mauritius, Mozambique, Somalia, South Africa, Susan, Tanzania.

3.1 THE AIM AND OBJECTIVE OF PRESENT WORK:

AIM:

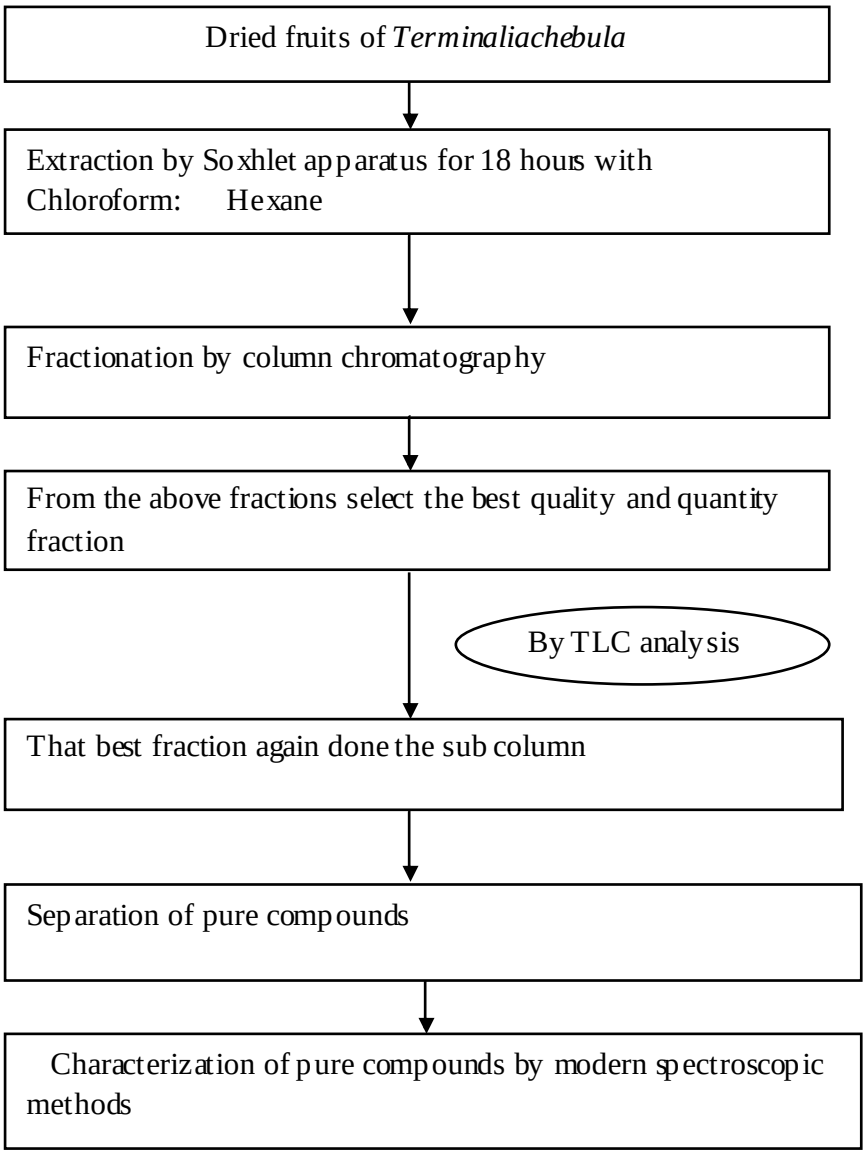
To extract, isolate and characterize the chemical constituents from the dried fruits of *Terminaliachebula*.

Objectives:

The main objective of the study was

- Phytochemical Investigation of the plant
- To isolate chemical constituents by using column chromatographic techniques.
- To characterize the various isolates using modern spectrometric methods.

3.2 PLAN OF WORK:



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4.0 MATERIALS AND METHODS

4.1. COLLECTION AND AUTHENTICATION

Fruits of *Terminaliachebula*(4 Kg) was collected from Srikakulam mangrove forest in the Andhra Pradesh (Latitude 15º 54' N and Longitude 80º 43' E) of India in March 2009 and was authenticated by Prof. SaluguBodayya pedal, Department of Botany, Andhra University, Visakhapatnam. A voucher specimen (# IIC-MG-111) has been deposited at the Herbarium of Natural Products Chemistry, I.I.C.T.

After drying fruits of *Terminaliachebula* under shade, segregated, pulverized by a mechanical grinder and passed through a (40 mesh) sieve at "Indian Institute of chemical technology", Tarnaka.

4.2 EXTRACTION PROCESS:

The coarsely powdered shade – fruits of *Terminaliachebula* (2.5Kg) were successively extracted by Soxhlet with chloroform, acetone and methanol in a soxhlet apparatus for 36h. The resulting extract was then concentrated under vacuum to obtain a residue (1.5g, 8.5g) respectively. The hexane extract (6.5g) was subjected to vacuum liquid chromatography over a column of silica gel (230-400 mesh) and eluted with n-hexane / ethyl acetate combinations of increasing polarity. Hexane extract furnished one compound pmgsgsl. The structure of these compound were elucidated by using ^{1}H NMR, ^{13}C NMR, IR and Mass data. Separation and purification of the compounds were discussed in the schematic representation of the isolation procedure is given below.

Soxhlet apparatus:

It is a hot percolation extraction method.

It has three main sections:

- A percolator (reflux and boiler) it circulates the solvent.
- A thimble (commonly made of thick filter paper) which retains or hold the solid to be loved.
- A siphon mechanism, which periodically empties the thimble.

Procedure

- The powdered material (fruits) containing the compound to be separated was put inside the thimble with in the primary assembly of the Soxhlet extractor. Refining flagon was loaded with required dissolvable and was set on warming mantle.
- The Soxhlet extractor was set ontop of the cup and reflux condenser was put on of the extractor.

4.1 Soxhlet apparatus

Rotary Evaporator:

The main components of a rotary evaporator are:

- An engine unit that turns the vial containing the client's example or dissipation cup.
- A vapour channel that is a vacuum-tight conductor for the vapour being drawn off the example and is the hub for test revolution.
- A vacuum framework, to limit the weight inside the evaporator framework.
- A liquid shower which is warmed (by and large containing water) to warm the example.

4.2 **Rotary Evaporator**

- A condenser into which coolant blends, for example, CH3)2CO and dry ice are set is furnished with either a "chilly finger" or a loop passing coolant.
- A condensate-a gathering carafe at the base of the condenser, to gather the refining dissolvable after it re-consolidates.
- A mechanized or mechanical to rapidly lift the vanishing from the warming shower.

5.0 RESULTS AND DISCUSSION

5.1 The phytochemical screening was done by *Terminalachebula* extracts with chloroform solvent. Chloroform was done by colour test. The results were presented as below.

Table 5.1 Phytochemical present in the Chloroform extract of *Terminaliachebula* fruits

Phytochemical screening	Chloroform extract
Alkaloids	+
Tannins	+
Saponins	+
Steroids	+
Triterpenoids	+
Glycosides	+
Flavonoids	+
Reducing Sugar	+

SUMMARY AND CONCLUSION

Isolation and Identification studies conducted on fruits of *Terminaliachebula*were indicated in this thesis.

Chapter I

In this chapter emphasis has been laid on the role of medicinal potential source of therapeutic agents and the role played by natural products in drug discovery.

Chapter II

This chapter contains a brief introduction on mangroves. This chapter also contains review of literature like botanical information, plant description, and previously reported phytochemical work *Terminaliachebula*has been incorporated.

Chapter III

This chapter describes the aim and objectives of the present work. It also contains brief explanation about the plant of the work.

Chapter IV

This chapter describes the collection, authentication and extraction procedure employed and also describes the chromatographic technique employed in isolation of molecules; from the Chloroform extract of *Terminaliachebula*fruits.

Chapter V

This chapter explains isolate the bioactive molecules from the mangrove plant *Terminaliachebula*fruits using gradient elution technique by column chromatography structural elucidation and characterization of molecules using spectral data (^{1}H, ^{13}C NMR Mass and IR) of

isolated compounds. The compounds that were isolated are **Stigmasterol, Chebulinic acid, Gallic acid and Vanillin.**

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