IMPACT OF DIOECISM ON QUALITY AND EFFICACY OF SANSHAMNI VATI

Thesis Submitted for the Award of the degree of

DOCTOR OF PHILOSOPHY

in

Ayurvedic Pharmacy

By

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2025

DECLARATION

I hereby declare that the presented work in the thesis entitled "Impact of Dioecism on Quality and Efficacy of Sanshamni Vati" is submitted in fulfilment of the degree of **Doctor of Philosophy (Ph.D)** is outcome of research work carried out by me under the supervision of Dr Manish Vyas, working as Professor, in the school of pharmaceutical sciences of Lovely Professional University, Punjab, India. In keeping with general practice of reporting scientific observations, due acknowledgements have been made whenever work described here has been based on findings of other investigator, this work has not been submitted in part or full to any other university or institute for the award of any degree.

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CERTIFICATE

This is to certify that the work reported in the Ph.D thesis entitled "Impact Of Dioecism On Quality And Efficacy Of Sanshamni Vati" submitted in fulfilment of the requirement for the award of degree of **Doctor of Philosophy (Ph.D)** in the school of pharmaceutical sciences, is a research work carried out by Madhurima,41900801, is Bonafide record of her original work carried under my supervision and that no part of thesis has been submitted for any other degree, diploma or equivalent course.

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ABSTRACT

A popular Ayurvedic formulation called Shanshamni Vati is well known for its medicinal properties in treating ailments including fevers and infections. The botanical source of its constituents, however, may have an impact on its quality and effectiveness. The presence of separate male and female plants within a species, or dioecism, is particularly significant for the pharmacological characteristics of medicinal herbs. This study investigates how dioecism affects Shanshamni Vati's effectiveness and quality.

Methods: Shanshamni Vati preparations made from the male and female plants of important dioecious species that were employed in the formulation were compared. The phytochemical makeup, the amounts of bioactive compounds, and the pharmacological actions in vivo were among the parameters evaluated. To guarantee uniformity, standardized techniques were applied to the extraction, quantification, and efficacy testing processes.

Result: The phytochemical profiles and quantities of bioactive compounds in the Shanshamni Vati made from male and female plants near about same, according to the research. Plants that are female in nature typically provide larger amounts of active ingredients like alkaloids, steroids, which are associated with more potent pharmacological effects. In vivo experiments showed that Shanshamni Vati derived from female plants had stronger immunomodulatory and anti-inflammatory effects compared to those from male plants. Discussion: The results imply that dioecism has an important impact on Shanshamni Vati's therapeutic efficacy and quality. Female plants provide more effective formulations due to their little higher phytochemical profiles. This emphasizes how crucial it is to use the right botanical sources for Ayurvedic formulations in order to ensure maximum therapeutic benefits. Moreover, these findings support the uniformity of plant gender during the medicinal use growing and harvesting phases. Conclusion: Shanshamni Vati's effectiveness and quality are greatly influenced by the sex of dioecious plants used in its preparation. The effectiveness and quality of Ayurvedic preparations are greatly influenced by the gender of dioecious plants. Proper identification and use of the ideal gender can enhance their medicinal value, thereby contributing to the development of more efficacious and standardized herbal remedies. For Ayurvedic pharmacology to completely profit from gender-specific plant qualities, more study and standardization procedures are advised.

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Date: August 2025 Dr Madhurima

Place: Jalandhar

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LIST OF TECHNICAL TERMS

ABBREVIATIONS / SYMBOLS	FULL FORM
T. cordifolia	Tinospora cordifolia
LOD	Loss on drying
TA	Total ash value
TNF-β	Tumour necrosis factor
IL-6	Interleukin-6
TGF-β	Transforming Growth Factor-Beta
IFN	Interferon
LOD	Loss on drying
AIA	Acid Insoluble ash
WSA	Water soluble extractive
WSA	Water soluble ash
RI	Refractive index
ASE	Alcohol soluble extractive
SA	Sulphated ash
KOH	Potassium Hydroxide
2N HCl	Hydrochloric Acid (2 Normal solution)
CHCl3	Chloroform
H2SO4	Sulfuric Acid
FeCl3	Ferric chloride
Ph	Potential of Hydrogen
Ppm	Parts Per Million
Hg	Mercury
Cd	Cadmium
As	Arsenic
Pb	Lead
HNO3	Nitric Acid
$HCLO_4$	*Perchloric Acid*

AAS	Atomic absorption spectroscopy
µg/ml	Micrograms per milliliter
AAS	*Atomic Absorption Spectroscopy*
GC-MS	*Gas Chromatography–Mass Spectrometry*
v/v	Volume by Volume
µL	Microliter (10^{-6} liter)
Mm	Millimeter
CAMAG	Swiss company
TLC	Thin Layer Chromatography
w/v	Weight by Volume
ELISA	Enzyme-Linked Immunosorbent Assay
SD	Standard Deviation
N	Sample size(number of observations)
P	Probability value (p-value)
ANOVA	Analysis of Variance
A.P.I.	Ayurvedic Pharmacopia of India
fu/g	Forming Units per Gram
Nd	Not Detected
FM	Female
M	Male
TSC	Total Solid Content
Rf	Retention Factor
HB	Haemoglobin
RBC	Red Blood Cells
WBC	White Blood Cells
ESR	Erythrocyte Sedimentation Rate
Bw	Body weight
NSP>0.001	Not Significant

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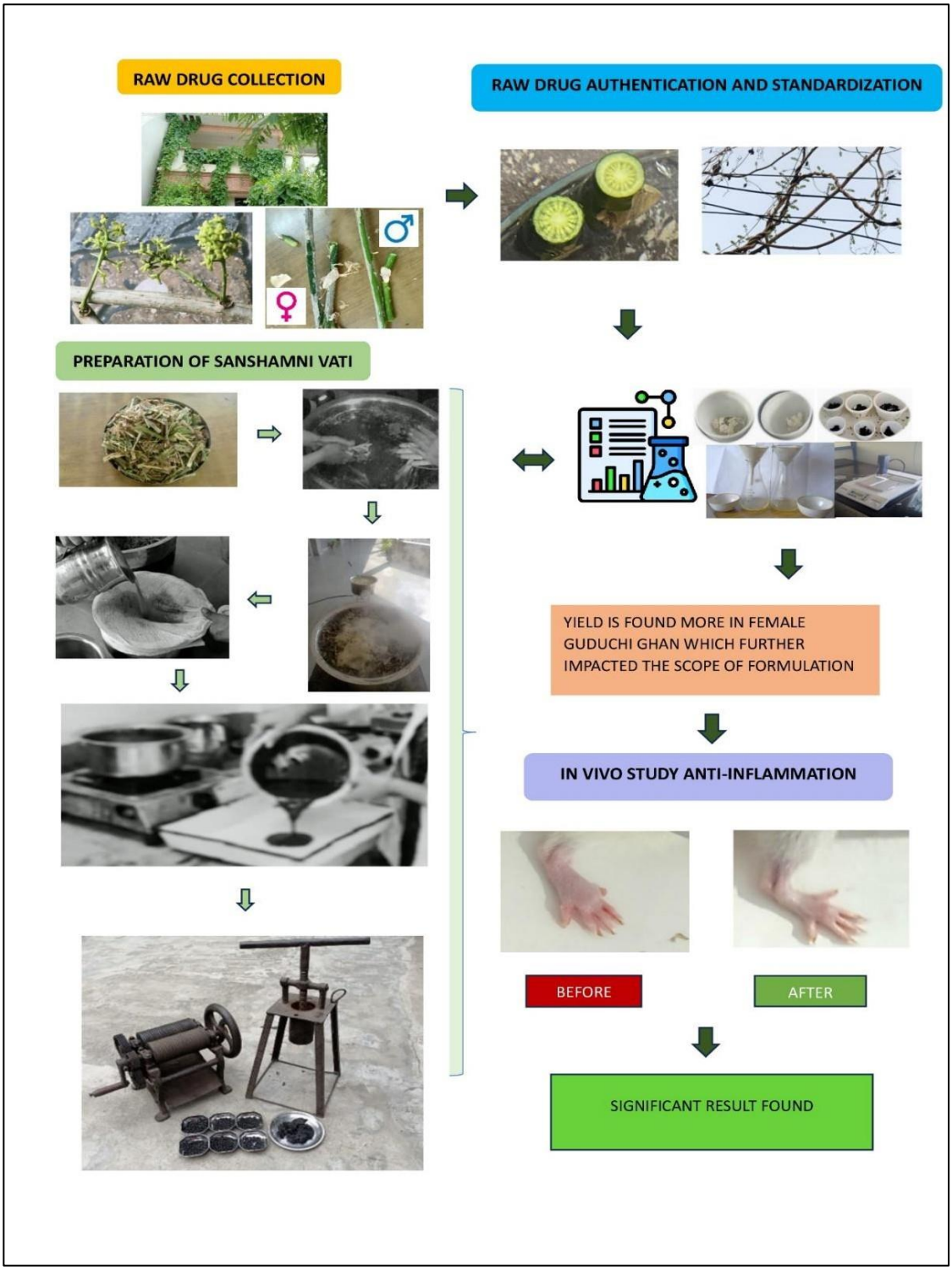

IMPACT OF DIOECISM ON QUALITY AND EFFICACY OF SANSHAMANI VATI

CHAPTER -1
INTRODUCTION

Medical encyclopedias of Ayurveda de-lineated customize several therapeutic formulations for future generations, for a holistic approach to wellness and science. Medicinal plants are utilized as raw materials for therapeutic formulations. Accurate identification of these plants solely through ancient literature and classical texts has become challenging due to the presence of various synonyms and vernacular names that lead to misidentification of various herbs. Therefore, 'there is a need for scientific investigations into various medicinal properties related to morphological characteristics of these plants. Correlating these findings with information from Ayurvedic classics is crucial to ensure accurate identification for efficacy in Ayurvedic preparations of contemporary era.

The production of primary and secondary metabolites varies according to growth of plants. The efficacy of plant-based formulations is directly related to the concentration of active pharmaceutical ingredients present in the processed plants or herbs. The biosynthesis of secondary metabolites in plants depends on the environment, growth, function, and activities of the individual part of the plant. (Fernandez et al., 2016) These factors will also affect the number of secondary metabolites synthesized throughout the season and also in the developing stage of the plants. Generally, the collection of a specific part of the plant depends on the season and also its optimum state of development. Just 6% of plant species are dioecious, or have separate male and female parts, whereas the majority, or 96%, are angiosperms, or blooming plants. By comparison, 37% of conifer species belong to the 50% of gymnosperms (non-flowering seed plants) that are dioecious. Moreover, only 7% of the total taxa have been known for the phenomenon of dioecy. (Kumar, Researcher and Khatoon, 2020) (Choudhry et al., 2014)

Dioecy is very rare in plants and widely distributed in various orders and families. Direct references for the dioecy or its importance is not available in the classical texts of Ayurveda. But, the nomenclature of individual plants indicating their gender either masculine or feminine; might be a selection criterion for the collection of best quality plants. Gender can also be considered as one of the factors which influence the quality

of plants along with other morphological features. Dioeceous plants, identified with separate male and female reproductive parts and surrounding somatic tissue that modulates the general properties of plants. It is considered an important factor for morphological identification. However, the effect of gender is not predominantly acknowledged.

Presently, around twenty-five plants are being used in Ayurveda including *Amrita*, *Pippali* and *Vijaya*, popularly identified based on dioecism. (Choudhry et al., 2014). Environmental variables and differences in growth rate modulate secondary metabolite synthesis frequently leading to gender-specific responses in these plants. These reactions are best shown by the distinguished dioecious plant *Guduchi* (Tinospora cordifolia) (Thumb.) Miers, a member of Menispermaceae family. Its synonym Amrita directing its application for revitalization. It is considered as one of the important *Rasayana* drugs in Ayurveda. (Health and Welfare) Various researches published based on their chemical constituents, therapeutic properties, and validations are well-known for the broad spectrum of its biological actions including immunomodulatory, hepatoprotective, apoptogenic, anti-inflammatory, anticancerous, antiallergic, antioxidant, antidiabetic, and diuretic qualities. (Upadhyay et al., 2010)

Moreover, *Guduchi,* it is also found effective as an adjuvant for chemotherapy in cancer. (Malabadi et al., 2024) It incorporates alkaloids, flavonoids, cardiac glycosides, saponins, lignans, steroids, terpenoids, tannins, as chief chemical constituents. (Kirti Sinha, N P Mishra, 2015) (Sharma and Pradesh, 2023) Berberine is one of the most important chemical constituents that belongs to the Isoquinoline alkaloid, reported for its anticancer, immunomodulatory, and antidiabetic property. (Ghosh and Saha, 2012) Moreover, Tinosporaside is also reported to have antihyperglycemic activity (Choudhry et al., 2014) *Guduchi Satva* and *Sanshamni Vati*, well known Ayurvedic formulations of *Guduchi (Tinospora cordifolia)* are reported to have immunomodulator, antipyretic and antimetastatic activity. (Kirti Sinha, N P Mishra, 2015). Clinicians have recently been interested in this magical herb due to its immunomodulatory property. The Indian Ayurvedic Pharmacopoeia suggests the use of *Guduchi* (internal and external) to treat a variety of illnesses.

Numerous classical Ayurvedic formulations have been effectively utilized by Ayurvedic physicians for the treatment of various ailments. Among these, *Sanshamani Vati* is a well-

known and widely prescribed medicine. It is used in the management of several conditions such as *Jvara* (fever), burning sensation (*Daha*), *Daurbalya* (general weakness), and *Pandu* (anemia), (inflammation) shotha, depletion (Kshaya), urinary disorders (*Mutraroga*), and semen loss (*Viryastrava*). It was also extensively employed during the COVID-19 pandemic for both preventive and therapeutic purposes. Due to its proven efficacy. The efficacy of the potential of plant-based formulations depends upon the concentration of effective constituents of the ingredients. Gender may be an important factor to impact formulations. *Guduchi's* chemical composition and biological properties have always been the subject for its use in various ailments without considering dioecism. *Sanshamani Vati* was first introduced by Acharya Yadavji Trikamji in the 20th century through his renowned text *Siddhayoga Sangraha* due to its primary ingredient in *Guduchi* (*Tinospora cordifolia* Miers.), acclaimed in Ayurveda as an excellent *Sanshamana Dravya,* recognizing this unique therapeutic property(Vyas, 2023). Concentrated extract of Guduchi was named as sanshamni vati by Acharya Yadavji Trikamji. Later on, Bheshaja Samhita, referencing Rasoddhara Tantra *and* vaidhyaka chikitsa sara and *d*escribed four variants of *Sanshamani Vati*: No. 01, No. 02, No. 03, and *Special*. *Sanshamani Vati. respectively. Rasatantrasara evam Siddha Prayoga Samgraha* cites the same *Sanshamani Vati (Special)* formula from *Vaidhyaka Chikitsa Sara*, with a slight modification—*Svarna Makshika Bhasma* (6 Masha) is included, while *Pravala Bhasma* is omitted(Vyas, 2023).

The Ayurvedic Formulary of India adopts this from *Siddhayoga Sangraha*, prescribing it for fever, chronic fever, tuberculosis, weakness, anemia, and irregular fevers in a dose of 250–500 mg. Hence, the present study has been designed to determine the impact of dioecism in the phytoconstituents of *Sanshamni Vati* and its anti-inflammatory activity. *Sanshamani Vati* remains a frequently recommended remedy by Ayurvedic practitioners.

Chapter 2
Literature Review

2.1 Vati Kalpna

2.1.1 Definition of Vati

In Ayurveda, "*Vati*" is a formulation prepared in the form of tablets or pills. It is a common dosage form used for administering herbal medicines. The word Vati literally means "pill" or "tablet" in Sanskrit.

2.1.2 Synonyms of *Vati*

Gutika, Vati, Vatika, Modak, Pindi, Guda and Varti.

2.1.3 Types of *vati Kalpana*

2.1.3.1 Sa-Agni Vati

Vati is crafted using heat. The process involves preparing sugar, jaggery, or *Guggulu* into a semi-solid *Leha* by gently heating them. Once the *Leha* reaches the right consistency, powdered ingredients are mixed in. The mixture is then shaped into circular tablets, or *Vati*, by rolling.

2.1.3.2 Nir-Agni Vati

Vati is made without the use of heat. Instead, powdered ingredients are combined with *Guggulu*, *Guda* (jaggery), or another recommended liquid. This mixture is then thoroughly blended, or triturated, to form the *Vati* into its final shape (Article, 2016).

2.2 Ghana

Ghana is a concentrated form of the liquid material from plants, such as *Swarasa* and decoctions. It is essentially a dried aqueous extract, where the liquid content of *'Kwatha*' is evaporated using heat. In Ayurvedic literature, *Ras kriya* is considered synonymous with *Ghana*, *Phanita*, and *Avaleha*, due to their similar preparation methods with only minor differences. *Avaleha* is a semi-solid that can be licked, while *Ghana* is in a solid form.

In Ayurveda, "*Ghana*" generally refers to a concept or substance that is dense, thick, or solid. The term is often used in various contexts, such as describing certain physical properties of substances or the form in which herbal medicines are prepared.

1. Substance Density *Ghana* can describe the density or thickness of a substance. For instance, herbal extracts that are concentrated and thick are sometimes referred to as

"*Ghana*".

2. *Ghana Satva*: This refers to a concentrated, solid extract derived from plants. For example, "*Ghana Satva*" is used in preparing certain Ayurvedic medicines where the active ingredients are extracted and condensed into a dense form, often used in the form of tablets or pills.

3. *Ghana Vati*: This is a term for tablets or pills made using the concentrated extracts (*Ghana*) of herbs. These are commonly used in Ayurvedic practice for their potent therapeutic effects.

4. Qualitative Description: *Ghana* can also be used to describe the quality of being dense, heavy, or solid in a more general sense. It is sometimes used in describing bodily tissues or substances within the body.

Various references are available in *Astanga Sangraha*, *Bhavaprakash*, Ayurveda *Prakash* and *Sharangdhara Samhita* which can be correlated with *Ghana Kalpana*.

Table 2.1 Description of Ghana available in *Sangraha Kala*

Classical text	Description
Astanga Sangraha	Rasakriya is one of the type of anjana kalpana.
Bhavaprakash	Description about rasanjana is available. Decoction of darvi added with an equal quantity of milk and heated till it becomes thick termed as rasakriya.
Ayurveda Prakash	The method of preparation of rasanjana with aja milk
Sharangadhara Samhita	Examples of rasakriya under anjana kalpana is mentioned i.e., davryadi rasakriya, babul rasakariya.

2.2.2 *Adhunik Kala* (*Sidha Yoga Samgraha*):

Guduchi Ghana Vati is mentioned by *Acharya Yadavaji Trikamaji* as Sanshamni Vati in *Jwaradhikar.*

2.2.3 Definition of *Ghana*

When *Swarasa* and *Kwatha* from plant parts are further heated to concentrate them, this process is known as *Rasakriya*. "Aqueous solutions of plants, such as *Swarasa* and *Kwatha*, when concentrated using heat to achieve a semi-solid form, are referred to as *Ghana*."

2.2.4 Synonyms of *Ghana*

While there are no synonyms for *Ghana*, the common pharmaceutical procedure *Sharangdhara* applies to all these terms, each having its own specific meaning and therapeutic value. *Avaleha* and khanda are concentrated liquid forms with sweet substances, whereas *Ghana* and *Raskriya* are concentrated forms of aqueous solutions without sweet substances.

2.2.5 Advantages of *Ghana*

Ghana or concentrated herbal extracts in Ayurveda, offers several advantages:

Potency: *Ghana* extracts are more potent than raw herbs or simple decoctions because they are concentrated, providing stronger therapeutic effects.

Convenience: The solid form of *Ghana* makes it easier to store, transport, and consume compared to liquid decoctions or semi-solid forms like *Avaleha*.

Standardization: *Ghana* allows for more consistent dosing and standardization of herbal medicines, ensuring uniform therapeutic benefits.

Shelf Life: The drying process used to create *Ghana* reduces moisture content, which helps in preserving the extract for longer periods, extending its shelf life compared to liquid preparations.

Ease of Use: *Ghana* can be conveniently formulated into tablets or capsules, making it user- friendly and increasing patient compliance.

Reduced Dosage Volume: Since *Ghana* is a concentrated form, a smaller quantity is required to achieve the desired therapeutic effect, making it easier for patients to take.

Enhanced Absorption: The concentration process may enhance the bioavailability of certain active constituents, improving their absorption and efficacy in the body.

Customization: *Ghana* can be combined with other herbal extracts or formulations to tailor treatments to specific health conditions and individual patient needs. (Bhatt, Deshpande and Chaskar).

2.3 *Ghana Vati Kalpana*

Ghana Vati is a type of pill created by compressing *Ghana*, a semi-solid extract. Traditional texts reference different formulations of *Ghana Vati*, including *Sarpagandha Ghana Vati* and *Guduchi Ghana Vati*. The *Charaka Samhita* specifically notes that *Vati* is made from a semi-solid state. The pill form of the medicines is a convenient form for the patient as well as a physician in treatment. *Vati* is made in the shape of flat circular mass hence it is similar to the pills. [Reference: *Agniveshacharya*, *Charak Samhita*, elaborated by *Charak* and *Drudhabala* edited by *Priyavrat Sharama Chikistha Sthana,* 26/206. Chaukhambha Orientalia, Varanasi 2007;]

2.3.1 Dose of *Ghana Vati*

Classical texts do not specify a clear dosage foer *Ghana*. However, the *Siddha Yoga Sangraha* provides guidance for *Sanshamni Vati*, recommending a dosage of 5 to 10 Vati, each weighing 2 Ratti (approximately 250-500 mg), taken four to five times daily. This translates to a total daily dose of *Ghana* ranging from 5 to 10 grams.

(API Part -2 vol-1) Some formulations of *Ghana Vati* with references:

Table no.2.2: Examples of Ghana Vati with reference

S. No.	Formulation	Reference
1.	*Kutaja Ghana Vati*	*Siddhayogasangraha, atisara-pravahika- grahanyadhikara*
2.	*Sarpagandha Ghana Vati*	*Siddhayogasangraha,bhrama-anidra-unmadadhikara*
3	*Sanshamni Vati* (*Guduchi Ghana Vati*)	*Siddayogasangraha, jvaradhikara*

2.4 Sanshamni Vati

In Ayurvedic texts, *Sanshamni Vati* is identified as a safe and effective formulation. The references for *Sanshamni Vati* can be found in the *Siddhayoga Sangraha* and *Rasoddhara Tantra*. According to Yadavji Trikamji Acharya, *Guduchi Ghana Vati* is synonymous with *Sanshamni Vati*. However, the *Rasoddhara Tantra* suggests that while the ingredients of *Sanshamni Vati* differ, its therapeutic indications remain consistent.

2.4.1 *Review of Literature Guduchi Ghana Vati*

The available literary sources indicate

1. In the *Siddhayoga Sangraha* by Yadavji Trikamji Acharya, *Guduchi Ghana* is identified as Sanshamni Vati.
2. According to the Rasoddhara Tantra, the constituents of Sanshamni Vati include *Guduchi*, Ativisha, Pippali, and Loha Bhasma.

2.5 Guduchi

Botanical Name: Tinospora cordifolia (willd.) Miers

Family: Menispermaceae

Guduchi (*Tinospora cordifolia*) is also called an Amrita in traditional Ayurvedic text. It is one of the most often utilized medications in Ayurvedic pharmacopoeias. Recently clinicians have been interested in this medication due to its immunomodulatory properties. The Indian Ayurvedic Pharmacopoeia suggests using *Guduchi* both internally and externally to treat a variety of illnesses.

2.5.1 Properties

Rasa: Tikta, kasaya

Guna: Laghu

Virya: Usna Vipaka: Madhura

Dosh Karma: Tridoshshamak, Samgrahi, Balya, Dipana, Rasayana ,Raktashodka, jwara Ghana.

2.5.2 History

Focusing on the past provides a blueprint for growth across all aspects of life. This historical perspective, particularly regarding the quality and uniqueness of drugs, is invaluable, as it helps guide future generations in making more informed decisions.

2.5.3 Vedic kala

Sayan noted that *Giloy* is traditionally kept in every household to ward off snakes and scorpions, as referenced in the (Sounakiya Atharvaveda 6/56) and (Panini Upadhi Bhojavrtta-2/2/80). In the *Kaushika Sutra*, *Giloy* is referred to by the name *'Kudruchi'*

2.5.4 Mythological review

Acharya Bhavamishra was the first to share the mythological significance of *Giloy* in his text *Bhavaprakasha Nighantu*. According to legend, *Ravana*, driven by desire, kidnapped *Sita*, the wife of *Rama*, sparking a fierce war between *Rama* and *Ravana*. During the battle, many monkey warriors who supported *Rama* lost their lives. As a gesture of victory and respect, Lord *Indra* showered elixir upon the fallen monkey warriors, granting them rebirth. Wherever the elixir fell on the ground during this process, *Giloy* plants are said to have originated.

2.5.4.1 *Guduchi* in Samhitas

Charaka Samhita (1000B.C to 4th Century A.D)

According to *Acharya Charaka*, a substance, or "Dravya," can be recognized through its name, either in Sanskrit or vernacular languages, as well as by its morphological characteristics or properties, a concept known as "*Namarupaguna Jnana*." "*Rupa*" pertains to the external morphological features, primarily relying on vernacular names and synonyms.

Sushruta Samhita (c. 1000 BC to c. 5 AD)

In Sushrut Giloy can be traced be traced in 41 locations and under 9 Ganas in this description. Additionally, it is grouped with lesser plants like Vallipanchmula. Giloy ’s multiple medicinal uses have led to its inclusion in several Ganas or groupings.

Astanga Hridya (5th century A.D)

Giloy is referenced either alone or combining with other remedial herbs for the treating of Jwara, Shleepada,Prameha, and other conditions.

2.5.4.2 *Guduchi* in *Nighantus*

- ***In Kala of Nighantu***

The majority of *Nighantu* has extensive documentation on *Giloy*, suggesting that it is a widely accessible and often used plant. There were also descriptions of other synonyms that showed its main morphological characteristics. Only its varieties and a range of medicinal effects were recorded during this time.

- ***Dhanvantari Nighantu***

Giloy has been first stated in one out of seven *Vargas*. Two varieties known as *Giloy* and *Kanda Giloy* are defined with different vehicle in congruence of *Doshas* i.e. *Vata, Pitta, Kapha*. this *Nighantu* listed 34 synonyms of *Giloy*.

- ***Kaidev Nighantu****Kaidev Nighantu*: *Ausadha Varga* mentions *Giloy* and describes both *Pinda Giloy* and *Giloy*, along with 19 synonyms.

Table No. 2.3: Description in Samhitas with references

Samhita	Description	References
Charaka Samhita	*Sandhaniya*	Cha.Sut. 4/5
	Tripti-ghna	Cha.Sut 4/11
	Stanya-shodhana	Cha.Sut 4/18
	Sneha-paga	Cha.Sut 4/21
	Trishnana-nigrahana	Cha.Sut 4/29
	Daha-prashamana	Cha.Sut 4/41
	Vaya Sthapana	Cha.Sut 4/50
	Tikta skandhas	Cha.Sut 8/143
	Sirovirechana	Cha.Sut 8/151
	Madhura skanda	Cha.Sut 8/139
Sushruta Samhita	*Vatasan-shaman -Gana*	Sus.Su.37/7
	Pitta-sanshamana -Gana	Sus.Su.37/8
	Shlesham-sanshaman -Gana	Sus.Su.37/9
	Shodhana -Varga	Sus.Su.37/12
	Ropana -Gana	Sus.Su.37/24
	Aragvadhadi	Sus.Su.38/29
	Syamadi	Sus.Su.37/7
	Patoladi	Sus.Su.38/33
	Kakolyadi	Sus.Su.38/35
	Guduchyadi	Sus.Su.38/50
	Valli- panchmula	Sus.Su.38/73
	Shaka -Varg	Sus.Su.46/270
Astanga Sangraha	*Shaka -Varg*	Ast.H.Su 6/77
	Aragvadhadi -Gana	Ast.H.Su 15/18
	Shyamadi -Gana	Ast.H.Su 15/45
	Padmkadi	Ast.H.Su 15/12
	Patoladi	Ast.H.Su 15/15
	Guduchyadi -Gana	Ast.H.Su 15/16

- ***Bhavaprakash Nighantu***

Giloy 's mythical origins and 21 synonyms have been explained under *Guduchyadi Varga.*

- ***Raj Nighantu***

A description of the two varieties of *Giloy* and *Kanda Giloy*, together with a list of their 31 synonyms and medicinal use, is provided. In addition to its use as a drug, the leaves of *Guduchi* (*Tinospora cordifolia*) are also described under *Shaka Varga* (group of vegetables) in classical Ayurvedic texts such as *Brihatrayee*, *Bhavaprakasha*, and *Gunaratnamala*. These texts highlight that the leaves of *Guduchi* possess similar properties as the stem, which is commonly used for its medicinal benefits.

- ***Shaligram Nighantu***

Guduchyadi Gana provides nine synonyms for *Giloy* and six for *Kanda Giloy* in its description of *Giloy*.

- ***Shabd Chandrika***
- In Shabd Chandrika, Guduchi is mentioned in *Vrikshadi Varga*; one out of 09 *Varga*. *Aacharya* have described about its 09 Synonyms with properties and therapeutic uses. (***Shabd Chandrika-*** 1**. vriksha adi varg** /122,123,124)

- ***Saraswata Nighantu***

In *Saraswata Nighantu*,Guduchi is mentioned in Lata Varga; one out of 06 Varga. Aacharya have described about its 18 Synonyms.

- ***Siddha Sara Nighantu***

In Siddha Sara Nighantu,Guduchi is mentioned with about its 07 Synonyms

- ***Raja Vallabha Nighantu***

In Raja Vallabha Nighantu,Guduchi is mentioned in Aushadhashrya Parichheda; one out of 06 Parichheda. Aacharya have described about its properties and therapeutic uses.

In ancient times, medicinal plants were typically described using a variety of synonyms that reflected their physical attributes, properties, effects, habitat, medicinal uses and unique traits. Therefore, understanding these synonyms is crucial for accurately identifying plants in today's botanical context.

Table No. 2.4: Description of *Guduchi* in *Nighantus*

Nighantu	***Varga***
DhanvantariNighantu	*Guduchyadi varga*
BhavaprakashaNighantu	
Raja Nighantu	
Shaligrama Nighantu	
Shodhala Nighantu	
Nighantu Adarsh	
Kaiyadeva Nighantu	*Aushadiya Varg*
Priya Nighantu	*Pippalyadi Varga*

2.5.5 Synonymsof *Guduchi*

In ancient times, medicinal plants were typically referred to by various names and synonyms, reflecting the diverse languages, cultures, and traditions in which they were utilized. These synonyms serve as important historical references and provide insight into the widespread use and recognition of plants across different regions and civilizations.

2.5.5.1 Nirukti of synonyms (Ninama et al., 2022)

Synonym of *Nirukti* and its meaning

- *Amrita valli*: the creeper of the plant never dies.
- *Chinnruha*: When cut, *Giloy* grows again.
- *Kundali*: The *Giloy* plant spirals around its support as it grows upward.
- *Chkralkakshna*: When the stem of is cut across, it reveals a circular pattern.
- *Jwarnashin: Very* efficacious drug fever.
- *Amrita*: *Giloy* possesses qualities like that of nectar.
- *Jivanti: Giloy* protects life due to its *Rasayana* property.
- *Madhuparni*: When the leaves are crushed, they produce sticky juice similar to honey.
- *Tantrika*: The stem of the *Giloy* plant has a rope-like appearance.
- *Mandali*: The *Giloy* plant ascends its supporting plant in a circular, spiraling fashion
- *Vishalya: Giloy* protects by removing toxins
- *Rasayani: Giloy* has rejuvenating properties
- *Chandrahasa:* The seeds are semilunar in shape.
- *Kandoddhava: Giloy* can be propagated with stem.

2.5.6 Distribution

- Endemic to India throughout tropical and sub-tropical zones at an altitude of about 600m. & throughout sub-tropical regions at 1200 m above sea-level
- Dry districts, at low elevations, Deciduous and dry forests. Range- East-Asia, India, Sri- Lanka, Myanmar

2.5.7 Chemical Constituents

- *Terpenoids:*Tinosporide,Diterpene,Tinocordioside,Cordioside,sesqiterpene, tinocordiofolioside a,b,c,d,e, Terpenoside, Furanoid, Diterene etc
- *Alkaloids*: Tinosporine, magnoflorine, Berberine, Palmitine, Tembetarine, Choline, Jatorrhizine, isocolumbine, Tetra Hydropalmatine etc.
- *Steroids*: Gilonsterol, β-sitosterol etc.
- *Others*: Giloin, Tinosporine acetate, Tinosporidine,Hepatocosanol,
- Arbinogalactan are immunologically (Singh et al., 2003)

2.5.8 Botanical Characteristics

It's a large, smooth Vining Shrub that often reaches significant heights, producing long, thread-like aerial roots. The fresh or tender stems are greenish, have a smooth surface, and swell at the nodes. The branches have smooth, heart-shaped leaves. There are longitudinal clefts with spaces in between, typically dotted with large rosette patterns and lenticels.

2.5.8.1 **Stem**

The fresh or tender stems are greenish with smooth surface and swelling at nodes. Branches bear smooth heart shaped leaves. Longitudinal clefts, the space between the cleft being usually dotted with large rosette lines and lentils.

2.5.8.2 Leaf

The leaves are simple, alternate, and lack stipules. They have relatively long petioles and emerge from short, swollen nodal projections on the stem. These leaves are short-

lived and fall off soon after sprouting. The petiole is slender, rounded, and measures 2.5 to 7.5 cm in length. Its base is thickened (pulvinate) for a short distance, with this swollen section exhibiting a slight twist. There is also a similar swelling, typically not twisted, at the top or distal end of the petiole. The blade is broadly ovate to round-cordate, measuring 5 to 10 cm in diameter. It is thin, entire, and quite smooth on both surfaces, with a slightly glaucous underside. The base features a wide, deep depression (sinus), while the tip is acute or short and sharply pointed (acuminate) and having five to seven nerves.

2.5.8.3 Floral Arrangement

The plants are dioecious, meaning that the female and male flowers grow on different individuals. The stamin ate inflorescences are typically drooping or pendulous, the flowers are the longer than leaf inflorescences are grouped in clusters of two to six. The pistillate inflorescences are generally shorter than the leaves, with flowers mostly borne singly but densely packed along the rachis.

2.5.8.3.1 Flowers

The flowers are small, unisexual, numerous, and shed early. They are precocious, bracteate, greenish-yellow, and delicate with tiny, delicate pedicels. The lower pedicels are occasionally with some leafy structures. There are six sepals, which are free and deciduous, placed in two series, the inner three being bigger and membranous and the outer three being tiny, ovate-oblong, and pointy.

- Stamen ate flowers have six stamens with free, spreading filaments that are slightly longer than an enclosed by the petals. The anthers are oblong, connivant, and embedded in the thickened fleshy tip of the filament, dehiscing longitudinally by an oblique, nearly marginal slit.
- Pistillate flowers Pistillate flowers possess six vestigial staminodes alongside the gynoecium. The gynoecium consists of three free carpels, with the ovaries positioned on a swollen prominence of the receptacle. The styles are very short and simple, and the stigma is expanded and either forked or deeply lobed.

2.5.8.4 Fruit

The fruit consists of one to three sessile drupelets, aggregated together, though it is rare for all three carpels to ripen. When ripe, these drupelets are smooth, bright red or scarlet, roundish to somewhat ovoid or oblong, and approximately the size of a large

pea. They are apiculate, having a convex dorsal side and a flat ventral side, with the styles located sub-basally and resting on a swollen projection of the receptacle. The mesocarp is very sticky and pulpy. The endocarp is dorsally keeled, ventrally concave, and smooth or somewhat rough due to a few isolated tubercles.

2.5.8.5 Seed

Each fruitlet contains a single seed, which is meniscus- and kidney-shaped, with a deep ventral groove curving around a slightly two-lobed intrusion of the endocarp. The seed is endospermic, with the endosperm ruminate on the ventral side. The embryo is slightly curved, with ovate, spreading or divaricated, leaf-like cotyledons.

2.5.9 *Flowering and Fruiting season*

Flowering: May- June

Fruiting: September- October

2.5.10 Chief advantages of the plant:

- Plant itself can grow in different environmental and soil conditions.
- Plant itself is a weed and survive in most of the environmental conditions.
- Cultivation practices is not required.
- Grazing animals does not consume this plant.

2.5.11 Formulations of *Guduchi*

1. ***Churna:*** *Rasayan churna, Sudarshan churna*
2. ***Kwatha:*** *Guduchyadi kwath,Manjisthadi kwath, Punarnavastaka kwatha etc.*
3. ***Arista:*** *Amritarishta*
4. ***Ghrita:*** *Giloy ghrita, Amritadi ghrita, Panchatikta ghrita*
5. ***Taila:*** *Guduchyadi taila*
6. ***Vati:*** *Sanshamni vati, Chandraprabha vati*
7. ***Lauha:*** *Guduchyadi lauha*
8. ***Rasa- aushadhi:*** *Gandhak rasayan, Chandrakala rasa*

Table No. 2.5: Macroscopic Characteristics of *Guduchi*

Habitat	Dry districts, at low elevations, deciduous and dry forests
Stem	Terete, sparcel tentaculate. The young stem has smooth surfaces, a green color, and swollen-looking nodes. The stem becomes somewhat protruding as it ages and takes on a light brown appearance dotted with round lenticels.
Leaves	Leaves are membranous, 7-9 veined, typically 5-10cm in size (rarely up to 12 by 10cm), rounded to subdeltoid, cordate with a broad sinus and prominent basal lobes. They are obtuse or slightly cuspidate, with a reticulate veining pattern. Petioles measure 2.5 to 7 cm long. Racemes are relatively loose, starting around 5cm in length and extending, found in axillary, terminal, or older wood location.
Floral Characteristics	Male flowers are clustered in the axils of small subulate bracts. Female flowers are typically solitary, resembling male flowers but with green sepals and unreflexed margins; staminodes are short and linear.
Calyx	Sepals consist of three outer ones that are very small, ovate-oblong and acute, and three inner ones that are larger, membranous, broadly elliptical, concave, and yellowish, measuring 3 to 4 mm.
Corolla	There are six equal petals, each about 2 mm long, broadly spathulate in shape. When young, each petal loosely surrounds a stamen, with a cuneate claw and a lamina that is triquetrous or somewhat trilobed, reflexed at the apex.
Gynoecium	The carpels number 1-3, widely spaced on a short, flexible gynophore. They are dorsally convex and ventrally flat or nearly so, scarlet in colour and approximately the size of a large pea. The style scar is subterminal. The stone is broadly ellipsoid, with a slender dorsal ridge.

Fruits	Fleshy, single seeded
Seeds	Curved
Root	Slender, fleshy, rooting pedicels are sent down by aerial branches.

2.5.12 Microscopy of *Guduchi* Stem

T.S. Of stem shows 2-3 cells of cork followed by 2-3 layers of collenchymatous cortex and 4-6 layers of parenchymatous cortex consisting of circular to isodiametric type of cells and below lenticels, group of sclereids present in secondary cortex, cortex cells are filled with plenty of starch grains which are simple irregularly ovoid-elliptical pericyclic fibers are lignified with wide lumen and pointed ends associated with large number of crystal fibers containing a single prism in each chamber. Vascular zone is composed of 10-12 wedge shaped strips of Xylem externally surrounded by semicircular strips of Phloem, alternating with wide medullary rays. Phloem consists of sieve tube companion cells and Phloem parenchyma contains calcium oxalate crystals. Xylem consist of vessels, tracheids, parenchyma and fibers. Secondary Xylem elements are thick walled and lignified. Vessels cylindrical with pits on walls. Large vessels possess several tylosis with transverse septa. Medullary rays 15-20 or more cells wide containing rounded hemispherical, oblong, ovoid starch grains with faintly marked concentric striations and central hilum. Pith composed of large thin-walled cells with starch grains. (API, Part-1, volume-1, first addition)

Some of the microscopy data has also been sourced from the Research papers.

The transverse section of *Guduchi* stem, have a distinctive wheel-like appearance, which is the main characteristic of the Menispermaceae family. The section shows radial lines of medullary rays alternating with vascular strands, topped with sclerenchyma cells. The epidermis, covered by a thick cuticle, forms a single outer layer. Beneath it lies a collagenous hypodermis. The inner cortex in the notch region is parenchymatous, while the middle cortex is composed of chlorenchyma. The pericycle is prominent, continuous, and sclerenchyma Tous, forming wavy rings with distinct ridges and furrows. Multiple vascular bundles, either closed or open, bicollateral, and

ring-shaped, are present (Khatoon et al., 2018). The well-developed phloem consists of companion cells, parenchyma, and sieve tubes. The xylem comprises vessels, tracheid's, fibres, and parenchyma, with cambium located in the outer regions. Most parenchymatous cells contain embedded starch grains of varying shapes and sizes. Additionally, the hypodermal and cortical regions feature mucilage canals (Pandey, Sourabh and Gautam, 2021).

2.5.13 Different types of phytochemicals that are found in different parts of *Guduchi*

Alkaloids

- Berberine, Palmatine: Found in the stem

Glycosides

- 18-norclerodane glucoside, Furanoid diterpene glucoside, Tinocordiside, Tinocordifolioside: Found in the stem
- Cordioside, Cordifolioside A, Cordifolioside B, Syringin, Syringin-apiosylglycoside, Palmatosides C, Palmatosides F, Cordifoliside A, Cordifoliside B, Cordifoliside C, Cordifoliside D, Cordifoliside E: Found in the stem

Diterpenoid Lactones

- Furanolactone, Clerodane deriVatives: Found in the all parts of plant
- [(5R,10R)-4R-8R-dihydroxy-2S-3R:15,16-diepoxy-cleroda-13(16),14-dieno-17,12S:18,1S-dilactone], Tinosporon, Tinosporides, Jateorine, Columbin: Found in the whole plant

Steroids

- β-sitosterol, δ-sitosterol, 20 β-Hydroxy ecdysone, Ecdysterone, Makisterone A, Giloinsterol: Found in the aerial part and stem

Sesquiterpenoid

- Tinocordifolin: Found in the stem

Aliphatic Compounds

- Octacosanol, Heptacosanol: Found in the whole plant

Other Rare Compounds

- Nonacosan-15-one: Found in the whole plant.

- 3(α,4-di-hydroxy-3-methoxy-benzyl)-4-(4-hydroxy-3-methoxy-benzyl)-
- tetrahydrofuran: Found in the all plant parts.
- Jatrorrhizine, Tinosporidine, Cordifol, Cordifelone, N-trans-feruloyl tyramine as diacetate, Giloin, Giloinin, Tinosporic acid: Found in the whole plant. (Sharma et al., 2019)

2.5.14 Reported Pharmacological Activity of Stem of *Guduchi*

Anti-inflammatory and Immunomodulatory Activities

A 2020 study emphasized the strong anti-inflammatory and immunomodulatory properties of *Tinospora cordifolia* stem extracts. The research demonstrated that these extracts significantly decreased the levels of pro-inflammatory cytokines, indicating their potential effectiveness in treating inflammatory conditions (Mathew et al., 2020)

T. cordifolia alcoholic extract exhibits anti-inflammatory effects in both acute and subacute inflammation models. When grown on neem (Azadirachta indica), it significantly reduces acute inflammation at a dose of 50mg/100g administered both orally and intraperitoneally. The dried stem of *T. cordifolia* is effective in treating both acute and subacute inflammation, surpassing acetylsalicylic acid in acute inflammation, although it is less effective than phenylbutazone in subacute inflammation. (Roy et al., 2020)

A subsequent study in 2021 supported these findings, featuring the role of *Tinospora cordifolia* in enhancing the immune response. This research demonstrated that *T. cordifolia* boosts both innate and adaptive immunity, further emphasizing its immunomodulatory capabilities (Singh et al., 2021) *Guduchi* stem is abundant in alkaloids, glycosides, diterpenoid lactones, sesquiterpenoids, and phenols. These bioactive compounds are responsible for its significant anti-inflammatory and antioxidant properties, which help mitigate oxidative stress and inflammation, providing protection against various diseases (Singh et al., 2003; Sreenivasa Reddy et al., 2009).

Research conducted in 2022 revealed that the stem extracts of *Tinospora cordifolia* exhibit strong antioxidant properties. The study demonstrated that these extracts effectively scavenge free radicals, indicating their potential in managing oxidative stress-related disorders (Kumar et al., 2022).

Table No. 2.6: Pharmacodynamics (action) and Therapeutic indication (uses) of *Guduchi* (Upadhyay and Kumar, 2010).

Classical reference	**Pharmacodynamics Action**	**Therapeutic indication**
Bhav Parkash Nighantu,(Yadav, 2017) Guduchyadivarg: 8-10	Rasayan,Sangrahi,Balya, Agnideepan,Tridoshshamak	Daha,Kash,Kamla,Kusth,Pandu, Vatrakta,jwar Krimi prameh, Meha, Swas, Arash,Hridyarora
Astang Sangreha{10} Sushrut Sathan7-149 ,16-10	Vata-pita-kaphanashak,trishnanashak, Agnideepaka	Jwar ,chardi, daha
Charak Samhita {11} Sutrasathan25-40	Sangrahi, vatahara agnideepana,shleshma-shonit-prashmana	vivanda
Raj Nighantu Guduchyadi varga17-18	Tridoshnashak,vishghani,jwa r-bhootghani	Jwar daha,Trishna,vatarakta,prameha, pandu bhrama,balipalitya
ArkParkash,Trity ashatak	Dipna, Grahi	Kash, Pandu Jwar
Sidh Bhasijya ManiMala Dvitya Gucch70	Balya Tridoshnashak	Balya Tridoshnashak Laghu jwara, Meha,Daha .Kasa,Pandu, Visarna
Sodhal Nighantu Guna Sangreha,Guduch aydi varg-120	Tridoshghani, Grahi ,Rasayan,Dipna	Jwar,Daha,Kamla,Vatarakta
Madan pai Nighantu HartikyadiVarga-39,40,41	Sangrahi ,Balya, Agni Deepan	Kamla,kustha,Vatarakta ,jwara,Pitta,Vivandha, Krimi
Kaidev Nighantu , Aushadhi Varg 9,10,11	Sangrahi .Vrishya. Balya,Rasyana,Dipna, chakchsya,vaya sathapan, Medhya,Tridosh nashak	Kusth, Krimi, Chardi,Daha,Vatarakta, ,Pandu ,Jwar, Kamla, Meha, Trishna ,Kasa
Dhanvantri Nighantu Guduchyadi varg 5,6,7,8	Tridoshnashak , Ayushyaprada, Medhya , Sangrahi	Jantu ghan ,Rakatarsh, Raktavata, Kandu, Visarpa Kustha Visha , bhoota , bali palitya chardi , Meha Jwar
Shaliggram Nighantu Guduchyadi varg-215,251	Grahi Valya, Rasyana ,Dipna ,Hridya, Ayushyaprada , Chakshusya, Tridoshghana	Jwar , Chardi , Kamla, Daha , Trishna, Brham , Pandu, Prameha, Kasa , kustha,Krimi, Vatarakta Kandu, Meda,

,253		Visarpa, Aruchi, Hikka, Arasha , ,Mutrakricch, Pradra, Somroga
Sushrta Samhita, sutra sathan 46;270, Chikitsa, 18:5,46	*Pitta- Kaphahar*	Vatja Granthi Vatja Galgand

Table No. 2.7: Some of the important Active components of *Guduchi* (*T. cordifolia*).

Sr.no.	Active components	Compounds	References
1. .	Terpenoid	Furanolactone clerodane diterpene, cordifoliside D and E, furanoid diterpene, Tinosporide, ecdysterone, Tinosporaside, Tinocordioside, polyacetate, phenylpropene disaccharides cordifolioside A, B, and C, makisterone, cordioside, Furanolactone diterpene, palmatosides C and F, Sesquiterpene glucoside tinocordifolioside, and Sesquiterpene tinocordifolin..	(Khuda M.Q.I., Khaleque A., 1964) (Word, 1995) (R K BHATT, 1987), (Hanuman, Bhatt and Sabata, 1986), (Choudhary et al., 2013) (Gagan et al., 2010)
2.	Alkaloid	Tinosporine, (S), Berberine, (S), Jatrorrhizine, (S), Magnoflorine, (S), 1,2-Substituted pyrrolidine (S), Choline, (S), Alkaloids, viz. jatrorrhizine, palmatine, berberine,tembeterine, choline.	(Hanuman, Bhatt and Sabata, 1986), (Choudhary et al., 2013), (Bisset and Nwaiwu, 1983)
3.	Lignans	3(a, 4-dihydroxy-3-methoxybenzyl)-4- (4-hydroxy-3-methoxybenzyl), (S)	(Sharma et al., 2019)
4.	Steroids	Giloinsterol, (S), ß-Sitosterol, (S),20aHydroxy ecdysone, (S).	(Upadhyay and Kumar, 2010)
5.	Others	Tinosporal acetate, Octacosanol, Tinosporan acetate, sinapic acid, Tinosporidine, Heptacosanol, Tinosponone, two Phyto ecdysones, Giloin, an immunologically active arabinogalactan.	(Rakesh Maurya, Versha Wazir, Anjulika Tyagi and More, 1995), (P Pradhan, V.D. Gangan, A.T. Sipahimalani, A. Banerji, VD Gangan, P. Sipahimalani, no date)

Anti-diabetic Effects

In 2023, researchers investigated the potential of *Tinospora cordifolia* stem extracts to combat diabetes. Their findings revealed a notable decrease in blood sugar levels in diabetic rats, suggesting that these extracts could be effective in treating diabetes mellitus (Sharma et al., 2023).

Studies have suggested that *Guduchi*, also known as *Tinospora cordifolia*, aids in controlling blood sugar levels by influencing insulin secretion and enhancing insulin sensitivity. This indicates its potential benefits in managing diabetes (Jantan et al., 2015).

- **Neuroprotective Effects**

A study published in 2021 demonstrated that stem extracts of *Tinospora cordifolia* possess neuroprotective properties, potentially useful in addressing neurodegenerative conditions. This research underscored the plant's capacity to shield neurons from oxidative harm and enhance cognitive abilities (Patel et al., 2021).

Research indicates that *Guduchi* exhibits neuroprotective characteristics, potentially advantageous in managing neurodegenerative ailments like Alzheimer's disease. This is attributed to its antioxidative attributes and capability to regulate neuroinflammatory reactions (Raveendran Nair et al., 2004).

- **Hepatoprotective Activity**

In 2022, a thorough investigation demonstrated the hepatoprotective effects of *Tinospora cordifolia*. The study revealed that stem extracts of the plant shield the liver from harmful substances and promote its optimal functioning (Gupta et al., 2022).

Guduchi is recognized for its ability to safeguard the liver, aiding in detoxification and shielding liver cells from harm induced by toxins and free radicals (Leyon and Kuttan, 2004; Prince and Menon, 1999).

- **Antimicrobial and Antiviral Activities**

Research conducted from 2019 to 2024 has consistently highlighted the antimicrobial and antiviral attributes of *Tinospora cordifolia* stem extracts. These investigations showcased its efficacy against numerous bacterial and viral strains, suggesting its potential utility in combatting infectious ailments (Reddy et al., 2021; Mehta et al., 2022).

Guduchi stem demonstrates wide-ranging antimicrobial activity against diverse

pathogens, rendering it efficacious in addressing infections stemming from bacteria, fungi, and viruses (Alrumaihi et al., 2019). Selecting the appropriate material is crucial for efficacy, especially if there are gender-related variations in the qualitative and quantitative contents of the medicinally active or significant ingredients and metabolites. Current research has demonstrated that these distinctions indeed occur. Plants that are male or female differ in the length, shape, and form of their petioles. Quantitative anatomical traits also provide the basis for distinguishing between male and female plants. The presence of starch grains, mucilage canals, and the size of the cortical area were among the crucial characteristics that varied considerably between the sexes. Female plants also had higher levels of carbohydrate, tannin, and total sugar. *Sanshamni Vati*, also known as *Guduchi Ghan* (concentrated form of decoction), is an Ayurvedic preparation that is employed as an adjuvant in a number of hospital studies against COVID.

2.6 Dioecy

The condition where male and female organs are found on separate individuals within a species, is a fascinating phenomenon in plant biology. This reproductive strategy has significant implications for genetic diversity, ecological interactions, and evolutionary dynamics. The reproductive organs are the sole features in dioecious plants that differentiate the sexes. These include surrounding somatic tissues, Floral structures, which are frequently formed from altered stems or leaves, include parts of angiosperm flowers such as the calyx and corolla as well as vital reproductive organs like stamens and pistils.

Sexual Dimorphisms and Adaptations: It is widely accepted that sexual dimorphisms in plants, such as variations in the reproductive systems of male and female individuals, are evolutionary traits. According to Moore & Moore (2017), females are involved in pollen or sperm collection, as well as systems for protecting and providing for developing embryos, whereas males are usually regarded as playing functions in the distribution of pollen or sperm. Evolutionary Implications of Dioecy: In order to encourage outcrossing and inhibit inbreeding, dioecy frequently develops as a method to increase genetic variety within populations. Furthermore, dioecious plants enhance their ability to adjust the shifting Environmental factors and lower the possibility of self-fertilization by segregating their male and female activities (Charlesworth &

Charlesworth, 1978).

Impact of Dioecism on Therapeutic Efficacy: Being a dioecious plant, *Guduchi* (*Tinospora cordifolia*) has distinct male and female individuals. However, the gender of the plant is frequently disregarded when gathering plant material for therapeutic purposes. Given that research indicates that a plant's gender might influence its biological activity, this oversight may have an impact on the extract's therapeutic efficacy (Charlesworth & Charlesworth, 1978).

Evidence of Dioecism: There is evidence to support *Tinospora cordifolia*'s dioecious nature from a number of studies. Using molecular markers, Saha and Mukherjee (2017) carried out a thorough analysis that verified the existence of Separate male and female plants in wild populations. The dioecious character of the species was further supported by Ghosh and Chattopadhyay's (2019) observation of sex-specific changes in floral morphology and reproductive components.

Impact on Medicinal Properties: The therapeutic properties of *Tinospora cordifolia* are significantly impacted by dioecism. Sharma et al.'s (2018) research revealed changes in the amounts of important bioactive components like flavonoids and alkaloids as well as variances in the phytochemical makeup between male and female plants. These results imply that the medicinal effectiveness of *Tinospora cordifolia* preparations may be influenced by the gender of the plant.

Genetic Diversity and Conservation: Dioecism is essential to the preservation of genetic variety in populations of *Tinospora cordifolia*. Mishra and Tripathi's (2020) research emphasized the significance of conserving both male and female plants in order to maintain the genetic variety of the species. Furthermore, knowledge of the genetic underpinnings of *T. cordifolia* dioecism may help with breeding initiatives targeted at improving therapeutic qualities.

Ecological Significance: *Tinospora cordifolia*'s dioeciousness has an impact on population dynamics and reproductive methods, among other ecological issues. The function of pollinators in moderating reproductive success in dioecious populations was examined by Singh etal.(2019) in their research, which provided insight into the ecological interactions influencing the distribution and abundance of the species.

In summary, *Guduchi*'s (*Tinospora cordifolia*'s) dioeciousness has significant ramifications for a number of areas of plant biology, including genetic diversity,

medicinal qualities, and ecological interactions.

In plant biology, dioecism—which is defined by the separation of male and female reproductive systems in separate individuals- is an amazing phenomenon. It emphasizes how reproductive methods have evolved throughout time and greatly increases genetic variety within populations.

Dioecious plants, such as *Guduchi*, improve their ability to adapt to shifting environmental conditions by encouraging outcrossing and inhibiting inbreeding, which ultimately ensures their survival and resilience.

It is unbearable to overestimate the effect of dioecism on the therapeutic effectiveness of *Guduchi* preparations. Research has indicated that the phytochemical content of male and female plants differs, indicating that a plant's gender could impact its therapeutic characteristics. Additionally, maintaining the genetic variety of *Guduchi* (*Tinospora cordifolia*) populations and guaranteeing the sustainability of medicinal resources depend on the maintenance of both male and female plants.

In terms of ecology, *Guduchi*'s (*Tinospora cordifolia*) dioeciousness is important for population dynamics and reproductive strategies since pollinators influence the success of reproduction in dioecious populations.

Ultimately, optimizing *Guduchi* (*Tinospora cordifolia*) medical benefits, preserving genetic variation, and comprehending its ecological relevance in natural environments all depend on our ability to comprehend and accept the plant's dioecious nature. Our understanding of this fascinating plant species will continue to be enhanced by additional investigation into the genetic, physiological, and ecological mechanisms behind dioecism in *Guduchi* (*Tinospora cordifolia*)

2.7 Inflammation

The initiation of inflammation involves multiple processes. Upon encountering an infection, vasodilation occurs, facilitating the movement of fluid exudates containing leukocytes to the site of infection. This response, triggered by infection, antigens, or tissue damage, aims to eliminate microbes and promote tissue healing. However, if inflammation becomes excessive, it can cause harm to the affected tissue. Depending on the duration and severity of the injury, inflammation is broadly categorized as acute or chronic. Chronic inflammation is observed in conditions such as rheumatoid arthritis, systemic *lupus erythematosus* (SLE), inflammatory bowel disease (IBD),

silicosis, and atherosclerosis, where tissue damage and repair processes occur concurrently over extended periods, ranging from weeks to years. Acute inflammation is characterized by vasodilation, protein-rich fluid exudates, and migration of neutrophils to the site of injury, Additionally, involving the activation of the coagulation cascade. Vasodilation, a hallmark of inflammation, leads to localized redness and warmth at the injury site, facilitating the recruitment of Endogenous mediators and inflammatory cells. Substances like nitric oxide and prostaglandins mediate this vasodilation.

Nitric oxide acts through cGMP-dependent mechanisms to induce smooth muscle relaxation. It is synthesized from L-arginine by nitric oxide synthase (NOS), leading to smooth muscle relaxation via cGMP-dependent pathways. Prostaglandins (PGs), derived from arachidonic acid through the cyclooxygenase pathway also contribute to vasodilation. When exposed to compounds made by microbes or proinflammatory cytokines leukocytes release NOS. Another characteristic of inflammation is edema, which results from the movement of protein-rich fluid from the blood vessels to the surrounding tissue. This is mediated by histamine, bradykinin, leukotrienes, complement components, substance P, and platelet-activating factor), which increase capillary permeability to water and proteins Additionally, inflammation-induced vasodilation increases capillary hydrostatic pressure, promoting the transfer of protein-rich fluid to the interstitial space. This, in turn, raises blood viscosity due to higher erythrocyte concentration in small vessels. Consequently, the loss of plasma proteins reduces intravascular oncotic pressure. Elevated vascular permeability, increased capillary hydrostatic pressure, and decreased plasma oncotic pressure collectively lead to the trans vascular movement of fluids and proteins into the interstitial fluids. This facilitates the migration of antibodies, proteins, and leukocytes to the site of injury. Neutrophils are the initial responders to infection, migrating from the blood vessels to the interstitial fluid space. This migration process involves distinct phases.

Steps: Adhesion, Margination, Chemotaxis and Diapedesis

- **Margination**

It involves neutrophils transitioning from the central bloodstream to the periphery of blood vessels. This movement occurs due to the stable fluid conditions at the inflammation site and physical interactions between red blood cells and neutrophils.

These interactions lead to the formation of weak bonds between neutrophils and the endothelial cells that line the capillaries, keeping the neutrophils near these cells.
The rolling of leukocytes was aid with molecules called selections and their corresponding ligands. Selectins are present on various cell types including leukocytes (L-selectins), endothelial cells (E- selectins), and platelets (P-selectins). When these selectins bind to specific carbohydrate structures called sialylated carbohydrates on nearby cells, they facilitate the movement of neutrophils from the bloodstream into the interstitial spaces through a process known as rolling.

- **Adherence**

It refers to the process where neutrophils attach to endothelial cells, facilitated by the strong binding between integrins and specific ligands. Integrins, which consist of alpha and beta subunits, are found on the surface of neutrophils, primarily beta-2 integrins. When these integrins interact with ligands present on endothelial cell membranes, notably ICAM-1, it leads to the adherence of neutrophils to the endothelial cells. This attachment is pivotal in transitioning neutrophils from margination to adherence onto endothelial cells, enabling them to penetrate these cells and enter the interstitial fluid environment. (38) Additionally, this activity enhances diapedesis (the passage of blood cells through the intact walls of capillaries) and chemotaxis (the movement of cells in response to chemical signals).

- **Diapedesis**

During diapedesis, the junctions between endothelial cells retract, and the platelet endothelial adhesion molecule (PECAM-1) is present on both neutrophils and endothelial cells. The binding of neutrophils to PECAM-1 reduces their affinity to attach to intercellular adhesion molecules (ICAM-1), thereby inhibiting adherence and promoting diapedesis. These interactions between adhesion molecules on endothelial and neutrophil cells aim to enable neutrophil migration from the bloodstream to the interstitial fluid, occurring at sites of infection or injury.

- **Chemotaxis**

It involves leukocytes being guided towards sites of injury or infection by chemo attractants. These chemo attractants, such as chemokines (chemoattractant cytokines), consist of alpha and beta types. Alpha chemokines include interleukins (e.g., IL-8), while beta chemokines encompass bacterial byproducts, complement components, and

more. Together, these chemokines orchestrate a pro- inflammatory response at the affected site. During infection or injury, locally produced chemokines recruit leukocytes to the site, along with fluid accumulation specific to the inflammatory process of that particular disease. Chemokines also regulate the composition of inflammatory fluid in the tissue. The migration of leukocytes hinges on the specificity of chemokine receptors and the chemokines they bind to. These receptors, which are structurally similar but functionally distinct, are G-protein-coupled proteins.

Moreover, acute inflammation triggers the activation of the coagulation system, which aids in the formation of inflammatory responses. This process, known as the coagulation cascade, consists of a series of events categorized into two pathways: Both the extrinsic and intrinsic pathways converge upon activation of thrombin, which converts fibrinogen into fibrin.

The intrinsic pathway is activated in response to direct tissue trauma. It involves a sequence of events initiated by plasma proteins, culminating in the activation of thrombin through factor XII. This activation occurs when proteins synthesized in the liver, they attach to collagen, the basement membrane, or activated platelets. During infection and systemic inflammation, the extrinsic coagulation pathway is primarily activated. Initiation occurs through tissue factors (TF) located on tissue surfaces that are not directly in contact with blood. Endothelial cells and activated monocytes generate tissue factor (TF) in response to inflammatory molecules like TNF-α, IL-1, IL-6, and C- reactive protein.TF subsequently activates factor VII, initiating a cascade of coagulation factor activations that culminate in thrombin formation.

Proinflammatory response: The coagulation process plays a crucial role in the proinflammatory response by triggering the formation of proinflammatory cytokines. Factors Xa, thrombin, and the TF- VIIa complex are particularly involved in this process, Thrombin and the TF-VIIa complex are the main producers of proinflammatory Cytokines. For instance, TNF-α is primarily produced by mononuclear cells and endothelial cells in response to these coagulation factors. To prevent excessive coagulation, various mechanisms inhibit the coagulation process. These include the formation of antithrombin, protein C, and tissue factor pathway inhibitor (TFPI), which counterbalance pro coagulation mechanisms. process, Thrombin and the TF-VIIa complex are the main producers of proinflammatory

cytokines. For instance, TNF-α is primarily produced by mononuclear cells and endothelial cells in response to these coagulation factors.

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Antithrombin

Anti-thrombin acts by binding to thrombin, thereby inhibiting the coagulation pathway. The presence of heparin and glycosaminoglycans on endothelial cell surfaces enhances this inhibition process. Antithrombin was recognized for its role in regulating coagulation and also exhibits anti-inflammatory properties. In rodents, the binding of anti-thrombin to endothelial cell surfaces triggers the release of prostacyclin (PGI2). Prostacyclin, in turn, inhibits the production of TNF-α by monocytes through the activation of NF-κB.

a) Protein C

The pathway begins with the thrombin-thrombomodulin complex found on endothelial cell surfaces. Protein C functions to inhibit this pathway by deactivating factors Va and VIIa. Moreover, it suppresses TNF-α production in monocytes by inhibiting the NF-κB and AP-1 pathways. Protein C is recognized as an anticoagulant with anti-inflammatory properties. During sepsis, a decrease in thrombomodulin due to inflammation leads to a depletion of protein C. Consequently, excess thrombin is formed, resulting in an accelerated coagulation process. The significance of protein C regulation of thrombin becomes evident in septic patients, where mortality rates increase if protein C levels are depleted.

b) TFPI

TFPI, also referred to as tissue factor pathway inhibitor, is present on endothelial cell surfaces where it is bound to plasma lipoproteins. It plays a crucial role in regulating the coagulation cascade by inactivating tissue factor (TF). This inhibition occurs through the formation of a complex involving TFPI, tissue factor, factor VIIa, and factor Xa, known as the TFPI-TF-VIIa-Xa quaternary complex. Inhibiting TF prevents activation of the extrinsic pathway of coagulation. The Complement activation pathway comprises a series of proteins that become activated in response to microbial presence, resulting in inflammation. Tissue injury can trigger complement system activation via

two primary pathways: the classical pathway and the alternative pathway. The classical pathway of the complement system is initiated by the presence of immunoglobulin M (IgM) and Immuno-globulin G (IgG) antibodies bound to the surface of microbes or other structures. Alternative pathway of the complement system is triggered when surface molecules of microbes directly interact with component C3 within the complement system, leading to the activation of complement proteins. Both the alternative and classical pathways play a role in cleaving the C3 component into C3a and C3b. C3a acts as a chemoattractant, drawing immune cells towards the site of infection, while C3b binds to the microbial surface, facilitating the recognition of microbes by phagocytes and promoting their engulfment (phagocytosis). Following this, C3b can form complexes with other components of the complement system. Subsequently, C5 is cleaved into C5a and C5b. C5a induces changes in vascular permeability and serves as a chemoattractant for neutrophils, guiding them to the site of infection. Conversely, C5b binds to microbial surfaces and collaborates with components C6, C7, C8, and C9 to form the membrane attack complex (MAC). The MAC disrupts microbial membranes, leading to cell death. through the formation of a complex involving TFPI, tissue factor, factor VIIa, and factor Xa, known as the TFPI-TF-VIIa-Xa quaternary complex. Inhibiting TF prevents activation of the extrinsic pathway of coagulation

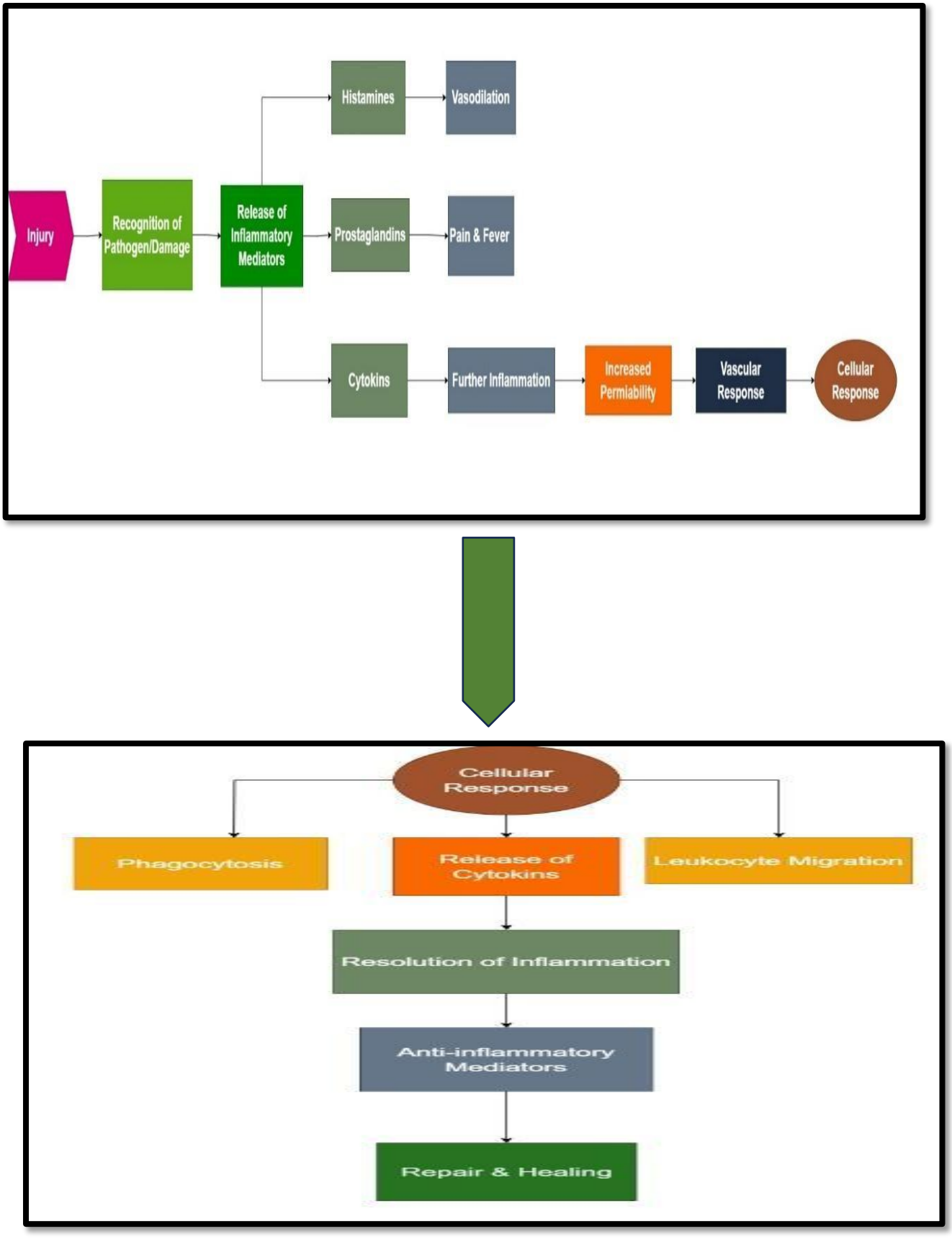

Figure 2.1: Mechanism of the Inflammatory Response

The Complement activation pathway comprises a series of proteins that become activated in response to microbial presence, resulting in inflammation. Tissue injury can trigger complement system activation via two primary pathways: the classical pathway and the alternative pathway. The classical pathway of the complement system is initiated by the presence of immunoglobulin M (IgM) and Immuno-globulin G (IgG) antibodies bound to the surface of microbes or other structures. Alternative pathway of the complement system is triggered when surface molecules of microbes directly interact with component C3 within the complement system, leading to the activation of complement proteins. Both the alternative and classical pathways play a role in cleaving the C3 component into C3a and C3b. C3a acts as a chemoattractant, drawing immune cells towards the site of infection, while C3b binds to the microbial surface, facilitating the recognition of microbes by phagocytes and promoting their engulfment (phagocytosis). Following this, C3b can form complexes with other components of the complement system. Subsequently, C5 is cleaved into C5a and C5b. C5a induces changes in vascular permeability and serves as a chemoattractant for neutrophils, guiding them to the site of infection. Conversely, C5b binds to microbial surfaces and collaborates with components C6, C7, C8, and C9 to form the membrane attack complex (MAC). The MAC disrupts microbial membranes, leading to cell death.

How is Immunity Modulated?

IL-6, produced by various cells during inflammation, supports B lymphocyte growth and differentiation. Although not directly involved in tissue injury, elevated IL-6 levels in sepsis patients often indicate a poor prognosis. IL-8 serves as a chemoattractant, drawing neutrophils to sites of inflammation. IFN-γ, a cytokine, accelerates the inflammatory response by stimulating cytokine secretion and macrophage activity. IL-12, produced by macrophages and dendritic cells, triggers NK cells to produce IFN-γ, amplifying the inflammatory response and enhancing immunity. IL-10 inhibits macrophage and dendritic cell activation, by blocking the production of IL-12 and IFN-γ, they have anti-inflammatory properties. PAF, Endothelial cells secrete endothelin, which controls the release of cytokines and enhances neutrophil adherence. Vasoconstriction and platelet aggregation are induced by thromboxane A2. Inflammation is decreased by preventing IL-12 and IFN-γ from being produced. IL-10 plays a key role in regulating the body's inflammatory response. In summary, there

exists a delicate balance between inflammatory mediators such as TNFα, IFN-γ, and IL-12, and anti- inflammatory mediators like IL-10, TGF-β, and prostaglandins (PGs). In mice lacking IL-10, hyperinflammatory changes occur, making them vulnerable to bacterial toxins. Conversely, external administration of IL-10 suppresses pro-inflammatory changes. G-CSF, or granulocyte colony- stimulating factor, encourages the bone marrow to produce and release neutrophils enhancing their killing activity. When G-CSF interacts with receptors on macrophages and monocytes G-CSF leads to a decrease in the levels of TNF-α, IL-1, and IL-12.

Acquired immunity

IL-12 plays a crucial role in activating acquired immunity, particularly by stimulating T-cells to differentiate into Th-1 cells. This process is initiated when antigens are presented to CD4 and CD8 T cells, triggering the release of cytokines that enhance both acquired and natural immunity in the body. Helper T cells are categorized into Th-1 and Th-2 cells based on the cytokines they produce. Th-1 cells release IFN-γ, which boosts pro-inflammatory functions by stimulating macrophages and cytotoxic T cells through the secretion of IL-12. B cells also contribute to acquired immunity by producing antibodies. External microbial cells can induce the production of IL-12, activating transcription factors in T cells to promote Th-1 differentiation. Th-1 cells play a crucial role in promoting phagocytosis When the body encounters an allergen, T cells differentiate into Th-2 cells instead of initiating a natural immune response. This differentiation is triggered by the production of IL-4. Although IL- 12 is also produced in this process, its quantity is minimal, resulting in minimal phagocytosis. IL-4 activates transcription factors that aid in the differentiation of T cells into Th-2 cells.53 release Numerous cytokines are important, such as IL-4, IL-5, IL-10, and IL-13. IL-4 collaborates with IL- 10 and IL-13 to regulate immunological responses, suppresses macrophages and promotes the formation of IgG and Ige antibodies, facilitating allergy and anti-parasitic responses.54 Allergic reactions are triggered by IL-4. Additionally, T cells also include Th-3 cells, which produce TGF-β and promote immune tolerance upon exposure to antigens in the gastrointestinal tract. 55Studies in mice have shown that the absence of TGF-β results in a hyperinflammatory response, leading to systemic inflammatory changes and death within weeks after birth.

References: (Sivakumar and Dhana Rajan, 2010), (Sharma et al., 2013), (Rajalakshmi et al., 2009), (Gupta, Gupta and Bajpai, 2024), (Pati Pandey, Kamra Verma and Anita, 2011), (Ahmad, Jantan and Bukhari, 2016), (Singh et al., 2022).

2.8 Carrageenan Paw Edema

The paw edema model caused by carrageenan is a commonly used technique to investigate acute inflammation. It involves multiple inflammatory mediators and is widely utilized to assess the anti- swelling properties of natural substances. Injection of carrageenan 1% (50 ml) in the mouse paw causes a biphasic response: One method for studying inflammation in research is the paw edema model caused by carrageenan. This technique involves injecting carrageenan, a material produced from red seaweed, into an animal's paw—usually a rodent—to cause localized inflammation and swelling. Using this model, scientists can monitor the body's inflammatory response and evaluate how well anti-inflammatory drugs work.

In 1962, Winter, Risley, and Nuss presented the model for the first time. The Upjohn Company in the US was the site of the groundbreaking research.

Significance: Their study proved that the technique could consistently cause inflammation, and so it became a standard in pharmacological investigations assessing the anti- inflammatory capacity of different drugs.

(Phyllis E Whitely 2001)

2.8.1 Studies to explore the anti-inflammatory effects of *Guduchi* using the carrageenan-induced paw edema model.

The research aimed to evaluate the anti-inflammatory effects of the alcoholic extract of *Tinospora cordifolia* using two experimental models: Male Wistar rats were used to evaluate the effects of carrageenan-induced hind paw swelling and cotton pellet-induced granuloma development. The findings indicate that T. cordifolia effectively mitigates inflammation in both short-term (acute) and longer-term (sub- acute) scenarios (J JOHN WESLEY 2008) The traditional Ayurvedic remedy for reducing fever, as detailed in the ancient Charak Samhita, underwent thorough scientific testing using established protocols. The study revealed that this formulation not only has significant anti-inflammatory properties but also provides strong pain relief both at the

site of inflammation (peripheral analgesic effect) and within the central nervous system (central analgesic effect), comparable to those of contemporary antipyretic medications. (Gupta et al., 2013)

- It was found that *Guduchi Ghana* prepared using traditional methods exhibited substantial anti- inflammatory effects. Therefore, the classical preparation method proved to be significantly superior compared to the commercial version. (Patgiri et al., 2014)
- The mention of "significant anti-inflammatory effects in the carrageenan-induced inflammation test" highlights the efficacy of *Tinospora cordifolia* extract in reducing inflammation, which complements its strong analgesic and antipyretic property. (Hussain, Liaqat,2015)
- *Tinospora cordifolia* (*Guduchi*) and Valeriana *wallichii* have notable analgesic and anti-inflammatory properties. Studies show that T. cordifolia provides a significantly stronger analgesic effect. (Deepika and Priyambada, 2016).
- Strong anti-inflammatory and antipyretic effects are demonstrated by *Tinospora cordifolia* (*T. cordifolia*). In contrast to NSAIDs, which may result in stomach problems, T. cordifolia provides immunomodulatory and gastro-protective properties without increasing the risk of drug dependency. It works by preventing the activity of the COX and LOX enzymes. This plant is readily accessible year-round, which makes its procurement and preparation simple and affordable. Even with these encouraging qualities, more extensive research is required to completely comprehend any possible negative effects and how it could interact with other medications. As a foundation for creating novel therapeutic medicines that combine COX and LOX inhibition with fewer side effects, *T. cordifolia* exhibits potential. (Suman et al., 2019)

A comprehensive review was conducted on Ayush recommended formulations and their constituent ingredients, widely utilized by the majority of the Indian population during the COVID-19 pandemic. Special attention was given to plants known for their antiviral, anti-allergic, anti-inflammatory, and immunomodulatory properties. (Ahmad et al., 2021) revealed that this formulation not only has significant anti inflammatory properties but also provides strong pain relief both at the site of inflammation (peripheral analgesic effect) and within the central nervous system (central analgesic effect), comparable to those of contemporary antipyretic medications. (Gupta et al., 2013)

Table2.8 Review of existing Literature on The Dioecy, Inflammation, Guduchi, prepration,Analytical, Pharmacological

Sr.No.	Authors	year	Category	Findings
1.	1877	Brian Charlesworth	Dioecy	A model for the Evolution of Dioecy and Gyno dioecy.
	1980	K.S. Bawa	Dioecy	Though less in proportion to world's flora, not as rare as generally considered.
	2004	Sherwood.E..et al,	Inflammation	Understanding immunology of inflammation & presented advanced outlook for basic inflammatory mechanisms
	2004	Sinha Kirti, Tiwari Prashant	Guduchi	Native drug participant for bioprospecting with scope to establish nutraceuticals from different parts of plant.
	2008	Vidyashree et al	Preparation	2008 Vidyashree et al Method of preparation of Ghana along with its benefits.
	2013	Sharma et al	Analytical	Parameters of optimum hardness, tablet weight, disintegration time, and friability considered to establish SMP for Guduchi Ghana and its tablet dosage forms (15 batches, 5% yield).
	2013	Sharma et al	Analytical	HPTLC peaks & characterization parameters observed for Guduchi Satva. Lack of published work for manufacturing guidelines & control parameters noted.

	2013	Patgiri et al	Pharmacologic al	Anti-inflammatory activity magnitude was less in marketed samples compared to classical samples.
	2013	Umeretia et al	Pharmacologic al	Guduchi Ghana vati (aqueous extract as per Siddha Yoga Samgraha) assessed for humoral & cell-mediated immunity → immunostimulatory action observed in marketed sample.
.	2013	Chaudhary Namrata et al	Guduchi	Stem is accepted for medicinal values due to high alkaloid content.
.	2013	Simpson et al	Dioecy	An important factor to be considered by researchers.
.	2013	Nagar katti	Immunomodul atory Effect of Guduchi	Immunomodulatory activity of Guduchi Ghana preparation as per classical texts (Aqueous extract of tinospora cordifolia)
.	0214	Rohit Sharma et al	Antimicrobial	Ghana and Satva found to have significant antimicrobial effect.
.	2014	Biswajyoti patigiri et al	Anti - Inflamatory effect of Guduchi	Prepared samples as per text and market samples of Guduchi exhibit anti-inflammatory activity.
.	2014	Kuchewar et al	Clinical	Kuchewar et al 500 mg Ashwagandha + Guduchi given to healthy volunteers under oxidative stress for 30 days; Guduchi + placebo for 6 months. Both helped prevent oxidative stress.
.	2015	Rohit Sharma	Guduchi	Effect of dioecy on Total alkaloid content – more in male Guduchi compared to female Guduchi

.	2016	Bajpai vikas et al,		Elicited the difference in abundance of alkaloids in male and female Guduchi
.	2017	Sayyada Khaton		• Elicited the difference in abundance of alkaloids in male and female Guduchi plant.
.	2017	Bajpai Vikas et al	Anti - Inflamatory effect of Guduchi	Effect of dioecy of Guduchi on upregulating anti-inflammatory cytokines
.	2018	Vidyashree et al	Preparation	Preparation Ghana is the most acceptable variety of Ayurvedic dosage forms. Discussed meaning of Ghana, preparation method, and therapeutic indications
.	2017	khatoon		Effect of dioecy on therapeutic applications stating importance of gender during plant Collection
.	2019	Tanvi Dayanand et al	Guduchi	Effect of seasonal variations on total alkaloid content – more in Grishma ritu compared to Sharad ritu.
.	2019	Sharma Priyanka et al	Anti - Inflamatory effect of Guduchi	Parameters to be used as an anti-inflammatory herb
.		Alushbani et al		• Elicited Guduchi to work as an Immunomodulator •
.		Upendra sharma	Gudachi in Covid-19	Isolated immune-modulatory active compounds from Tinospora cordifolia Guduchi Ghana vati was beneficial in

				asymptomatic patients of covid-19
.	2019	Kumar Abhimanyu et al	GUDUCHI IN COVID-19	Guduchi Ghana vati was beneficial in asymptomatic patients of covid-19
.	2019	Aman Sharma et al	Analytical	Physicochemical comparison of Guduchi Choorna, Satva, and Ghana showed marked variations. Alkaloid quantified.
.	2019	Dayanand Tanvi	Analytical	Standardisation of Guduchi Ghana in different Ritus. Yield & starch highest in Sharad, lowest in Grishma. Alkaloid max in Grishma.
.	2019	Saeed et al	Pharmacologic al	Humoral & cell-mediated immunity studied for anemia, gout, and aflatoxicosis.
.	2020	Deepa et al	Review	Reviewed Guduchi dosage forms from ancient compendia to recent years.
.	2020	Kumar et al	Clinical	Guduchi Ghana Vati useful in asymptomatic COVID-19 patients; more RCTs needed for confirmation.
.	2022	Sharma et al	Pharmacologic al	Guduchi noted as a natural herb with antimicrobial, anti-pyretic, and infection-preventive properties.

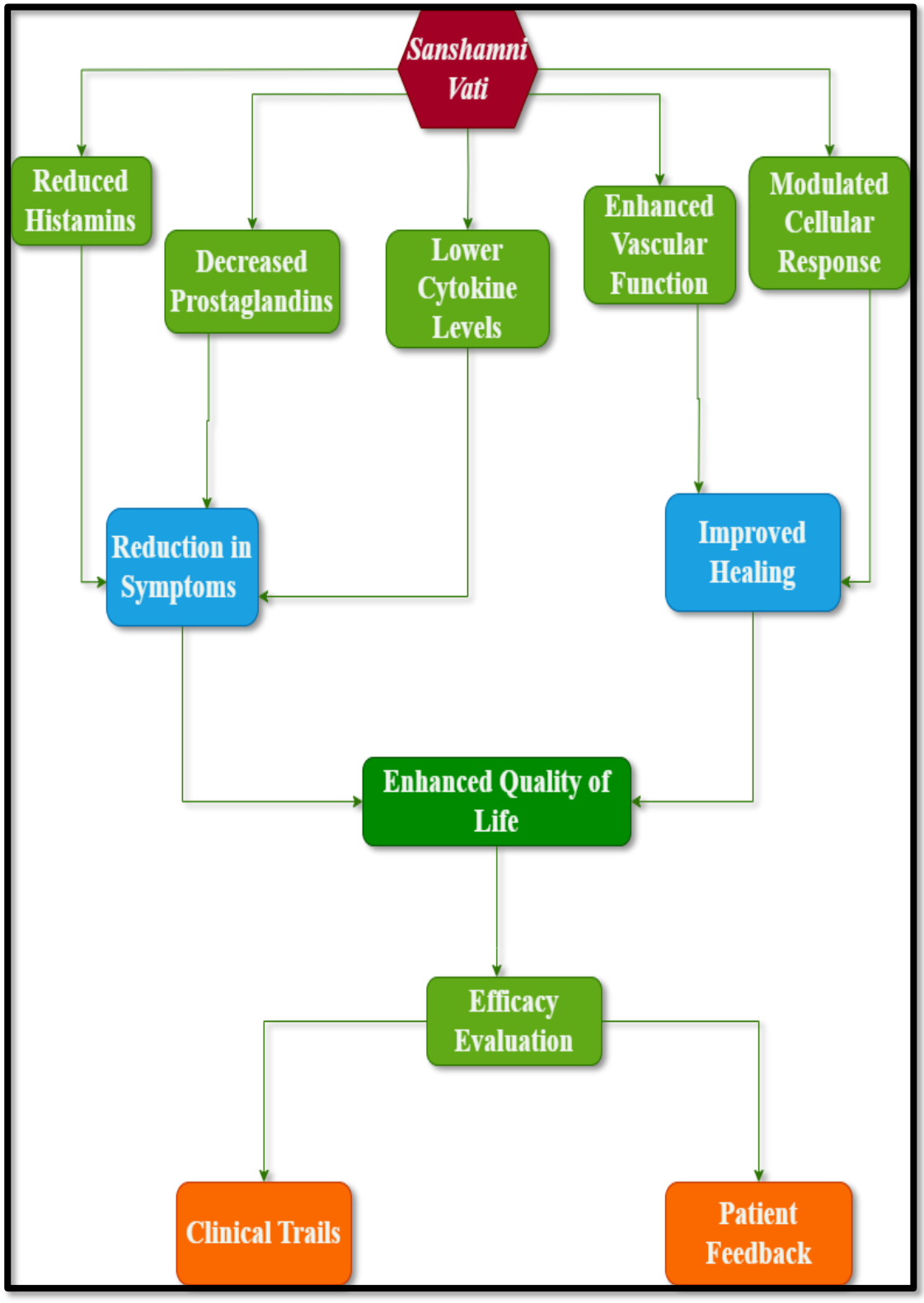

Figure 2.2: Impact of *Sanshamni Vati*

Chapter 3

Rationale

Dioecy is the term that describes the morphological and sexually difference of plants. The phenomenon of dioecy is rare and is restrained to only 7% of known taxa. This phenomenon affects the plant extracted yield, morphological, microscopical and pharmacological features. However, it is not considered as an appropriate consideration for the selection of raw materials. Few studies based on dioecy apparently suggest the role of dioecy in the quality of raw material. Ayurvedic texts do not highlight any direct reference for the dioecy. Moreover, the synonyms of the individual plant are gender-specific including Guduchi, Pippali, and Vijaya, etc. might be the direction for the collection of a specific gender of the dioecious plant. Ayurvedic name "Guduchi" signifies its feminine gender which might be considered as the selection criterion for its collection. However, classical literature does not provide any direct information about the dioecy of the T. cordifolia or other dioecious plants.

Research reveals the necessity of phytoconstituents for *Shanshamni Vati*'s therapeutic effects. Greater therapeutic results are attributed to the concentration of these bioactive chemicals, especially in terms of immunomodulatory and anti-inflammatory properties. The -present study has been designed to determine the effect of dioecism in the quality and efficacy of the formulations prepared by using dioecious plants as an ingredient. Aiming to improve the therapeutic efficacy of *Ghana* by choosing the right gender-specific plants, the manufacturing process will be guided by insights into the influence of dioecism. Hence, it is required to determine the impact of the dioecy in the phytoconstituents, formulations and its therapeutic activities. The present study has been designed to determine the impact of dioecism in quality and efficacy of Sanshamni Vati.

Aim of the study:

Impact of dioecism in the phytoconstituents of Sanshamni Vati, a formulation of *Guduchi* (*Tinospora cordifolia*) and its anti-inflammatory potential.

Objectives:

- Collection and Authentication of Both male and female plants of *Guduchi* (*Tinospora cordifolia*)
- Preparation of Sanshamni Vati

- Physicochemical, qualitative, and quantitative analysis of the collected plant materials and prepared formulations
- Evaluation and comparison of anti-inflammatory activities of the formulations

Chapter 4

Experimental work

4.1 Proposed methodology

4.1.1 Collection of drugs

Almost 40-50 kg of the fresh stem of both male and female plants of the *Guduchi* (*Tinospora cordifolia*) collected from Herbal Garden of Dayanand Ayurvedic college Jalandhar and Nursery of Horticulture Department Punjab at cantonment Road, near BSF campus Jalandhar (Punjab) during end June and July month.

4.1.2 Raw drug authentication Collected stems of both genders authenticated from the Government

The whole herbal plant was collected and identified according to the morphological features discussed in ayurvedic pharmacopoeia. A voucher specimen no. R.S-, 028 and 029 were deposited into the Herbal Health Research consortium, Amritsar, Ltd. Amritsar) for identification.

4.2 Morphological and Organoleptic and physicochemical standardization of *Guduchi (Tinospora cordifolia)*

The samples were evaluated based on method described by API standards and as mentioned in the research paper (Sharma, 2017). The parts used from the raw drugs were collected and subjected for morphological evaluations. Stem of both genders were observed for color, Odour and texture. Stems of guduchi plant tasted bitter. Fresh stems of raw drug were available in thickness ranging from 0.6 cm- 5cm in diameter. The young stems were tender and smooth, swelled at nodes. While the older stems were found light brown in color with warts like protuberances due to the circular shaped lentils. The transverse surfaces presented radial structures with noticeable medullary rays that pass through porous tissue. Physicochemical standardization of the herbal drugs was carried out to identify and assess the quality of phytoconstituents. All the parameters were evaluated as per described in section 4.5.1 to 4.5.11 described in the forthcoming paragraphs (Sharma, 2017)

4.2.1 Foreign Matter

100 gm of sample was spread in stainless steel tray and foreign matter was sensory observed. Foreign matter was removed and remaining quantity of the sample was weighed and foreign matter percentage is calculated.

4.2.2 Moisture content / Loss on drying (LOD)

10 grams of drug (without preliminary drying) were placed in an evaporating plate, The samples were dried at 105°C for 5 hours, followed by cooling in a desiccator, and then weighed until the difference between the result of two consecutive measurements did not exceed 0.25%. Weight is considered constant when there is less than 0.01g difference between the two weights. (Sharma, Amin and Prajapati, 2015) until the variation between 2 progressive observations is less than 0.25 % (LOHAR, 2011) (Gupta, A.K, 2008)

4.2.3 Total Ash Value (TAV)

Total Ash value was calculated as the Residue remaining after incineration of the 2gm of an air- dried sample of powdered and dried drugs (Sharma, Amin and Prajapati, 2015)

4.2.4 Acid Insoluble ash (AIA)

25 ml diluted HCL acid was mixed in the ash and boiled for 5minutes. Ashless filter paper was used for filtration (Whatman filter paper no.41). Hot water was used for the washing of the filtrate and subjected to muffle furnace for the ignition to achieve After reaching constant weight, the percentage of acid-insoluble ash was calculated.

$$Acid\ Insoluble\ ash\ = \frac{\text{weight of the ash residue}}{\text{weight of the sample}} X\ 100$$

4.2.5 Water soluble extractive (WSA)

Ash obtained from the above method (Acid insoluble ash) was mixed with 25ml water and kept boiling for 5 minutes. Filter the mixture through ashless filter paper (Whatman filter paper no.41), the filtrate was then washed with hot water. Weighed the obtained residue to Calculate the Percentage using the formula as below:

$$Water\ Insoluble\ ash\ = \frac{\text{weight of the ash residue}}{\text{weight of the sample}} X\ 100$$

Water Soluble = Total Ash − Water Insoluble Ash

4.2.6 Alcohol soluble extractive (ASE)

Measured 5gm sample (coarse powder) and added in a closed conical flask with 100

ml of alcohol, the conical flask was shaken frequently for 6 hours and then allowed to stand undisturbed for 18 hours. Afterward, 25 ml of the filtrate was taken into the evaporating dish, and the contents were allowed to evaporate.

The percentage was calculated after weighing the residue

$$\text{Water Soluble Extractive} = \frac{\text{weight of the residue}}{\text{weight of the sample}} X\ 100$$

4.2.7 Sulphated ash (SA):

Add 1 to 2 gm of accurately weighed substance, into heated and dried crucible at and ignite until thoroughly charred. Add 1ml of sulphuric acid and ignite again at 800^0C+25^0C. after the disappearance of black particles. allow particles to cool. weigh the crucible till the weight differences are not more than 0.5mg.

$$\text{Sulphated Ash (\%)} = \frac{\text{weight of the residue}}{\text{weight of the sample}} X\ 100$$

4.4 Microscopy Method

The Ayurvedic Pharmacopoeia of India (API) provides a standardized method for studying the anatomy of medicinal plants like *Tinospora cordifolia* through microscopy. To prepare a transverse section of the stem for microscopic observation, fresh and healthy stem samples are collected and cut into thin sections using a razor blade or microtome. These sections are placed in water to prevent drying. Staining the sections with agents like Safranin or Fast Green is essential to highlight the plant's cellular structures, with Safranin staining cell walls red and Fast Green highlighting other tissues. After staining, the excess dye is washed off, and the section is mounted on a glass slide using glycerin, which acts as a mounting medium. A coverslip is placed carefully over the section to avoid air bubbles, ensuring clear microscopic observation. Under the microscope, various anatomical features can be observed, such as the outer epidermis, the cortex containing parenchymatous cells, vascular bundles arranged in a ring (xylem and phloem), and the pith located centrally. This systematic method ensures accurate identification and quality control of the plant material, as specified in the Ayurvedic Pharmacopoeia.

4.5 Phytochemical Screening

Air-dried plant material extracts were prepared and analyzed for the presence and

absence of phytoconstituents which are responsible for producing therapeutic effects i.e. Tannins, Glycosides, Saponin, Protein, Carbohydrate, Alkaloids, Steroids, Phenols, and Flavonoids. **(Sharma, Amin and Prajapati, 2015) (Singh et al., 2003)**

4.5.1 Test of Tannin

4.5.1.1 Lead Acetate Test

3ml Test Solution was taken the 5% Lead Acetate was added which on addition of KOH produces Reddish Precipitates which will disappear on more addition of KOH.

4.5.2 Test for Alkaloids

4.5.2.1Dragondorff Reagent Test

A 2ml test solution was taken in a test tube in which few drops of 2N HCl were added then 0.5 ml dragon Dorff reagent (Mixture of Potassium Mercury Iodide and Bismuth Sub nitrate Solution) was added. Orange reddish precipitates appears at the bottom indicated presence of Alkaloids.

4.5.3 Test for Carbohydrates

4.5.3.1 Molisch's Test

In 3ml of hydroalcoholic extract, two drops of fresh 10% alpha naphthol was mixed then 2ml of conc. Sulphuric acid was added. After sometime formation of red purplish ring at the upper layer was seen indicating the presence of carbohydrates.

4.5.4 Test for Flavonoids

4.5.4.1 Shonda's test

3ml Test Solution was taken and 5m of 95%(w/v) Ethanol was added then few drops of conc. Hcl and 0.5 gm of $MgCl_2$ which gave the pinkish or magenta color to the solution. On addition of few drops of lead acetate light yellowish color precipitates appear.

4.5.5 TEST FOR REDUCING SUGAR

4.5.5.1 Benedict's Test

In 2ml Test Solution,2ml of Benedict's Reagent was added and shaken well. Then it was heated on water bath for 05 minutes solution appears dark green indicating the presence of Sugar.

4.5.6 TEST FOR STARCH

4.5.6.1 Iodine Test

3ml of Solution was taken and then few drops of iodine solution were added. Brownish green solution turns dark blue indicating

4.5.7.1 Proteins

4.5.7.1 Biuret's

1ml of the test extract was combined with a little amount of 1% copper sulphate solution and 4% sodium hydroxide solution. The appearance of a violet-red color signified the presence of proteins.

4.5.8 Test for Amino Acids

4.5.8.1Ninhydrin Test

Ninhydrin Test 3ml Test Sample was heated on water bath the 3 drops of 5% Ninhydrin Solution which is again heated on water bath for 10 mins Solution turns purplish indicative of presence of Amino Acids.

4.5.9 Test for Saponins

4.5.9.1 Foam Test

A prolonged froth formed in a test tube after vigorously shaking the extract with distilled water, suggesting the presence of saponins.

4.5.10 Test for Steroids

4.5.10.1 Salkowski Test Salkowski Test 2ml Test Solution was taken and 2ml of $CHCl_3$ and 2ml of H_2SO_4 were added which in turns appears Lower $CHCl_3$ layer dark red & Upper H_2SO_4 appears with yellow greenish fluorescence which is indicative of presence of Steroids

4.5.11 TEST FOR CARDIAC GLYCOSIDES

4.5.11.1 Keller Killani Test 2ml Test Solution was taken and 1ml Glacial Acetic Acid was added with 01 drop of 5% $FeCl_3$ and few drops conc. H_2SO_4. Solution turns Reddish Brown indicative of presence of Cardiac Glycosides.

4.6 Determination of pH value

Determine pH value of aqueous solution of drug by preparing a 1% solution of herbal drug w/v with the help of a glass electrode.

4.7 Determination of trace and heavy metals in the plant materials

Analysis was performed according to flame atomic absorption spectroscopy in three

herbal drugs. Calibration curves prepared from Standard Working solutions in range of 1ppm to 10 ppm from stock solutions of 1000 ppm of all standards of Hg, Cd, As and Pb purchased from Merck, Germany for reference purpose. 3gm of Accurately dried samples were treated with 3ml nitric acid for about 5 hr duration. Later proportionate to HNO_3, half in amount of $HCLO_4$ was added and heated for about 6 hrs till solution becomes clear and flames stop coming out of it. Then add milli-Q water and boil for 15 min to reduce to half the prior volume. Cool and filter with Whatman filter paper no.42. Make the volume up to 50ml with milli-Q water. Prepare blank in similar way and aspirate each was assessed for AAS. for AAS. (Kulhari et al., 2013.

Heavy metals are identified as Human carcinogens identified by the International Agency for Research on Cancer (IARC) and the U.S. Environmental Protection Agency (EPA). A variety of chronic illnesses can result from drugs tainted with heavy metals

4.8 Microbiological evaluation

Prepare two petri dishes by adding 15ml of liquified casein soyabean digest agar and 1ml of prepared sample is poured on this medium of not more than 450C temperature is kept on petri dish (dilute pretreated material if needed) On obtaining the colony count of near about 300. Incubate them for 48-72hrs. colony numbers are counted in colony counters up to 300 colonies are considered for good evaluation. For fungal count Saboraud dextrose agar is taken with chloramphenicol is used and a reliable count to a maximum of 100 colonies are considered for good evaluation. (Ezhilarasu et al., 2023). The microbial load test tells us what kinds and quantities of viable microorganisms are present. If the drug's microbial contamination is found to be higher than the recommended threshold, it is rejected for additional testing and standardization because the presence of microbes may make it unsafe to use.

The quantitative measurement of microorganisms is known as the microbial load. The official monograph's microbiological quality characteristics were compared with those observed in the microbial test. The findings of the research on microbial plate count and total yeast and mold test fall within the specified range, indicating that the sample meets the set standards for microbiological quality.

4.8.1 specific pathogens

1 ml of sample is transferred in 50 ml of nutrient broth and incubated. This method is employed for examining *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella*

typhimurium, and *Staphylococcus aureus*. For *Escherichia coli*, 1 ml of the prepared subculture is mixed with 5 ml of McConkey agar and then incubated at 37°C. The presence of red-colored growth indicates the presence of *Escherichia coli*.

Pseudomonas aeruginosa 1 ml of prepared subculture is taken with cetrimide agar and at 37°C incubation was done for 24hrs. then the plates are detected for pathogen growth.

Staphylococcus aureus inoculate subculture on Baird-parker agar, incubate at 37°C for 48 hrs. check for growth for the presence of microorganisms. (More and Pai, 2011) These substances impair the quality and cause instability in the medicine and its ingredients; thus, the sample must be clear of them. Plant diseases are caused by pathogens, so the primary criterion for examining any plant illness was the results of the pathogen test. Gram +ve (*Staphylococcus aureus*) and gram -ve (*Escherichia coli*, *Pseudomonas aeruginosa*) bacteria were cultivated by the Agar well diffusion technique.

4.8.2 Aflatoxin test

Dissolve 1.0 mg of crystalline Aflatoxin G1, G2, B1, and B2 in 50 ml of a toluene-acetonitrile (9:1) solution in a glass flask. Shake the mixture vigorously to achieve a 20 µg/ml standard stock solution. Weigh and dissolve this solution in 60 ml of a toluene-acetonitrile (9:1) solution, then shake the mixture in a glass flask. Store the standard solution in a refrigerator at 4°C. (Ministry for Health and Family Welfare, Ministry for Health and Family Welfare and Ministry for Health and Family Welfare, no date) (Thakur et al., 2020) Aflatoxins are organic compounds that, in minute amounts, can be harmful to humans and are generated by Molds and fungus. Aflatoxin-producing fungi are commonly found in nature and can infect plants. The most dangerous and poisonous forms of aflatoxin include B1 and B2, as well as G1, G2. This test was run to determine how much aflatoxin was present in the medication.

4.8.3 Pesticide residue (AYUSH, 2008)

Medicinal plant materials can harbor pesticide residues due to various agricultural practices, including spraying, soil treatment during cultivation, and the use of fumigants during storage. Therefore, the presence of these pesticides will be analyzed using GC-MS and the results will be compared to the standards set by the Ayurvedic Pharmacopoeia of India.

4.9 HPTLC analysis for male and female *Guduchi* (Khatoon et al., 2018)

(Priyanka Patil, 2016)

4.9.1 Preparation of Standard Solution

In a 2 ml volumetric flask, 2 mg of standard berberine was precisely weighed. The solution is sonicated after adding 1ml of methanol to the standard. After complete dissolution of standard, the volume is made up to 2ml using methanol as a solvent. This standard solution is used for fingerprinting on HPTLC plates.

4.9.2 Preparation of the test solution using male *Guduchi* (*Tinospora cordifolia*).

weighed samples of male *Guduchi* (*Tinospora cordifolia*). 100 mg of each sample is taken in a 250ml volumetric flask. The solution is mixed with 10ml of chloroform and methanol (9:1 v/v). the solution is cooled, filtered using Whatman filter paper and concentrated in an evaporating dish to 5ml on a water bath after reflux heating in a flask on a water bath for 30 minutes. This concentrate is poured in a volumetric flask. The test solution is used for fingerprinting in HPTLC procedure.

4.9.3 Preparation of the test solution using Female *Guduchi* (*Tinospora cordifolia*)

Weighed samples of female *Guduchi* (*Tinospora cordifolia*). 100 mg of each sample is taken in a 250ml volumetric flask. The solution is mixed with 10ml of chloroform and methanol (9:1 v/v). The solution is cooled, filtered using Whatman filter paper and concentrated in an evaporating dish to 5ml on a water bath after reflux heating in a flask on a water bath for 30 minutes. This concentrate is poured in a volumetric flask. The test solution is used for fingerprinting in HPTLC procedure.

Table 4.1: Chromatographic details of HPTLC of Male and Female *Guduchi* with the standard

Application mode	CAMAG Lino mat 5- Applicator
Applicator filtering system	Whatman filter paper no.1
Stationary phase	MERCK-TLC/HPTLC silica gel 60 F254 on aluminium sheets
Application (Y axis) start position	10mm
Development end position	75mm from plate base
Standard application volume	5.0 µL
Band length	6mm
Sample application volume	5.0 µL (Concentrate of male and female *Guduchi*)
Distance between tracks	15mm
Development mode	CAMAG TLC Twin Trough Chamber
Chamber saturation time	30 minutes
Mobile Phase (MP)	Chloroform: Methanol (9:1)
Visualization	@ 366nm for Berberine
Drying Mode, Temperature, and Duration	TLC plates were heated for three minutes at 100 ± 50°C in a prepared TLC plate heater.

4.10 Physicochemical standardization of *Sanshamni Vati*

Physicochemical standardization of the herbal extracts was performed according to the parameters and their procedures mentioned from section 4.6 to section 4.9.

4.11 Preparation of *Sanshamni Vati*

4.11.1. Cleaning of Authenticated *Guduchi* samples

The authenticated samples of *Guduchi* were cleaned. Only the stem part of *Guduchi* was taken. Circumference of the stem was measured with the help of a vernier caliper and those having a girth between 1.5-1.20 cm were taken. The stem washed and dried.

4.11.2 Size reduction

The cleaned stem was reduced into pieces of 3-4 inches long having a girth of 1.5 to

2.0 cm in diameter. These pieces are subjected to mixer grinder for crushing and reduced coarsely. The crushed stem was again weighed, and batches were made of 5kg each for further processing.

4.11.3 Overnight soaking

Six batches male and Six female *Guduchi* stem, 5kg of each stem were taken. Each sample was soaked in 20 liters of potable water in a container of 40 liters capacity for 18 hours i.e., 4pm to 8am at room temperature.

4.11.4 Heating and stirring for preparation of *Kwatha*

After overnight soaking the container along with soaked *Guduchi* subjected to heating on gas Bhatti on controlled temperature. The temperature is maintained between 95°C -100 °C. When the water is reduced to 1/4th the *Kwatha* is subjected to filtration with four- fold cotton cloth. The *Kwatha* obtained was brownish green, sticky liquid. The *Kwatha* was measured with 1 liter measuring cylinder and recorded in table (4.1). The evaluation of the *Kwatha* for organoleptic and physicochemical characters was recorded in table 4.2

4.11.5 Filtered *Kwatha* is further subjected to heat

The filtered *Kwatha* is subjected to container of 8 liters capacity and again heated at the temperature of 90°C -95°C and when semisolid form was achieved, container is removed from gas stove and the material was subjected to enamel tray for further drying. Observations of preparation were recorded in table (4.3).

4.11.6 Hot air oven drying

The glass tray having semisolid *Guduchi Kwatha* was placed in the hot air oven at 45°C centigrade for the preparation of *Guduchi Ghana* (*Sanshamni Vati*). After evaporation for 4 hours, the blackish brown mass is obtained. The evaluation of Ghana was recorded in table (4.4) When the material is completely dried, rolled into the 250 mg Vati.

4.12 Characterization of *Sanshamni Vati*

During the manufacture of *Sanshamni Vati*, the following quality control procedures are used to guarantee uniformity and effectiveness:

Appearance

- ***Colour*:** Depending on the herbal extract's concentration and any added additives, it might range from brownish to dark brown. The concentrated herbal extract, consistently ranging from brownish to dark brown.

- ***Taste*:** It is important to consider as an organoleptic feature.
- ***Texture*:** The surface is uniformly smooth and free of any obvious flaws or fissures.
- ***Consistency*:** The tablet maintains its shape when handled and stored since it is firm and compact.
- ***Weight Uniformity*:** Depending on the precise formulation and desired dose, each Vati should have a constant weight, 250 mg.
- ***Moisture level Testing*:** To guarantee the durability of the *Vati* and inhibit microbiological development, the moisture level should be low. (Patgiri et al., 2012)
- ***Hardness:*** (A.K Gupta, 2004), (Nagar, 1956), Usually determined with a tablet hardness tester, tablets should be sufficiently hard to endure mechanical stress during handling, packing, and transportation.
- ***Friability*:** Low friability is necessary to prevent the tablets from crumbling or breaking readily. Using a friabilator, which subjects' tablets to mechanical stress and measures weight loss, this is quantified.

- ***Disintegration Time*:** The tablet must dissolve in an aqueous environment at body temperature(37°C) in a specified amount of time (usually 15–30 minutes). A disintegration tester is used to measure the disintegration time to make sure the tablet releases its active ingredients efficiently. Quality of Raw Material Before processing, the identification, potency, and purity of the raw *Guduchi* plant is confirmed. Consistency in Batch: Consistency in weight, hardness, moisture content, and disintegration time is ensured by routinely sampling and testing vati from various batches.

4.13 In- vivo Study for evaluation of *Sanshamni Vati* on rat paw edema (Tiwari et al., 2018)

4.13.1 Healthy albino Wistar rats of either sex, weighing 195 ± 10g, were chosen for the study. The rats were maintained under a 12:12 h light-dark cycle at a temperature range of 18 to 20°C. Throughout the experimental period, they were housed in spacious, hygienic cages. The animals had unrestricted access to standard pellet diet and water

until the conclusion of the study. Animals were divided into 5 groups (n = 6/group). Group I was kept as normal untreated control (5ml/kg saline), group II received Carrageenan 0.1 ml of 2% (w/v) along with saline, group III received standard drug indomethacin (10 mg/kg) administered intraperitoneally 1 h before carrageenan suspension administration, group IV received sample A (500 mg/kg) orally for 10 days, group V received sample B (500 mg/kg) orally for 10 days. 60 min. before inducing inflammation, the final dose was administered. Following this, all animals received a subcutaneous injection of 0.1 ml of 1% (w/v) carrageenan solution into the plantar region of the right hind paw to induce edema. Paw volume was measured at 1, 2, 3, and 4-hour intervals after the injection using a Vernier calliper. (Halici et al., 2007) (Rauf et al., 2022)

4.13.2 Evaluation parameters for inflammatory activity (Shaikh, R.U., Pund, M.M. and Gauche, 2016) (Patgiri et al., 2014)

4.13.3 Hematological Parameters (Sharma and Pandey, 2010)

Blood parameters were quantified using an automatic hematological assay analyzer.

4.13.4 Cytokine analysis (Aranha, Clement and Venkatesh, 2012)

Blood was drawn from the retro-orbital sinus of experimental animals and transferred to Eppendorf Micro-centrifuge tubes. These tubes were promptly placed in a cooling microcentrifuge and centrifuged at 7000 rpm at 4°C for 15 minutes to obtain clear serum. The serum obtained was then transferred to fresh, sterilized Eppendorf Micro-centrifuge tubes and stored in a deep freezer, shielded from artificial and sunlight exposure, for biochemical parameter studies. The serum concentrations of IL-6 and TNFα were quantified using rat-specific ELISA kits, and absorbance readings were taken using an ELISA microplate reader. (Erba).

4.14 Histopathology

On the final day of the experiment, cross-sectional full-thickness paw specimens were collected for histopathological examination. During tissue collection for histology, skin tissue pieces were cut, washed, and transferred into 10% formalin solution. After fixation, the pieces were dehydrated in absolute alcohol, followed by a mixture of alcohol and xylene (1:1) for 15-20 minutes. Subsequently, the tissue pieces underwent a second embedding process and were kept at a controlled temperature. Filtered matured wax was poured into the lid up to 4/5th of its height. The tissues were then

gently placed in the wax and allowed to solidify at room temperature. Using a microtome, the wax block was cut into ribbon-like sections. The sections on slides were dewaxed with xylene, stained with aqueous hematoxylin, carefully mounted with Canada balsam under cover slips, and examined under a microscope.

4.15 Statistical Analysis

Results are provided as Mean ± SD (n=6). Results were analyzed statistically using one-way analysis of variance (ANOVA) followed by Bonferroni t-test. $P < 0.05$ was considered as level of significance while comparison between groups.

CHAPTER 5 new

RESULT AND DISCUSSION

5.1 Collection of Drug:

The initial and crucial step in evaluating drugs is the collection process. The locations from which the drugs are sourced were carefully selected to ensure they are clean, well-maintained, and free from pollutants. Fresh stem of both male and female plants of *Guduchi* (*Tinosphora. cordifolia*) was collected from the Herbal Garden of Dayanand Ayurvedic College in Jalandhar and The Nursery of the Horticulture Department Punjab, located at Cantonment Road near the BSF campus, Jalandhar (Pb).

During the collection process, several important considerations were taken into account:

Timing of Collection: Plants were harvested at the optimal time to ensure maximum potency and efficacy of the active compounds.

Plant Maturity: Only mature plants were selected to ensure the presence of fully developed phytochemicals.

Environmental Conditions: The plants were collected during favorable weather conditions to prevent any deterioration of the plant material.

Avoidance of Contaminants: Care was taken to avoid any contamination from soil, water, or air pollutants.

5.2 Authentication of Drug

The collected stems of both genders were authenticated as Guduchi plant by the Government Accredited Laboratory, HHRC (Herbal Health Research Consortium Pvt. Ltd., Amritsar). The results are discussed below. Assistance for identification and authentication was sought from various official databases and floras.

5.3 Organoleptic Evaluation

The evaluation of the drug is based on its color, odor, and texture. The organoleptic properties of the raw drug were examined in accordance with A.P.I. standards, and the findings are presented in Table 5.1.

The organoleptic characteristics of the raw *Guduchi* (T. cordifolia) stems, including sha pe, texture, color, odor, and taste, were meticulously studied for both male and female plants. While the shape and texture of the stems were found to be similar for both genders, notable differences were observed in their color, odor, and taste.

The male stem exhibits a light green color, contrasting with the dark green color of the female stem. When it comes to odor, the male stem has a mild bitter smell, especially noticeable after the outer covering is removed. In contrast, the female stem emits a relatively stronger and more pronounced odor.

Taste-wise, the male stem is distinctly bitter. The female stem, however, is even more bitter, emphasizing the differential features between the two. These organoleptic properties are critical in the evaluation process, as they contribute to the overall assessment of the drug's quality and potential efficacy.

Although the surfaces, forms, and textures of the male and female plant stems are similar, there are some significant differences. More ridges, less succulent texture, and lower mucilage production are found on the male stem. Along with a harsh flavor and smell, it has a pale green exterior covering. Alternatively, the female stem has a stronger scent, more mucilage, a more bitter flavor, and is more succulent and darker green. Their general structures are similar; however, the female stem is typically softer and moister.

Microscopically the stem of the Plant was observed. Results are discussed below in Table 5.2

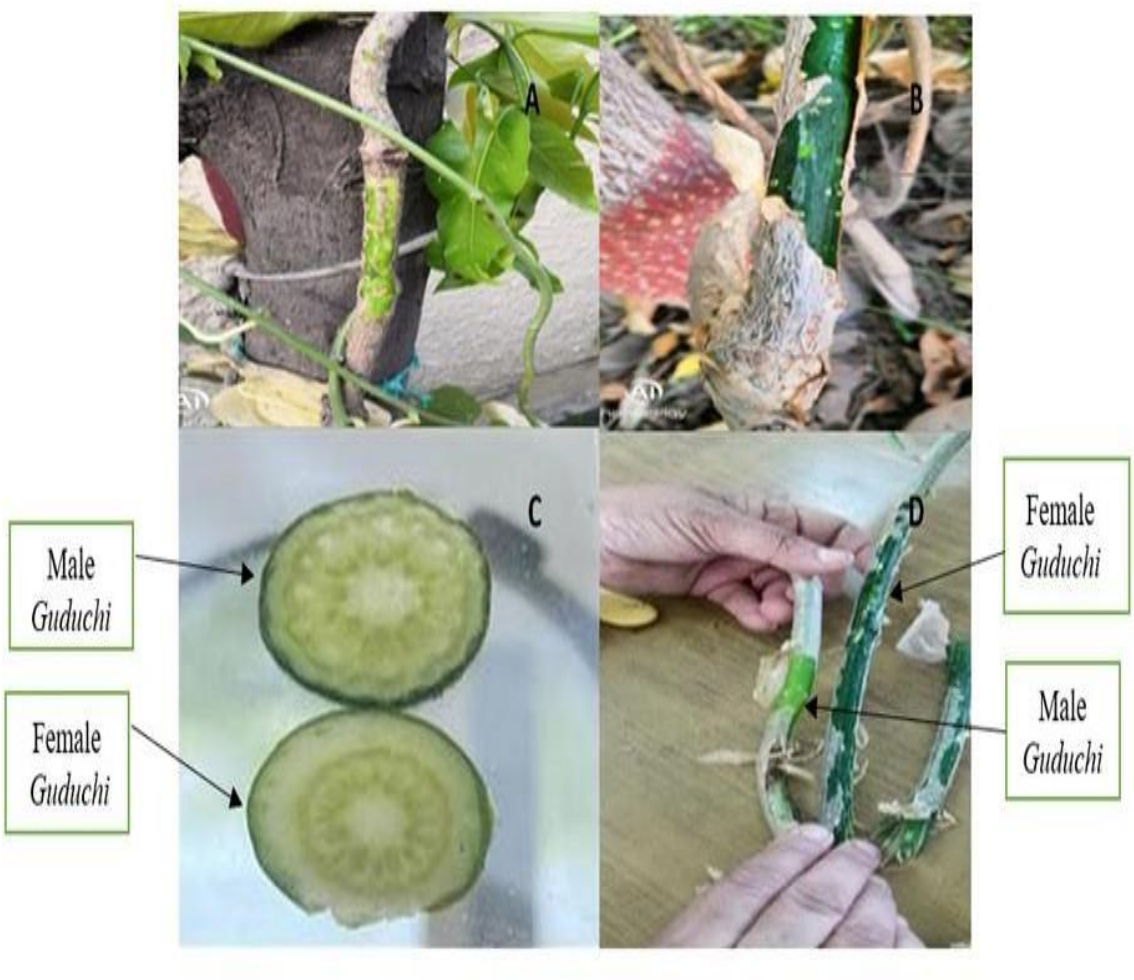

Fig: 5.1 Microscopy of *Guduchi* Stem

Table 5.1: Organoleptic features of Guduchi stem

Characters	Stem of Male plant	Stem of Female plant
The General Appearance	Possessing a warty feel tubercle o ginatin from the formation of vertical and longitudinal lenticels, succulent and ridged, featuring distinctive three beaked nodal swellings.	Similar to the male plant but more succulent and less flaky.
Texture	Ridged, irregular, continues, running ith lenticels	Similar
Shape	Slim rotates towards right side ombined twin, swelling at nodes	Similar
Surface	Sharp with longitudinal fissures ong the rows of lenticels.	Similar.
Mucilage	Less.	More.
Touch	Rough.	Similar.
Color	Light-green outer layer cr eamish-hite.	Dark green outer layer creamish-white.
Odor	Bitter smell after removal of bark of em	Relatively strong smell.
Taste	Bitter	More Bitter

Table 5.2: Anatomical differences between male and female plant of *Tinospora Cordifolia*

S.No.	Characteristics	*Guduchi* Male	*Guduchi* Female
1.	**Hypodermis**	Collenchyma Tous cell having more angular Thickness	Collenchyma Tous cells less angular thickness
2.	**Cortical region**	highly broad (Around. 2600-3000 µm)	Less broad (Around. 1800-2400 µm)
3.	**Tannin**	less in number (1-3/mm2)	More in number (5- 10/mm2), scattered in cortical, phloem and pith regions
4.	**Mucilaginous canals**	Less (2-6 /mm2)	Comparatively much more (6-13/mm2)
5.	**Starch grain**	Up to 16 µm in size, less in amount	Up to 2µm in size, more in amount

These changes suggest that the female *Guduchi* is more adapted for storing nutrients and maintaining moisture because it has more mucilaginous canals and a higher tannin content. On the other hand, the male *Guduchi*'s broader cortical region and larger starch grains might be responsible for its structural resilience and greater short-term energy reserves. Hypodermal structure and cortical area size variations reflect the many roles that each plant may play in its environment or in its reproductive strategies.

The Guduchi sample microscopy images highlight various anatomical features. Section A shows a star-shaped central vascular structure with a thick outer layer, likely representing the epidermis or cortex. Section B presents a similar arrangement with distinct, red-stained internal tissues. Section C depicts the clearest view of vascular bundles, enhanced by reddish-purple staining for clear tissue differentiation. Section D shows a partial cross-section with a red tint, focusing on vascular arrangement on one side.

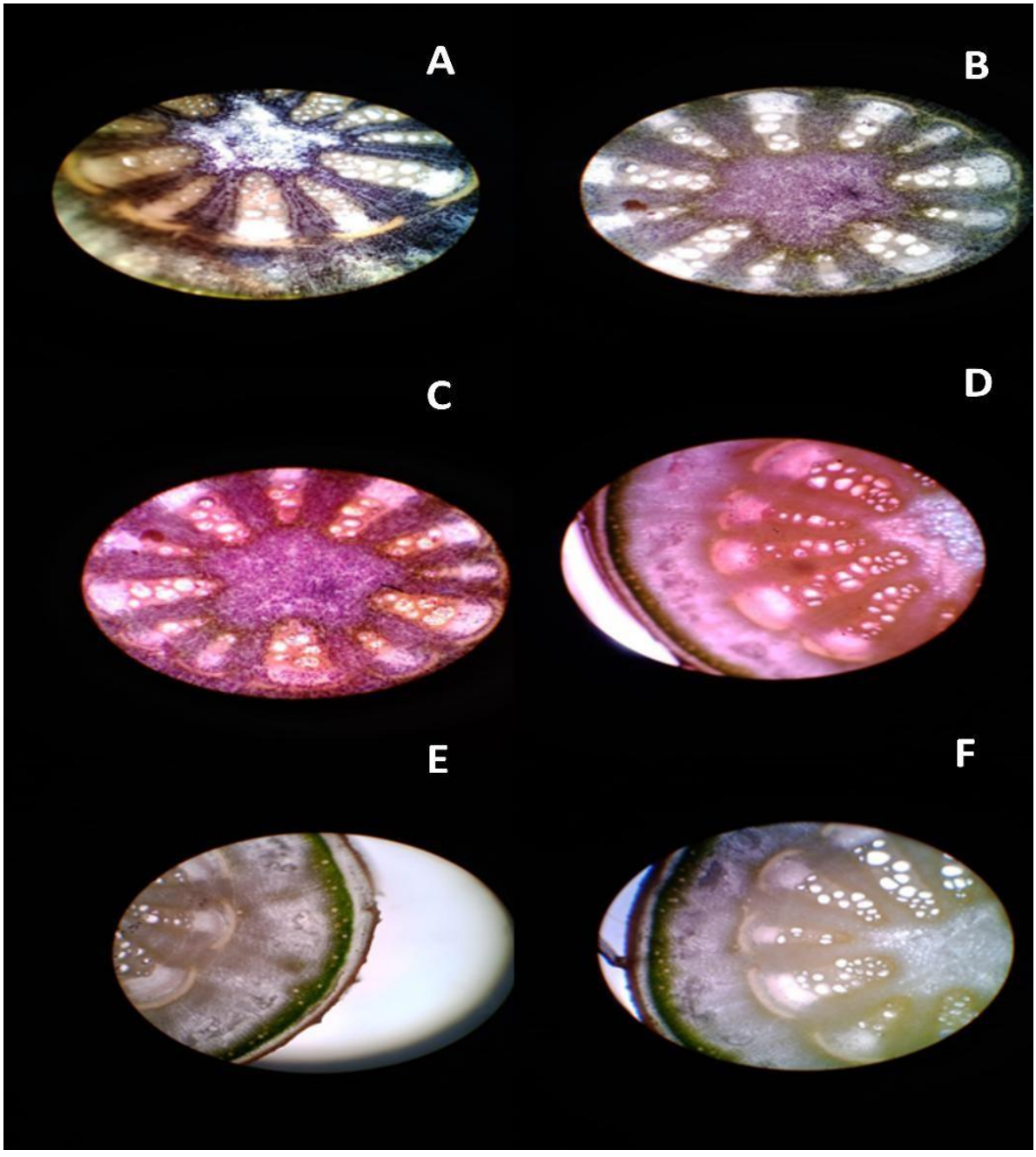

Fig:5.2 Microscopic View of *Guduchi* Stem Female (A, C, F) and Male (B, D, E).

Section E features a more natural tone, revealing both internal and external structures, including a visible green outer layer. Section F is lighter but maintains the central star-shaped structure with less defined outer tissues.

5.4 Physicochemical Analysis

Physicochemical analysis includes the comparison between male and female *Guduchi* plant. The Analysis includes various parameters, the result of all the parameters is depicted below in the Table 5.3.

The physicochemical parameters of *Guduchi* (T. cordifolia) were evaluated according to the procedures outlined by the A.P.I., with results falling within the prescribed limits specified in the monograph of *Guduchi*, as detailed in Table 5.3. Six batches were prepared and analyzed, with the standard deviation of the results also presented in Table 5.3. To ensure the quality of the collected samples, the physicochemical parameters were tested three times for both male and female *Guduchi* stems. No foreign matter was observed in any of the samples. The average values obtained for Loss on Drying (LOD), total ash, acid-insoluble ash, sulphated ash, water-soluble extractive, and alcohol-soluble extractive are as follows:

Male Guduchi Sample: 4.04%, 6.81%, 0.93%, 9.79%, 18.36%, 8.71%.

Female Guduchi Sample: 5.20%, 9.81%, 1.94%, 11.64%, 18.35%, 7.68%

The comparison of the results indicates that while both male and female samples adhere to the prescribed limits, there are distinct differences between the two. The female *Guduchi* samples generally exhibited higher values for LOD, total ash, acid-insoluble ash, and sulphated ash compared to the male samples. However, the water-soluble extractive content was nearly identical between the two, with the male samples having a slightly higher alcohol-soluble extractive content. These results, consistent with the A.P.I. standards, confirm the quality and authenticity of the collected *Guduchi* samples, as shown in Table 5.3 &5.4.

The physicochemical analysis of Male *Guduchi* stem indicates that key parameters such as Loss on Drying, Total Ash, Acid Insoluble Ash, Alcohol Soluble Extractive, and Water-Soluble Extractive are all within the specified limits, highlighting the batch's high quality and purity. The Sulphated Ash levels suggest the presence of valuable inorganic content, aligning closely with standard thresholds. The batch shows stability, rich extractive content, and minimal contamination, making it highly suitable for use.

The physicochemical analysis of female Guduchi stem demonstrates that parameters, including loss on drying, total ash, acid insoluble ash, alcohol soluble extractive and water-soluble extractive are within the specified standards, underscoring the batch’s

high quality and consistency. The loss of drying values indicates optimal moisture content, while the total ash and acid insoluble ash values highlight appropriate levels of inorganic and non-soluble mineral content

Table 5.3 Physicochemical analysis of Male *Guduchi*

Parameters	**Batch(M)**						**Std. Div.**	**Standard**
	I	**II**	**III**	**IV**	**V**	**VI**		
Loss on Drying (%w/w)	4.06	4.04	4.85	4.97	4.83	4.04	4.46± 0.42	NMT 10
Total Ash(%w/w)	6.88	7.10	7.97	8.65	6.88	6.81	7.38 ± 0.69	NMT 16
Acid insoluble Ash(%w/w)	1.15	1.45	0.94	0.90	1.36	0.93	1.12 ± 0.21	NMT 3
Alcohol Soluble Extractive(%w/w)	8.73	8.66	7.90	8.71	7.99	8.70	8.44 ± 0.35	NLT 3
Water Soluble Extractive(%w/w)	18.33	18.39	18.19	18.45	18.02	18.36	18.29±0.14	NMT 11
Foreign Matter	-	-	-	-	-	-	-	Nil
Sulphated Ash (%)	10.35	10.15	9.89	11.00	11.57	9.97	10.48±0.60	NMT 9.81

Table 5.4 Physicochemical analysis of Female *Guduchi*

Parameters	**Batch (FM)**						**Std. Div.**	**Standard**
	I	**II**	**III**	**IV**	**V**	**VI**		
Loss on Drying (%w/w)	4.84	4.70	5.00	5.18	5.05	5.20	4.9± 0.17	NMT 10
Total Ash(%w/w)	8.89	7.45	7.94	8.06	8.15	9.81	8.3±0.76	NMT 16
Acid insoluble Ash(%w/w)	1.00	1.92	1.93	0.99	1.78	1.94	1.5±0.42	NMT 3
Alcohol Soluble Extractive(%w/w)	7.70	7.66	7.00	7.10	7.20	7.68	7.3±0.29	NLT 3
Water Soluble Extractive(%w/w)	18.32	18.33	18.00	17.95	18.85	18.35	18.3±0.2	NMT 11
Foreign Matter	-	-	-	-	-	-	-	Nil
Sulphated Ash (%)	10.36	11.50	11.03	10.01	10.59	11.64	10.8±0.59	NMT 9.81

The Water and Alcohol Soluble Extractive values suggest a significant presence of active constituents. The Sulphated Ash content shows a suitable balance of inorganic compounds, within expected standards. Female *Guduchi* stem shows minimal impurities, stability, and purity.

5.5 Phytochemical analysis

Phytochemical analysis was conducted to assess the quality of the sample by verifying the presence of all its phytoconstituents. Various chemicals were used to detect these phytoconstituents through color changes and precipitation reactions. The stem of *Guduchi* was found to contain several pharmaceutically important phytochemicals, such as alkaloids, tannins, and flavonoids, glycosides, and saponin, reducing sugar etc. A phytochemical analysis of *Guduchi* male reveals a wide variety of bioactive compounds, including terpenoids, alkaloids, saponins, and reducing sugars, which together account for the plant's widely recognized medicinal properties. The plant's stable phytochemical profile is supported by the consistent results from all batches, demonstrating its utility for improving immunity, reducing inflammation, and boosting overall health in both traditional medicine and modern herbal formulations. Although flavonoids are not common in medicinal plants, *Guduchi*'s potential for improving health seems to be fueled by other potent bioactive ingredients, so the absence of flavonoids does not lessen its potential for therapeutic effects

Table 5.5. - Phyto-Chemical analysis of *Guduchi* stem

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S.no.	Qualitative Parameter	Analytical Test	*Guduchi* Male	*Guduchi* Female	Standard
1.	Tannins	Lead Acetate Test	+	+	Reddish Precipitates appears.
2.	Alkaloids	Dragendroff's test	+	+	Orange brown ppt
3.	Carbohydrates	Molish's test	+	+	Violet ring is formed
4.	Flavonoids	Shinoda Test	-	-	Yellow Ppts
5.	Reducing Sugar	Benedict's Test	+	+	Sol. turns green
6.	Starch	Iodine Test	+	+	Green Sol turns dark Blue
7.	Proteins	Biuret Test	+	+	Dark Purple Sol
8.	Amino acids	Ninhydrin Test	+	+	Solution dark turns Purplish coloured.
9.	Steroid	Salwoski Test	+	+	Upper H2So4 Layer appear yellow green
10	Saponin	Foam Test	+	+	Persistent Foam
11	Cardiac Glycosides	Keller Killani Test	+	+	Solution turns Reddish Brown

+ve =present, -ve =absent

Table 5.6. - Phyto-Chemical analysis of *Guduchi* Male

Compound	Test Performed	Result (Batch M)					
		I	II	III	IV	V	VI
Tannin	Lead Acetate Test	+	+	+	+	+	+
Alkaloids	Dragendroff's reagent	+	+	+	+	+	+
Carbohydrates	Molish's test	+	+	+	+	+	+
Flavonoids	Shinoda Test	-	-	-	-	-	-
Reducing Sugar	Benedict's Test	+	+	+	+	+	+
Starch	Iodine Test	+	+	+	+	+	+
Proteins	Biuret Test	+	+	+	+	+	+
Amino acids	Ninhydrin Test	+	+	+	+	+	+
Steroid	Salwoski Test	+	+	+	+	+	+
Saponin	Foam Test	+	+	+	+	+	+
Cardiac Glycosides	Keller Killani Test	+	+	+	+	+	+

Consistent results showed that important chemical components such proteins, hexose sugars, starch, reducing sugars, alkaloids, terpenoids, amino acids, and steroids were found in all six batches of the molecule.

These results suggest that the chemical has a considerable deal of potential for use in nutrition and medicine because of its high protein and carbohydrate content and abundance of bioactive chemicals such terpenoids, alkaloids, and steroid. The compound's antioxidant potential was limited due to the lack of flavonoid glycosides

Table 5.7.- Phyto-Chemical analysis of *Guduchi* Female

Compound	**Test Performed**	**Result (Batch M)**					
		I	**II**	**III**	**IV**	**V**	**VI**
Tannin	Lead Acetate Test	+	+	+	+	+	+
Alkaloids	Dragendroff's reagent	+	+	+	+	+	+
Carbohydrates	Molish's test	+	+	+	+	+	+
Flavonoids	Shinoda Test	-	-	-	-	-	-
Reducing Sugar	Benedict's Test	+	+	+	+	+	+
Starch	Iodine Test	+	+	+	+	+	+
Proteins	Biuret Test	+	+	+	+	+	+
Amino acids	Ninhydrin Test	+	+	+	+	+	+
Steroid	Salwoski Test	+	+	+	+	+	+
Saponin	Foam Test	+	+	+	+	+	+
Cardiac Glycosides	Keller Killani Test	+	+	+	+	+	+

However, because of its chemical composition specifically, the presence of terpenoids, alkaloids, and saponins there is a broad range of possible pharmacological effects that have been discovered. A detailed assessment and analysis of these properties was necessary to determine the compound's potential use in nutrition, medicine, and as a functional component of products connected to health.

Table 5.8. Microbial parameters & Limit tests for Male and Female *Guduchi*:

Parameters	Required amount	Male *Guduchi*	Female *Guduchi*
Total microbial plate count	1X105cfu/g	500cfu/g	400cfu/g
Total yeast and moulds	1X103cfu/g	80cfu/g	60cfu/g

The counts for the microbiological plate of Male &Female Guduchi were 500cfu/g & the 400cfu/g total counts for yeast and mold were 80cfu/g& 60cfu/g both of which are within the recommended limits outlined in the monograph. The official monographs defined range is met by the *Guduchi* stem. The outcome is displayed in Table No.5.8.

Table No. 5.9 Specific pathogens parameters & Limit tests for Male and Female *Guduchi*

Parameters	Required amount	Male *Guduchi*	Female *Guduchi*
Escherichia coli	-ve	-ve	-ve
Staphylococcus aureus	-ve	-ve	-ve
Pseudomonas aruginosa	-ve	-ve	-ve
Salmonella typhimuriu	-ve	-ve	-ve

Plant diseases are caused by pathogens, so the primary criterion for examining any plant illness was the results of the pathogen test. Gram +ve (*Staphylococcus aureus*) and gram -ve (*Escherichia coli*, *Pseudomonas aeruginosa*) bacteria were cultivated by the Agar well diffusion technique. There was no evidence of pathogen development in the *Guduchi* stem of both genders. The outcome is displayed in table 5.9

Table 5.10. Aflatoxins parameters & Limit tests for Male and Female:

Parameters	Required amount	Male *Guduchi*	*Guduchi* Female *Guduchi*
B1 & G1	NMT 0.5 ppm	ND	ND
B2 & G2	NMT 0.1 ppm	ND	ND
Pesticide residue	Absent	ND	ND

The most dangerous and poisonous forms of aflatoxin include B1 and B2, as well as G1, G2. T was not detected; Results are shown in Table.5.1

Table 5.11. Heavy metal analysis parameters & Limit tests for Male and Female *Guduchi*

Heavy metal	Required amount	Male *Guduchi*	Female *Guduchi*
Arsenic	3 ppm	ND	ND
Cadmium	0.3ppm	ND	ND
Mercury	1 ppm	ND	ND
Lead	10 ppm	ND	ND

The range of the stem of *Guduchi* of both samples is within the recommended limit and it was confirmed to be free of heavy metals. (*-ve Absent, *ND- Not detected)

5.6. Preparation and Evaluation Parameters of *Sanshamni Vati*

Preparation and evaluation of *Sanshamni Vati* includes the following steps.

5.6.1Preparation of *Guduchi Kwatha*

To prepare *Sanshamni Vati*, the ratio of *Guduchi* to water was maintained at 1:4 for each batch of the *Kwatha* to ensure a higher yield of Ghana. A total of six batches of *Guduchi Kwatha* were prepared for each gender. The fresh *Guduchi* stems were cut into uniform sizes, crushed using an *Udukhal Yantra*, and then transferred to a container for the *Kwatha* process. The quantities of ingredients used for the preparation of *Kwatha* are detailed in Table 5.12.

Guduchi pieces are taken diameter is almost 1.6 to 2.4. The ratio of *Guduchi* and water was 1:4 for each batch of *Kwatha*. Soaking time was 18 hours (4pm to 8am) other details are mentioned in table5.6. The coarse *Guduchi* was soaked for 18 hours and subjected to heat. Continuous stirring was done during the preparation of *Kawath* to get uniform concentration throughout the solvent. The temperature was maintained between 90-95 degree centigrade. The first batch took 5 hours to get reduced to 1/4th the average yield for all batches 5.15 litres (FM) and 5.05(M).

Table 5.12. Details of the preparation *Guduchi Kawath* in different batches

Batch No.	Weight of the *Guduchi* taken (Kg.)	Total quantity of water used (L)	Soaking time (hrs.)	Temperature Degree centigrade	*Kwatha* obtained (L)
1 FM	5	20	18	95	5.100
2 FM	5	20	18	95	5.150
3 FM	5	20	18	95	5.200
4 FM	5	20	18	95	5.100
5 FM	5	20	18	95	5.150
6 FM	5	20	18	95	5.200
Average	5	20	18	95	5.15
1 M	5	20	18	90-95	5.050
2 M	5	20	18	90-95	5.000
3 M	5	20	18	90-95	5.100
4 M	5	20	18	90-95	5.050
5 M	5	20	18	90-95	5.000
6 M	5	20	18	90-95	5.100
Average	5	20	18	90-95	5.05

The data shows that the extraction method was highly consistent, with just little differences in *Kwath* yield across batches FM and M. Though the difference is modest, the somewhat higher yield from FM batches may indicate that the female plant material is more effective in releasing extractable chemicals. Both Male and Female plant components behave consistently, in the case of the amount of *Kawath* that was created, suggesting that both could be utilized in decoction procedures under careful supervision. Additional investigation, such as *Kawath* chemical profiling, may shed

more light on the particular chemicals removed and any qualitative variations between the male and female *Guduchi*.

5.6.2Evaluation of *Guduchi Kwatha*:

Kwatha of each batch was evaluated by using organoleptic characters and physico-chemical parameters. The results of all the batches for different parameters were mentioned in Table 5.13.

Table 5.13. Evaluation parameters of *Guduchi Kwatha* (FM)

Sr. No.	Parameter	Batch						Std. Dev.
		1	2	3	4	5	6	
1.	Ph	6.41	6.00	6.30	6.40	6.32	6.35	6.29±0.13
2.	TSC (%)	6.25	5.78	6.15	6.13	5.9	5.6	5.96±0.22
3.	Viscosity	0.97	0.97	0.97	0.97	0.97	0.97	0.97±0
4.	Specific gravity	1.015	1.020	1.025	1.020	1.030	1.025	1.02±0.004
5.	Refractive Index	0.36	0.36	0.36	0.36	0.36	0.36	0.36+0.00

TSC- Total Solid Content

Overall support and consistency in the results are demonstrated by the limits computed for the Female *Guduchi* clusters. While stickiness remnants are typically consistent across all lots, there are some slight variations in the pH, TSC, refractive index, and

distinguishing importance. Together with the logical tangible and synthetic features seen, these findings plan that the distillation and readiness processes for Female *Guduchi* are reliable and repeatable. With every tiny variation in viscosity and refractive index, the precision of the calculations and the safety of the distillation process are highlighted.

Table no. 5.14. Evaluation parameters of *Guduchi Kwatha* (M):

Sr. No.	Parameters	Batch						Std. Dev.
		1	2	3	4	5	6	
1.	pH	6.41	6.00	6.30	6.40	6.32	6.35	6.29±0.13
2.	Total solid content (%)	6.25	5.78	6.15	6.13	5.9	5.6	5.96±0.22
3.	Viscosity	0.96	0.96	0.96	0.96	0.96	0.96	0.96±0
4.	Specific gravity	1.020	1.025	1.025	1.015	1.020	1.020	1.02±0.003
5.	Refractive Index	0.37	0.37	0.37	0.37	0.37	0.37	0.37±5

Overall support and consistency in the results are demonstrated by the limits computed for the Female Guduchi clusters. While stickiness remnants are typically consistent across all lots, there are some slight variations in the pH, TSC, refractive index, and distinguishing importance. Together with the logical tangible and synthetic features seen, these findings plan that the distillation and readiness processes for Male Guduchi are reliable and repeatable. With every tiny variation in viscosity and refractive index, the precision of the calculations and the safety of the distillation process are highlighted.

Table no 5.15. Organoleptic Properties of Guduchi Kwath (Male and Female)

Sr. No.	Parameter	Guduchi Kwath(M&F)
1	Color	Brownish- green
2	Oduor	Characteristic
3	Taste	Bitter

Both the male and female *Guduchi Kwath*a have similar sensory qualities, including a brownish-green color, a distinctive smell, and an unpleasant taste. These characteristics are typical of *Guduchi* extracts and reflect their inherent plant-based qualities.

5.6.3. Observation of *Guduchi Kwatha*:

The observed result for *Kwatha* prepared from Male and Female *Guduchi* is mentioned below in the Table 5.16

Table 5.16 Observations during process of *Guduchi Kwatha*

Sr. No.	*Guduchi Kwatha* (M)	*Guduchi Kwatha* (FM)
1	Evaporation commenced at 70°C and intensified with stirring.	Evaporation commenced at 70°C and intensified with stirring.
2	After two hours white color layer was appearing on liquid	After two hours white color layer was appearing on liquid
3	In the initial phase of heating heavy foaming was observed.	In the initial phase of heating heavy and dense foaming was observed.
4	The pH of *Kwatha* was 6.29 average	The pH of *Kwatha* was 6.29 average
5	Sticky nature of *kwatha* was observed	Sticky nature of *Kwatha* was observed
6	Time taken for preparation of *Kwatha* was 8.0 hours.	Time taken for Preparation of *Kwatha* was 8.0 hours
7	Quantity of Kwatha was obtained 5.15 L	Quantity of *Kwatha* was obtained 5.05L
8	In process Temp was 95°C	In process Temp 95°C

During the preparation of *Kwatha* not much difference is observed between male and

female plant, both have same pH i.e., 6.29.

5.7 Preparation of *Guduchi Ghana*

Ghana was prepared in 6 batches for both the plants. Detail of different batches is mentioned below in the Table 5.17.

Table 5.17. Details of preparation of *Guduchi Ghana* in different batches

Batch No.	***Kwatha* taken (liters)**	**Quantity of Semisolid *Ghana* (gm)**	**%age Yield of Semisolid *Ghana***	**Temperature in Hot air oven for drying (°)**	**Quantity of dried *Ghana* (gm.)**
1 FM	5.100	800	15.68	45	320
2 FM	5.150	750	14.56	45	298
3 FM	5.200	700	13.46	45	290
4FM	5.000	750	15.00	45	295
5FM	5.120	825	16.11	45	320
6FM	5.170	700	13.53	45	295
Std. Dev.	5.12±0.06	754.16±46.58	14.72±0.99	----	300±12.24
1 M	5.050	770	15.24	45	290
2 M	5.000	795	15.90	45	295
3 M	5.100	680	13.33	45	286
4M	5.030	770	15,30	45	290
5M	5.020	750	14.94	45	285
6M	5.170	700	13.53	45	280
Std. Dev.	5.06±0.06	744.17±1.00	14.58±0.00	--	287.66±4.71

The *Kwatha* was further boiled for the evaporation of water to get the solid content. The obtained solid content when stirred continuously throughout the process to avoid burning, a brownish green semi solid mass was obtained. This was transferred to

enamel tray for further evaporation in hot air oven to get a solid content. Average yield of all the batches was 312grams (FM) and 287grams (M).

Table 5.18. Observation of prepared *Guduchi Ghana*

S. No.	Male Guduchi Ghana	Female Guduchi Ghana
1	The Liquid brownish green in bitter in taste	The Liquid brownish green in bitter in taste
2	Bitter in taste	Bitter in taste
3	Kwatha was observed sticky in nature after 3.5 hours	*Kwatha* was observed sticky in nature after 3 hours
4	The liquid was stickier and stick to stirrer and vessel After 4 hours	The liquid was stickier and stick to stirrer and vessel After 4 hours
5	Brownish green semi solid mass was obtained	Brownish green semi solid mass was obtained
6	After further evaporation in hot air oven it was a solid content, black in color	After further evaporation in hot air oven it was a solid content, black in color

The comparison between male and female *Guduchi* in the preparation of *Guduchi* Ghana highlights the female *Guduchi*' s advantage in extraction efficiency. Both plants yield a similar brownish-green semi-solid mass and black solid concentrate, offering comparable medicinal properties. However, the female *Guduchi* thickens faster, reaching a concentrated state in 3 hours compared to 3.5 hours for the male plant.

This faster processing makes the female *Guduchi* more efficient and time-saving, without compromising the final product's quality. For commercial and traditional applications, where efficiency is crucial, the female *Guduchi* is the preferred choice. Its ability to quickly release bioactive compounds makes it an optimal option for producing high-quality *Guduchi Ghana*.

5.7. 1 Preparation of *Sanshamni Vati*:

Vati was prepared as per the reference in "*Siddha Yoga Sangraha*" The *Guduchi Ghana Vati* was prepared with the help of manual Hand operated tablet Making Machine, having die of 250 mg. After cutting with the die the pieces rolled into round *Vati* with the help of hands. Then placed into the hot air oven at 45°C for further drying. Then checked the average weight of 10 tablets (2.5gm)

Fig:5.3 Preparation of Sanshamni Vati
a) *Guduchi* pieces b) Crushed *Guduchi* c) Soaking d) *Kwath Nirman* E) Temperature 95-degree f) Filtration G) *Ghan* shifted to tray H) Oven drying at 45 degree i) Prepared *Ghan* J) *Vati Nirman*.

Table 5.19. *Sanshamni Vati* preparation from *Guduchi* Male and Female *Guduchi Ghana.*

S.no	*Guduchi**Gahana* Male(gm)	No. of *Vati* prepared	*Guduchi Ghana* Female (gm)	No. of *Vati* prepared
1	290	1084	320	1200
2	295	1096	298	1112
3	286	1116	290	1088
4	290	1100	295	1108
5	285	1070	290	1084
6	280	1056	295	1100
Std.Dev.	287.67±4.71	1087±19.79	298±10.25	115.33±39.15

Shanshamni Vati made from female *Guduchi* has an average mass of 298±10.24 grams, producing about 1115.33±39.15 Vati, which is higher than the male *Guduchi,* with an average mass of 287.66±4.71 grams and an output of 1087±19.79 Vati. While the female *Guduchi* shows slightly more variation in both mass and Vati production, its consistently greater yield makes it the favoured choice.

5.7.2. Evaluation of *Sanshamni Vati*:

Table No.5.20: Evaluation of *Sanshamni Vati* (M)

Sr. No.	Parameter	1 M	2 M	3 M	4 M	5 M	6M
1.	LOD(%w/w)	8.4	8.2	8.5	8.3	8.2	8.2
2.	pH	6.00	6.48	6.53	6.54	6.57	6.52
3.	Hardness (kg/cm3)	3.98	4.50	4.00	4.35	4.43	4.67
4.	Disintegration Time (min)	17	18	20	17	17	18
5.	Friability%	o.6	0.5	0.6	0.7	0.7	0.6

Table No. 5.21: Evaluation Parameters of *Sanshamni Vati* (FM)

Sr. No.	Parameter	1 FM	2 FM	3 FM	4 FM	5 FM	6FM
1.	LOD(%w/w)	8.5	8.6	8.3	8.3	8.6	8.3
2.	pH	6.43	6.44	6.42	6.41	6.44	6.41
3.	Hardness (kg/cm3)	3.78	4.47	4.00	4.35	4.66	4.45
4.	Disintegration Time (min)	15	17	16	18	16	17
5.	Friability%	0.6	0.5	0.6	0.7	0.5	0.6

Table No. 5.22: Evaluation 0f Organoleptic parameter of *Sanshamni Vati* (M&F)

Sr. No.	Parameter	*Sanshamni Vati* (M&F)
1	Color	Blackish- brown
2	Odor	Characteristic
3	Taste	Bitter

No significant variation was observed in organoleptic and physico-chemical parameters evaluated so far. Average disintegration time was 16.5 minutes. All the parameters are recorded in table 5.20 & 5.21.

5.7.3. Physicochemical analysis of *Sanshamni Vat*i

Table 5.23. Physicochemical analysis of male *Sanshamni Vati*

Parameters	Batch(M)						Std. Dev.
	I	II	III	IV	V	VI	
Total ash (%w/w)	11.65	11.89	12.00	11.78	11.90	11.9511	11.86±0.11
Alcohol Soluble Extractive(%w/w)	7.54	7.18	7.37	7.43	7.35	7.64	7.41±0.13
Water Soluble Extractive(%w/w)	27.40	26.97	27.39	26.99	27.39	27.55	27.28±0.22
Sulphated Ash (%)	28.00	270	27.3	28.05	27.90	28.14	27.95±0.13
Foreign Matter	-	-	-	-	-	-	-
Total Alkaloids	0.80	0.85	0.63	0.79	0.70	0.74	0.75±0.07

Table 5.24 Physicochemical analysis of Female *Sanshamni Vati*

Parameters	Batch (FM)						Std. Dev.
	I	II	III	IV	V	VI	
Total Ash(%w/w)	11.10	11.60	11.73	12.01	12.05	12.23	11.79±0.37
Alcohol Soluble Extractive(%w/w)	2.10	2.35	2.50	2.14	2.43	2.69	2.36±0.20
Water Soluble Extractive(%w/w)	31.00	32.00	31.45	32.15	31.73	32.99	32.99±0.13
Sulphated Ash (%)	16.25	16.90	16.73	16.84	16.73	16.96	16.73±0.23
Foreign Matter	-	-	-	-	-	-	-
Total Alkaloids	1.50	1.54	1.43	1.35	1.26	1.59	1.44±0.11

Both the male and female formulations of *Sanshamni Vati* generally meet the quality standards set by the Ayurvedic Pharmacopoeia of India.

Higher water-soluble compounds (e.g. glycosides, polysaccharides, tannins etc.) reflects stronger presence of phytochemicals extractable in water due to which female Guduchi may offer a balanced spectrum of hydrophilic phytoconstituents harmonizing better with other ingredients in the formulation. This may be correlated with therapeutic effectiveness of female guduchi in water-based preparations like Sanshamni vati and use of male guduchi in alcohol-based formulations such as Amrita Rishta, enhancing overall pharmacological action. But the data may not be sufficient to directly correlate with therapeutic potency in aqueous and alcohol-based formulations. Furthermore, preclinical and clinical studies can be conducted to gather more evidence.

5.8 HPTLC-Study:

The HPTLC fingerprint confirms the plant's identity. Quantitative analytical estimation requires hyperspectral data, densitograms, or image profiles. This analytical technique involves sophisticated instrumentation, standardized and documented processes, and validated methods. One of the strengths of HPTLC is the ability to use the same sample and plates for multiple detections. A marker compound, a chemically defined constituent, is used for botanical identification, detection of adulteration, and as an indicator of product quality, but not for therapeutic purposes. In the HPTLC study, the Rf value of the extract was compared with the standard Rf value. Berberine, known to be present in the stem of Guduchi, was used as the marker compound. The identical Rf value of both samples and standard confirms the quality of sample Male and Female.

The data reveal that both *Guduchi* Male and Female share similar phytochemical profiles, but female *Guduchi*'s closer alignment with standard Berberine suggests it might contain higher levels of this compound, which is known for its therapeutic benefits. The slight variations in Rf values between *Sanshamni Vati* (M) and *Sanshamni Vati* (FM) formulations indicate that although the overall chemical composition is similar, the female-based formulation may offer marginally enhanced therapeutic potential due to its different retention factors, particularly with regard to key compounds like Berberine.

This analysis supports the potential preference for female *Guduchi* in medicinal applications where these compounds are critical, as well as the possible superior efficacy of the female-based *Sanshamni Vati* formulation.

The berberine content is relatively low in both formulations, with the female formulation (0.17%) having a higher content than the male formulation (0.06%). Berberine is known for its antimicrobial and anti-inflammatory properties.

5.8.5. The beta-sitosterol content is also higher in the female formulation (4.26%) compared to the male formulation (2.02%). Beta-sitosterol is a plant sterol known for its potential cholesterol-lowering effects and other health benefits. The higher content in the female formulation might contribute to different therapeutic properties.

S. No.	Sample	Rf value
1.	*Guduchi* Male	0.58
2.	*Guduchi* Female	0.60
3.	Standard Berberine @366nm	0.60
4.	*Sanshamni Vati* (M)	0.66
5.	*Sanshamni Vati* (FM)	0.68
6.	Standard Berberine @366nm	0.69
7.	*Sanshamni Vati* (M)	0.89
8.	*Sanshamni Vati* (FM)	0.90
9.	Standard β- Sitosterol @366nm	0.92

Table 5.25. Rf value of *Guduchi* extract, *Sanshamni Vati* prepared with both genders, standard Berberine and Standard β- Sitosterol.

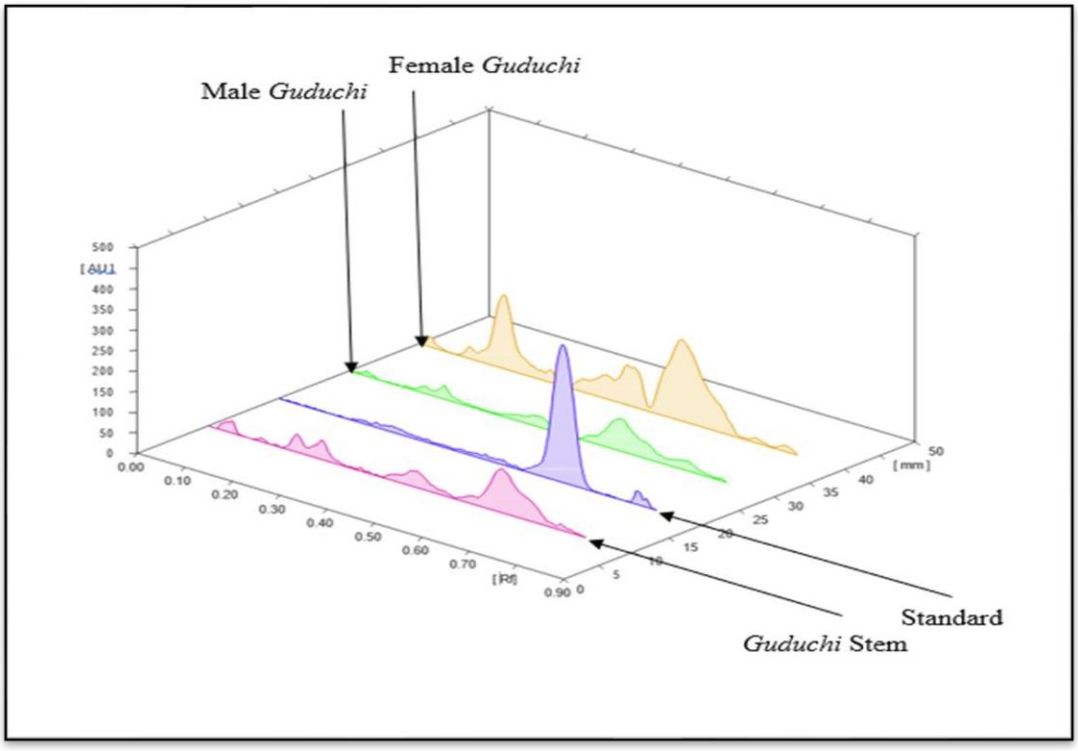

Fig:5.4 Chromatogram for HPTLC analysis of Guduchi male and Guduchi Female stem for Berberine.

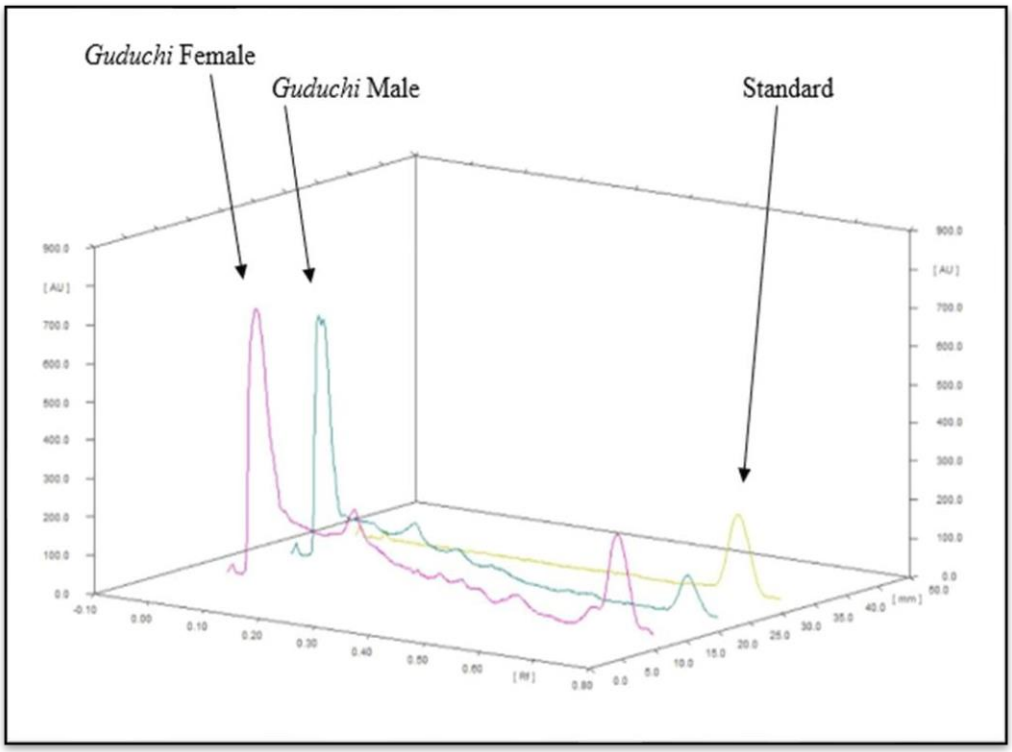

Fig: 5.5 Chromatogram for HPTLC analysis of *Guduchi* male and *Guduchi* Female prepared *Sanshamni Vati* for Berberine.

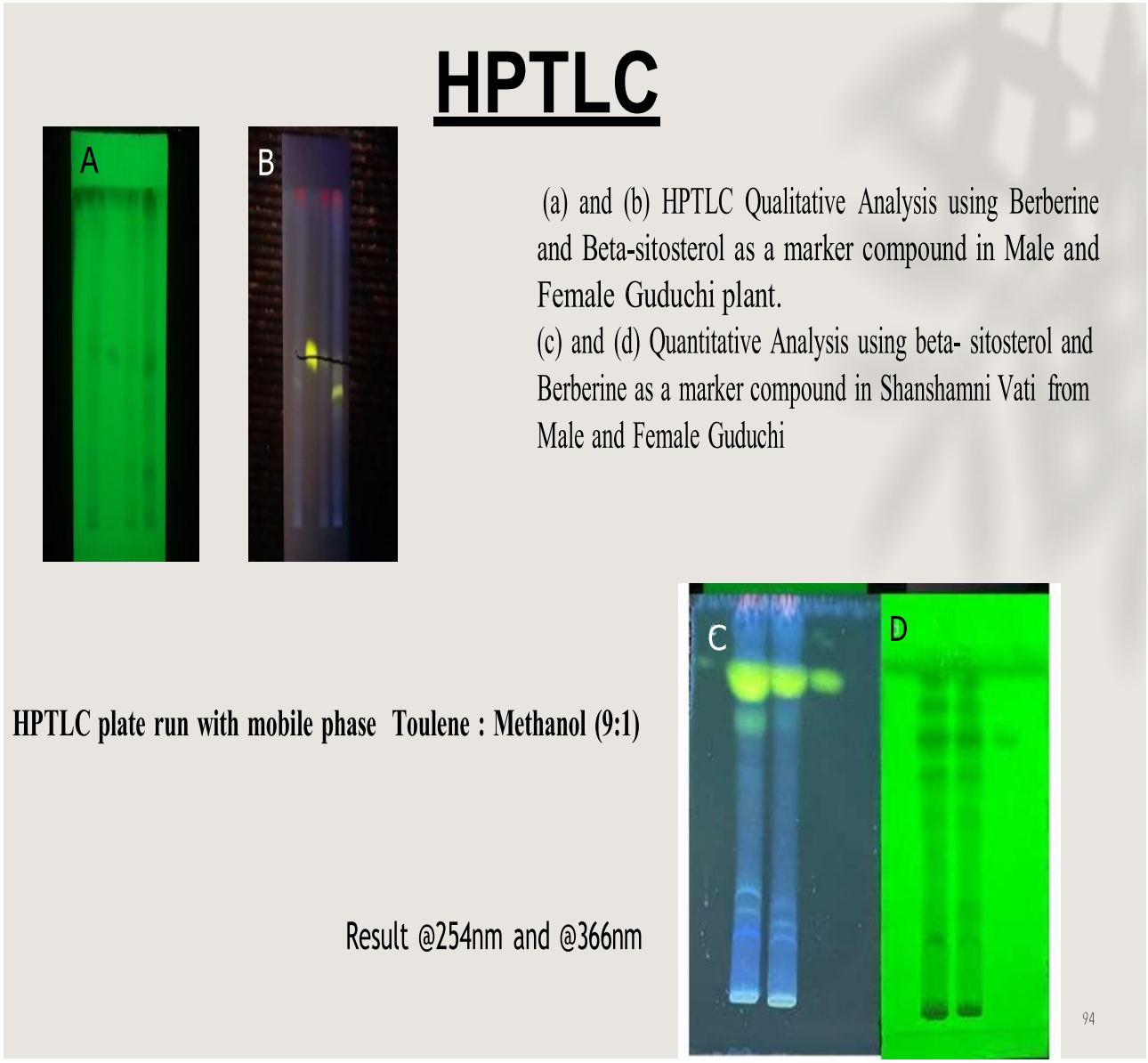

Fig 5.6 Pictorial presentation of thin layer chromatography of Guduchi & sanshamni vati

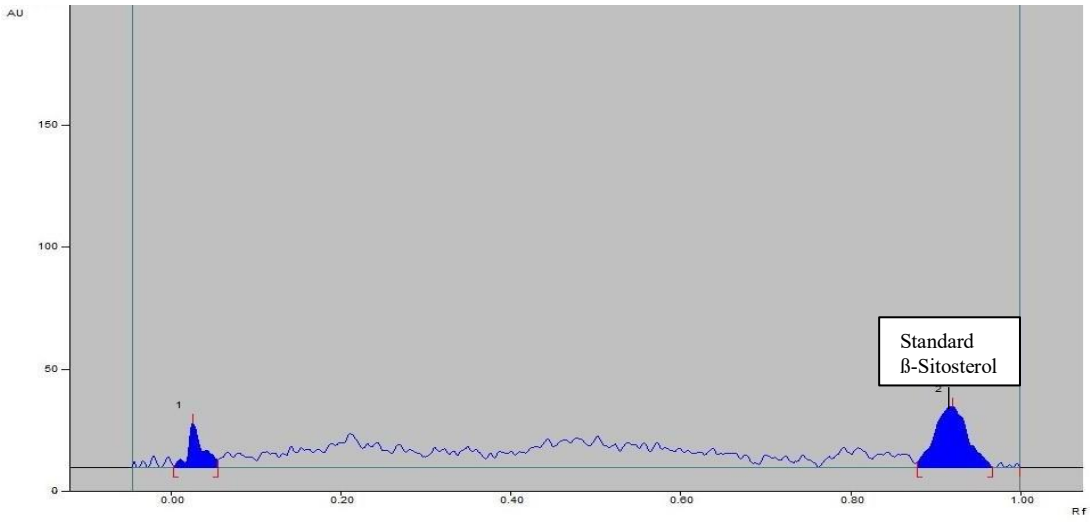

Figure 5.7: ß-Sitosterol standard

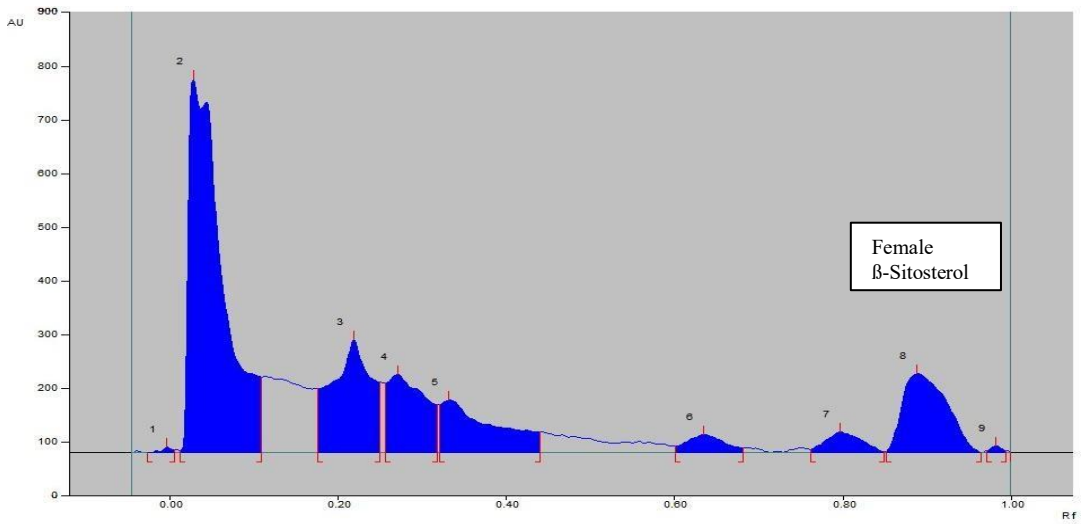

Figure5.8: Female ß-Sitosterol

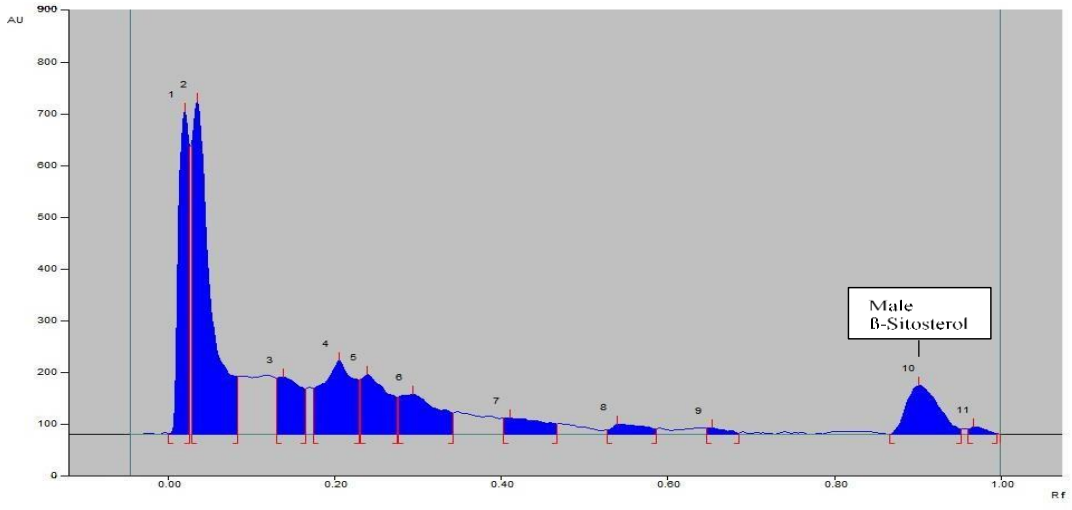

Figure 5.9: Male ß-Sitosterol

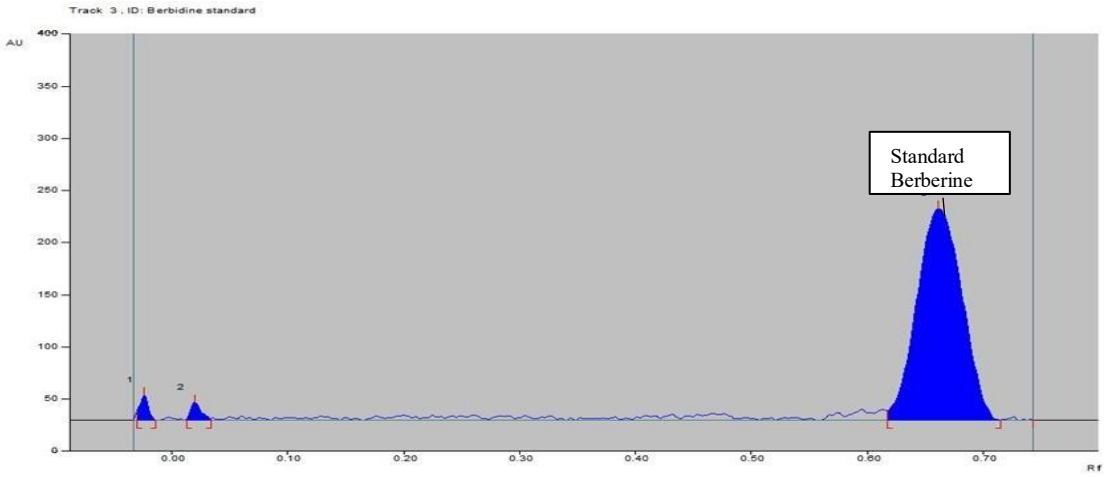

Figure 5.10: Standard Berberine

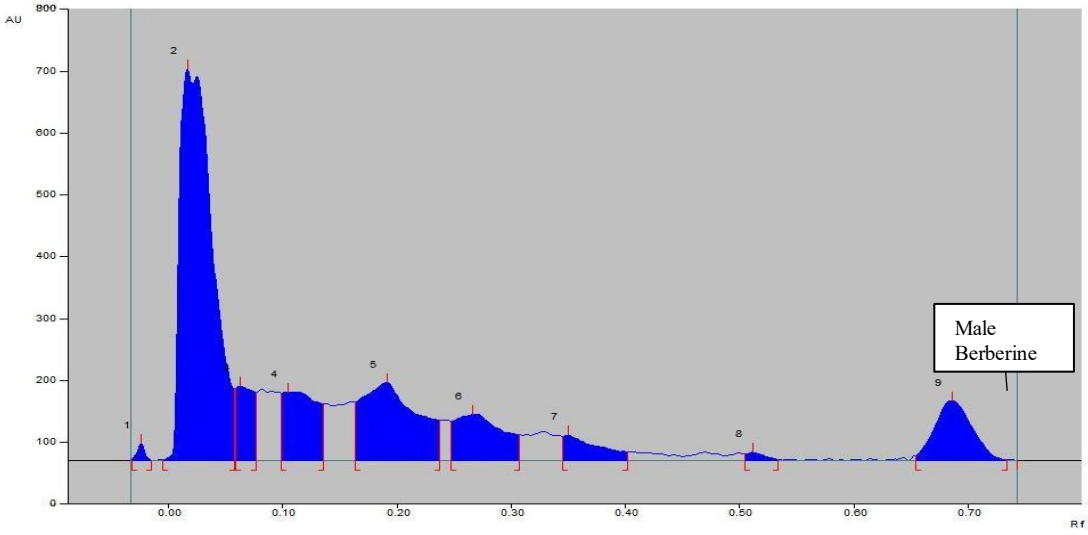

Figure 5.11: Male Berberine

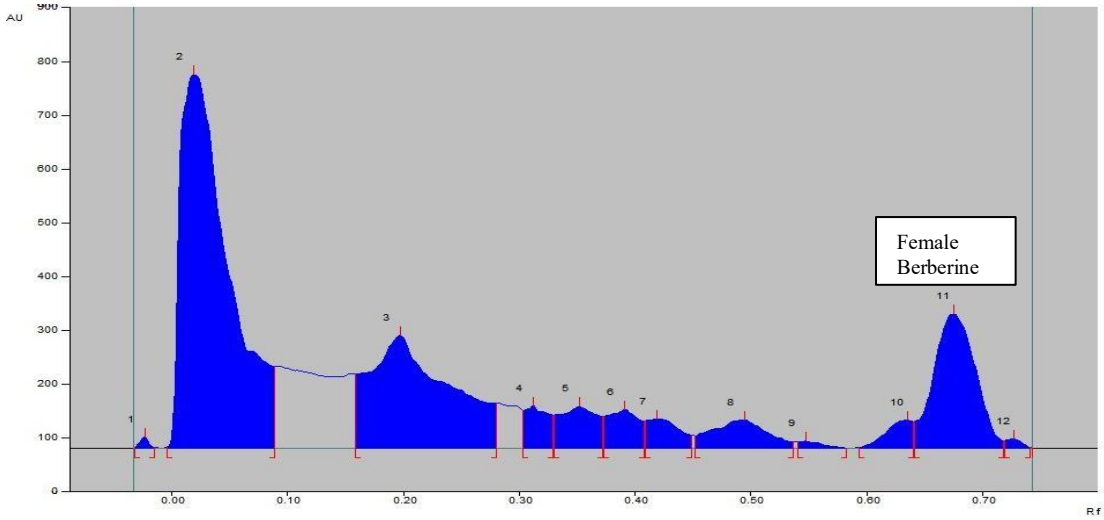

Figure5.12: Female Berberine

5.10 Animal Study:

Table No. 5.26: Paw measurement at different time Intervals

S.No.	Groups	Time				
		1	2	3	4	% Inhibition
I	Normal Control	0.00±0.000	0.00±0.000	0.00±0.000	0.00±0.000	-
II	Carrageenan control (0.1 ml of 2% (w/v)	1.80±0.299	2.02±0.273	2.10±0.297	2.33±0.182	-
III	Indomethacin (10mg/kg bw)	1.43±0.152 NS	1.20±0.150*	0.94±0.319*	0.46±0.220**	80.25
IV	Sample A (500 mg/kg bw)	1.52±0.164 NS	1.76±0.188 NS	1.40±0.136 NS	0.71±0.200**	69.52
V	Sample B (500 mg/kg bw)	1.46±0.130 NS	1.53±0.142 NS	1.00±0.062*	0.62±0.125**	73.39

Table no 5.27: Paw Measurements at 1 hour

PAW MEASUREMENTS AT 1 HR

Source of variation	DF	SS	MS	F	P
Between the groups	4	12.084	3.021	16.107	<0.001
Residual	25	4.689	0.188		
TOTAL	29	16.774			

The Differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

Table no5.28: Paw Measurements at 2 hours

PAW MEASUREMENTS AT 2 HR

Source of variation	DF	SS	MS	F	P
Between the groups	4	14.897	3.724	20.347	<0.001
Residual	25	4.5760	0.183		
TOTAL	29	19.473			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

Table no 5.29: Paw Measurements at 3 hours

PAW MEASUREMENTS AT 3 HR

Source of variation	DF	SS	MS	F	P
Between the groups	4	14.009	3.502	13.747	<0.001
Residual	25	6.360	0.255		
TOTAL	29	20.379			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

Table no 5. 30: Paw Measurements at 4 hours

PAW MEASUREMENTS AT 4 HR

Source of variation	DF	SS	MS	F	P
Between the groups	4	18.805	4.701	28.565	<0.001
Residual	25	4.1140	0.165		
TOTAL	29	22.919			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

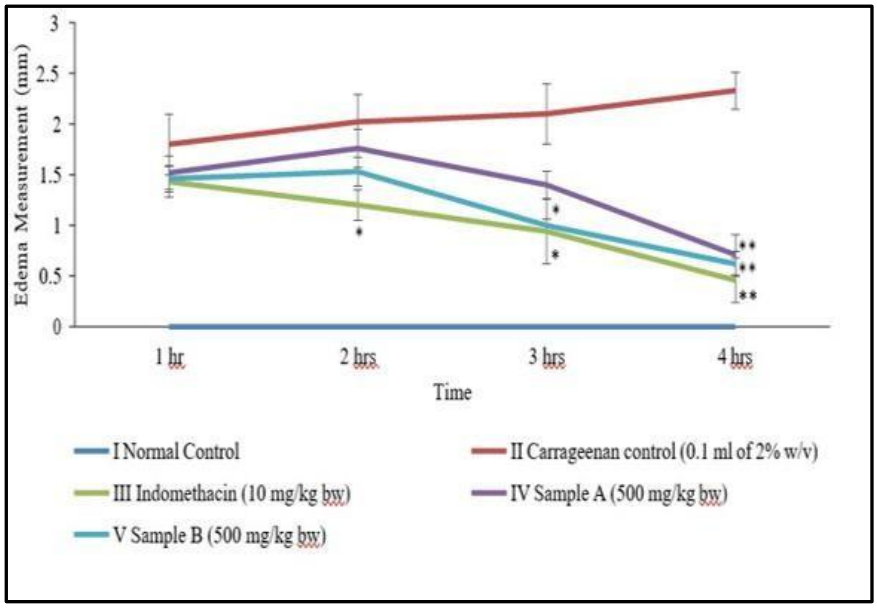

Fig 5.13: Graphical representation of Paw measurement at different time

The graph demonstrates that both Sample A and Sample B have considerable anti-inflammatory properties, with **Sample B being slightly more effective than**

Sample A. However, both are slightly less effective than the standard drug Indomethacin. This suggests that the formulations derived from the plants have potential therapeutic benefits in reducing inflammation.

Summary:

- Indomethacin: Highly effective in reducing edema, serving as the positive control.
- Sample A and Sample B: Shows promising anti-inflammatory effects, reducing edema compared to carrageenan control
- Carrageenan Control: Effectively induced inflammation, as expected.
- Normal Control: Baseline with no induced inflammation, serving as a reference for natural paw measurements.

5.11 Hematological Analysis

Table No. 5.31: Blood Analysis after Treatment with Sample A(male)& (Female)

S. No.	Groups	HB	RBC	WBC	ESR
I	**Normal Control**	15.43±1.023	11.30±0.674	5.28±0.382	5.08±0.624
II	**Carrageenan Control (0.1 ml of 2% (w/v))**	7.72±0.601	5.87±0.635	11.67±0.516	11.50±0.548
III	**Indomethacin (10mg/kg bw)**	14.67±0.410**	10.72±1.340*	5.27±0.375**	4.85±0.505**
IV	**Sample A (500 mg/kg bw)**	11.48±0.679^{NS}	8.82±0.897^{NS}	6.31±0.540**	5.35±1.138**
V	**Sample B (500 mg/kg bw)**	12.20±1.675*	9.34±0.448^{NS}	6.66±0.413**	6.58±0.492**

Values are expressed as MEAN±SD at n=6, One-way ANOVA followed by Bonferroni test, *P<0.050, **P<0.001 and **NS**P>0.001 compared to the Carrageenan control

Table no 5.31: Hematological Analysis HB

HAEMATOLOGICAL ANALYSIS Hb

Source of variation	DF	SS	MS	F	P
Between the groups	4	222.436	55.609	9.570	<0.001
Residual	25	145.275	5.811		
TOTAL	29	367.710			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

Table no 5.33: Haematological Analysis RBC

HAEMATOLOGICAL ANALYSIS RBC

Source of variation	DF	SS	MS	F	P
Between the groups	4	107.837	26.959	6.141	<0.001
Residual	25	109.752	4.390		
TOTAL	29	217.589			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = 0.001).

Table no 5.34: Haematological Analysis WBC

HAEMATOLOGICAL ANALYSIS WBC

Source of variation	DF	SS	MS	F	P
Between the groups	**4**	170.068	42.517	34.908	**<0.001**
Residual	**25**	30.449	1.218		
TOTAL	**29**	200.51			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

Table no 5.35: Haematological Analysis ESR

HAEMATOLOGICAL ANALYSIS ESR

Source of variation	DF	SS	MS	F	P
Between the groups	4	185.519	46.380	15.573	**<0.001**
Residual	25	74.454	2.978		
TOTAL	29	259.974			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

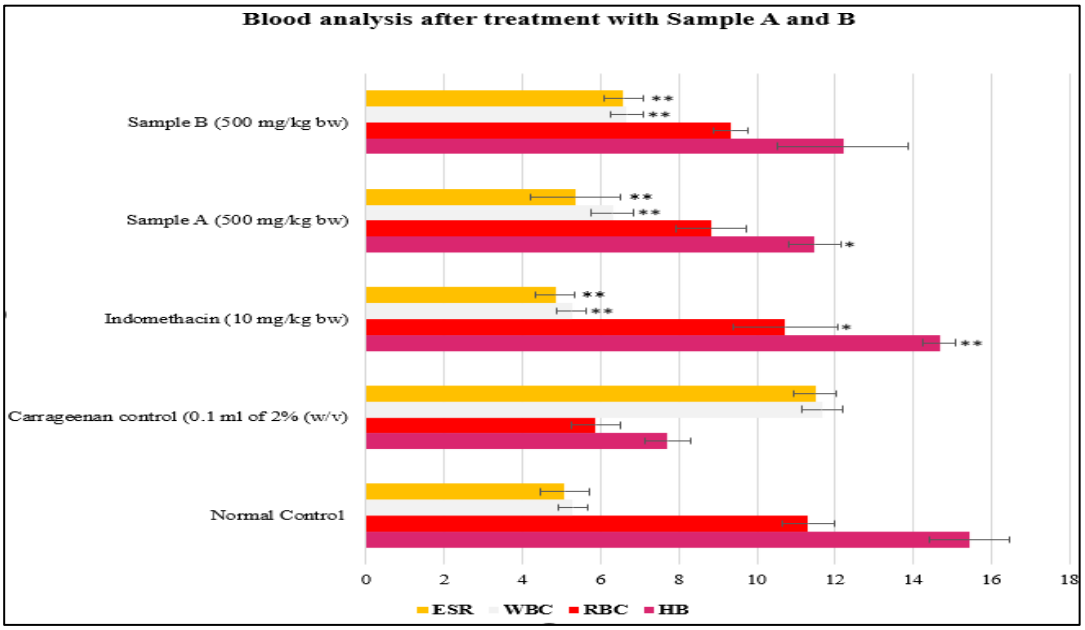

Fig 5.14: Graphical representation of Blood Analysis After Treatment with Sample A (male) & B(female)

For all metrics, the "Normal Control" condition is the baseline of health. Indicating inflammation and its effect on blood health, the carrageenan control group exhibits increased ESR and WBC and decreased RBC and HB. Indomethacin: Good for bringing RBC and HB back to normal, while effectively lowering WBC and ESR. Sample A: Restores blood health parameters partially and exhibits mild anti-inflammatory effects. Sample B: Shows a similar trend in restoring RBC and HB and notable anti- inflammatory benefits to those of indomethacin. According to this graph, Sample A and Sample B may both have anti-inflammatory properties; however, Sample B appears to have more potent effects that are on par with those of the prescription medication indomethacin. These results are essential for assessing the samples' medicinal potential for lowering inflammation and preserving blood health.

5.12 Cytokine Estimation

Table 5.36 TNF-α level in Sample A and Sample B treatment groups

Sr. No.	GROUPS	TNF-α (pg/ml)
I	**Normal Control**	60.68±1.225
II	**Carrageenan control (0.1 ml of 2% (w/v)**	120.37 ± 0.512
III	**Indomethacin (10 mg/kg bw)**	70.11 ± 1.198**
IV	**Sample A (500 mg/kg bw)**	93.75±2.047**
V	**Sample B (500 mg/kg bw)**	81.28±1.867**

Values are expressed as MEAN±SD at n=6, One-way ANOVA followed by Bonferroni test, *P<0.050, **P<0.001 and ^{NS}P>0.001 compared to the Carrageenan control

Table no 5.37: Cytokine Analysis TNF

CYTOKINE ANALYSIS- TNF-α (Pg/ml)

Source of variation	DF	SS	MS	F	P
Between the groups	4	12925.974	3231.494	247.650	**<0.001**
Residual	25	326.216	13.049		
TOTAL	29	13252.191			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

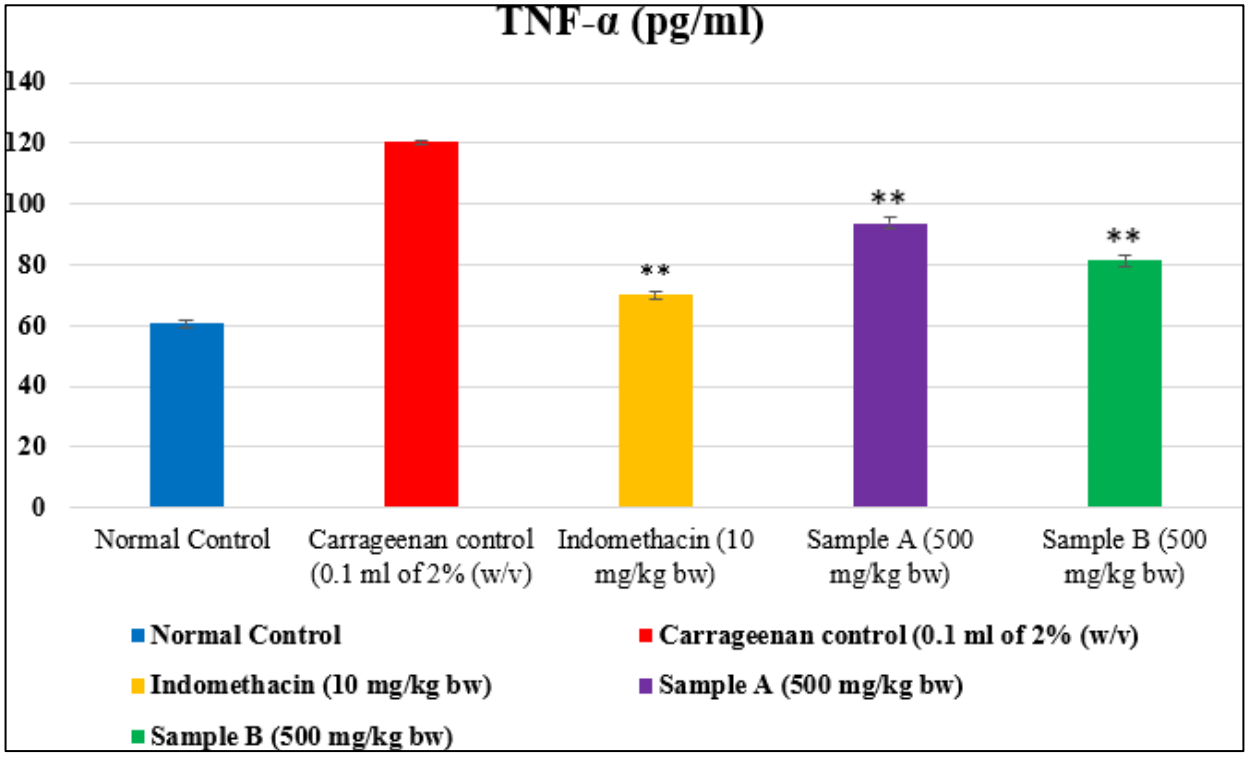

Fig 5.15: Graphical representation of TNF-α level in Sample A and Sample B treatment groups

The graph above displays the TNF-α levels in different sample groups, along with their respective standard deviations. Here's a breakdown of the data presented:

The Carrageenan control group shows the highest TNF-α level, indicating a significant inflammatory response. Indomethacin and both samples (A and B) show a reduction TNF-α levels compared to the Carrageenan control, with Indomethacin being the most effective in reducing TNF-α levels, followed by Sample B and Sample A

Table no.5.38: IL-6 Levels in Sample A & B

Sr.no.	**Groups**	**IL-6 (Pg/ml)**
I	Normal Control	89.02±1.069
II	Carrageenan control (0.1 ml of 2% (w/v)	213.85±1.828
III	Indomethacin (10 mg/kg bw)	112.77±0.977**
IV	Sample A (500 mg/kg bw)	129.68±1.180**
V	Sample B (500 mg/kg bw)	117.11±2.353**

Values are expressed as MEAN±SD at n=6, One-way ANOVA followed by Bonferroni test, *P<0.050, **P<0.001 and ^{NS}P>0.001 compared to the Carrageenan control.

Table no 5. 36: Cytokine Analysis-IL-6(Pg/ml)

CYTOKINE ANALYSIS- IL-6 (Pg/ml)

Source of variation	DF	SS	MS	F	P
Between the groups	4	54854.456	13713.614	924.007	**<0.001**
Residual	25	371.036	14.841		
TOTAL	29	5525.492			

The differences in the mean values among the treatment groups are greater than would be expected by chance; there is a statistically significant difference (P = <0.001).

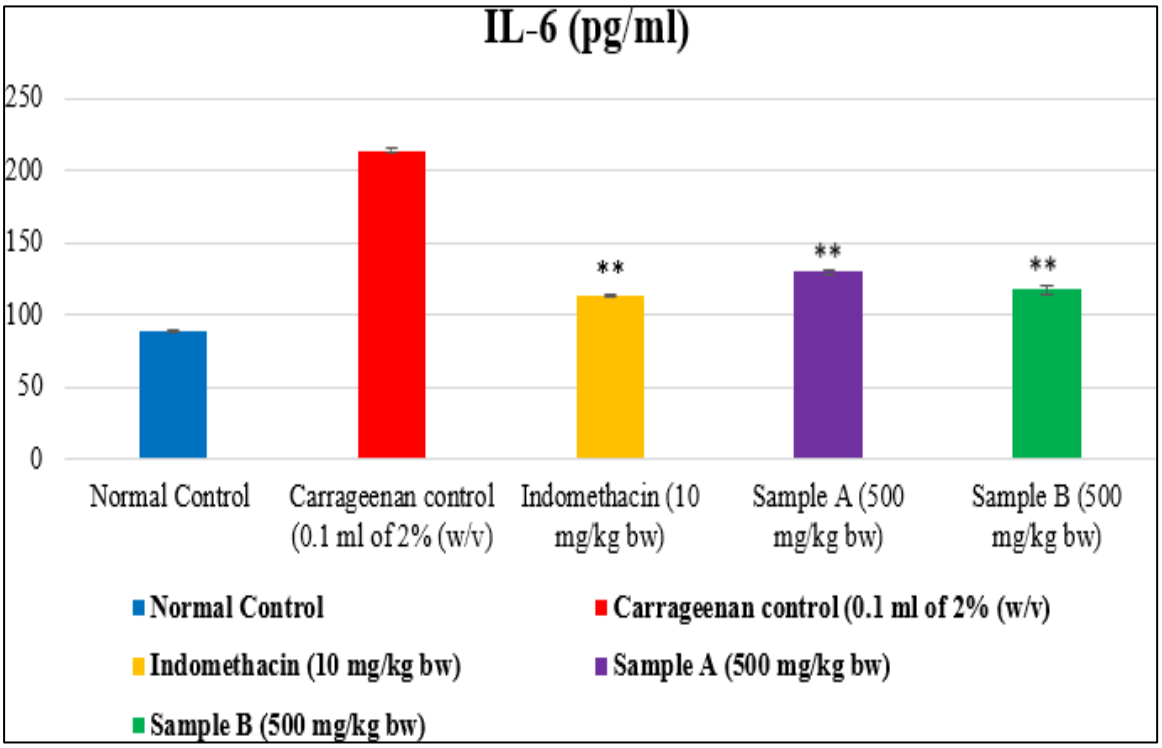

Fig 5.16: Graphical representation of IL-6 Levels in Sample A and Sample B

The graph above displays the IL-6 levels in different sample groups, along with their respective standard deviations. Here's a breakdown of the data presented. The Carrageenan control group shows the highest IL-6 level, indicating a significant inflammatory response. Indomethacin and both samples (A and B) show a reduction in IL-6 levels compared to the Carrageenan control, with Indomethacin being the most effective in reducing IL-6 levels, followed by Sample B and Sample A. To investigate the effects of samples A&B on the regulation of immune function in the immunosuppressed mice, the levels of TNF-α and IL-6 was determined in serum. This data showed that TNF-α and IL-6 serum levels were significantly increased in rats injected with a Carrageenan. The samples A and B treatment reduced serum levels of IL- 6, and TNF-α compared with the Carrageenan group.

5.12 Histopathological findings

Group I Normal control showing normal histological architecture

Group II carrageenan induced with detachment of epidermal layer severe inflammatory reaction in dermal layer, oedema, necrosis.

Group III Indomethacin treated rats almost retaining its normal paw size with normal tissue architecture.

Group IV Sample A treated mild hyperplasia, Meta tarsus, sweat glands and almost retaining its normal paw size degeneration of muscle fibers, moderate dermal inflammation.

Group V Sample B showing mild oedema

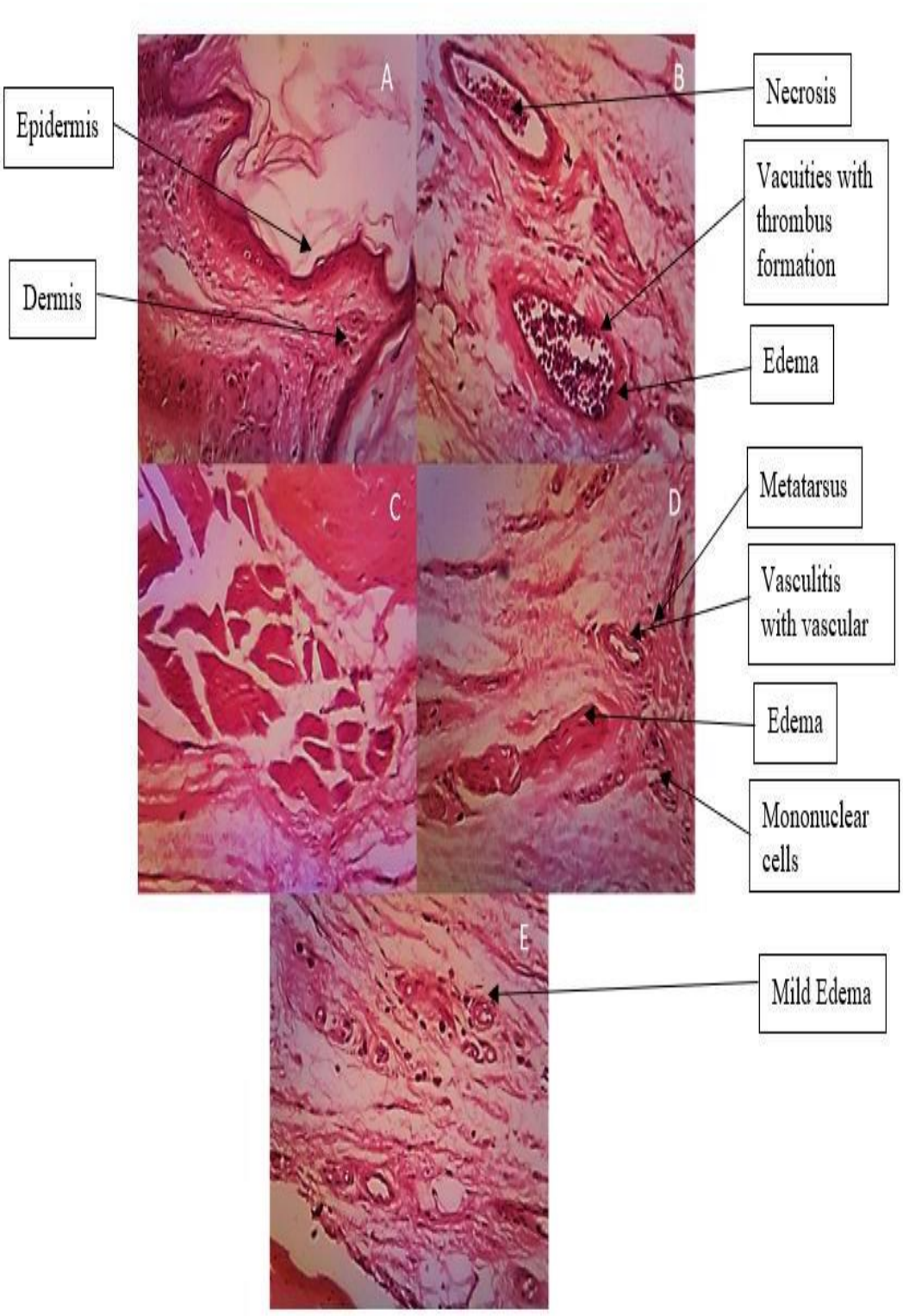

Fig: 5.17: Histopathological findings

(a) Normal control (b) Carrageenan induced (c) Indomethacin treated (d) Sample A treated (E) Sample B treated

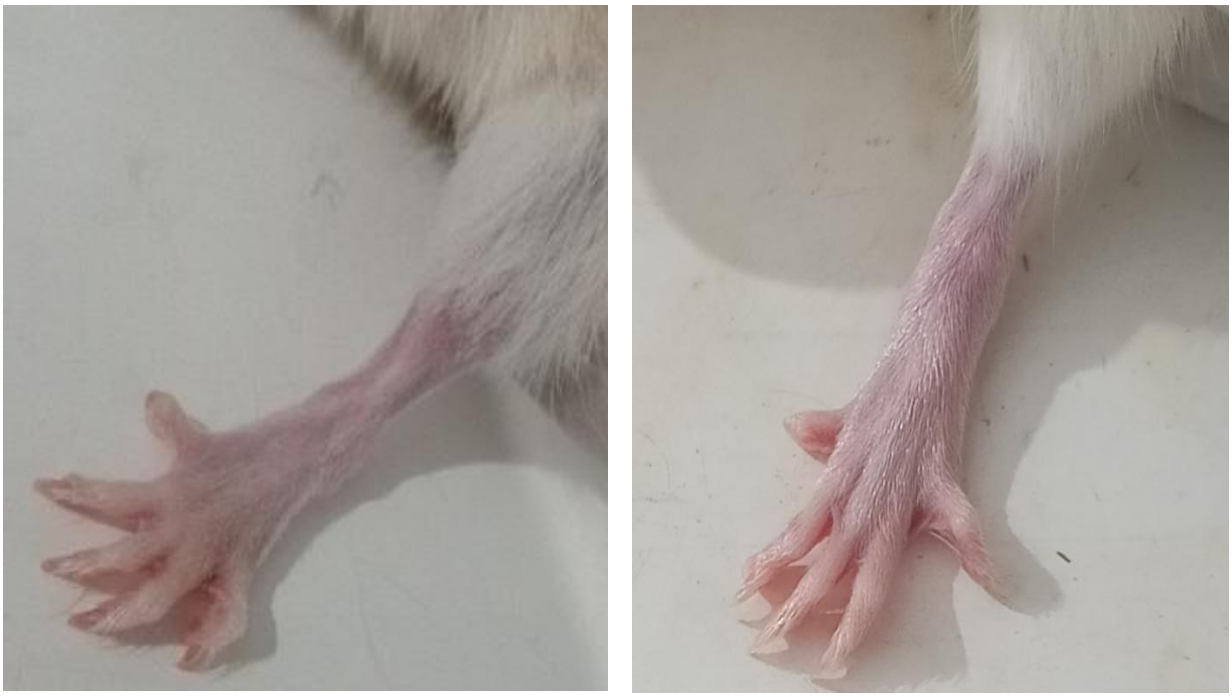

Fig: 5.18: Group I: Normal control at 1 hr and 4 hr

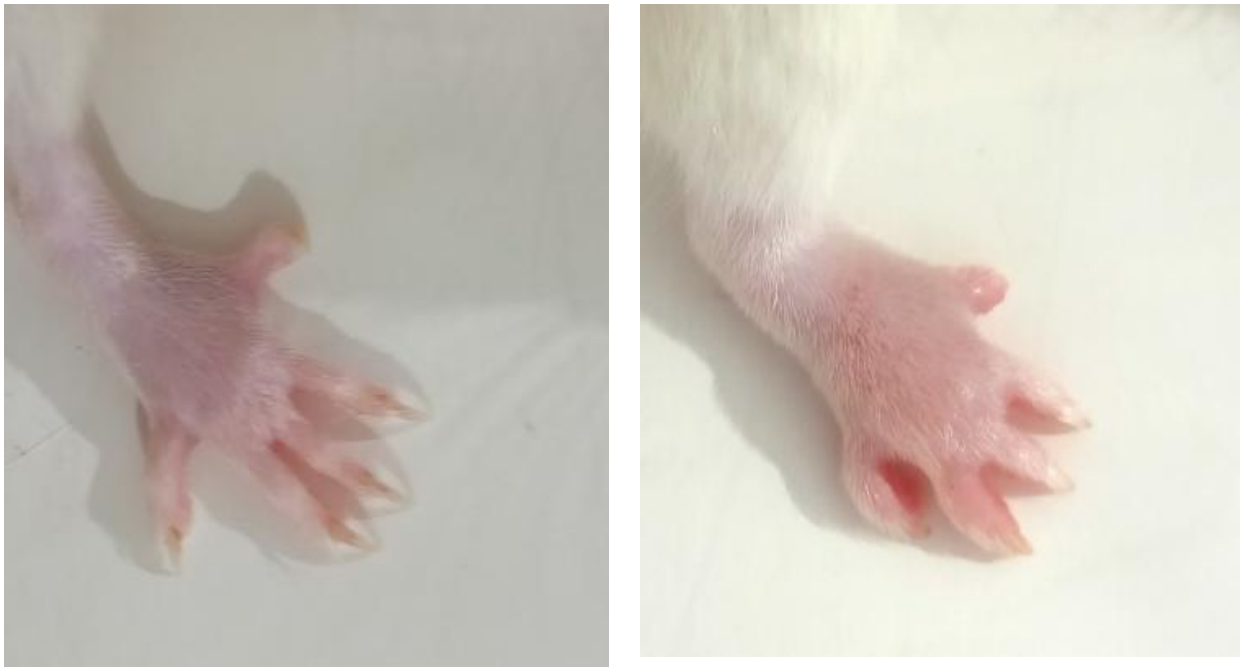

Fig: 519: Group II: Carrageenan control (0.1 ml of 2% w/v) at 1hr and 4 hr

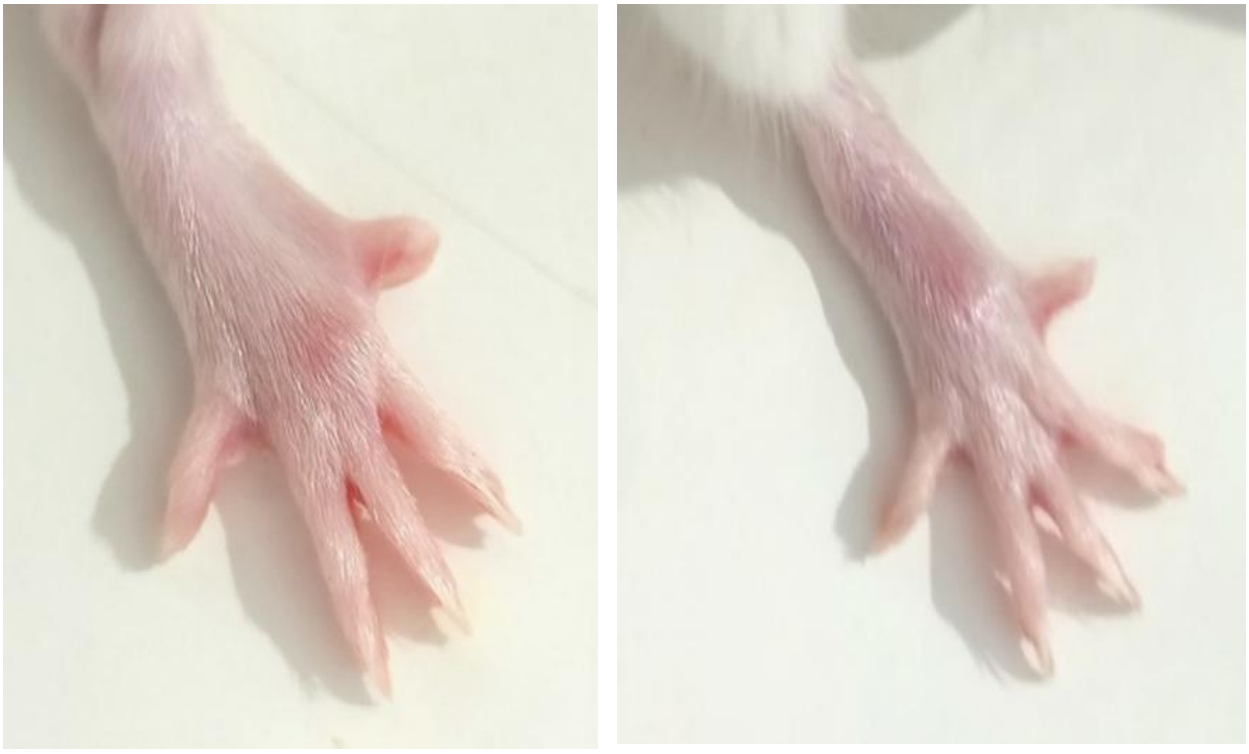

Fig 5.20: Group III: Indomethacin (10mg/kg) at 1 hr and 4 hr

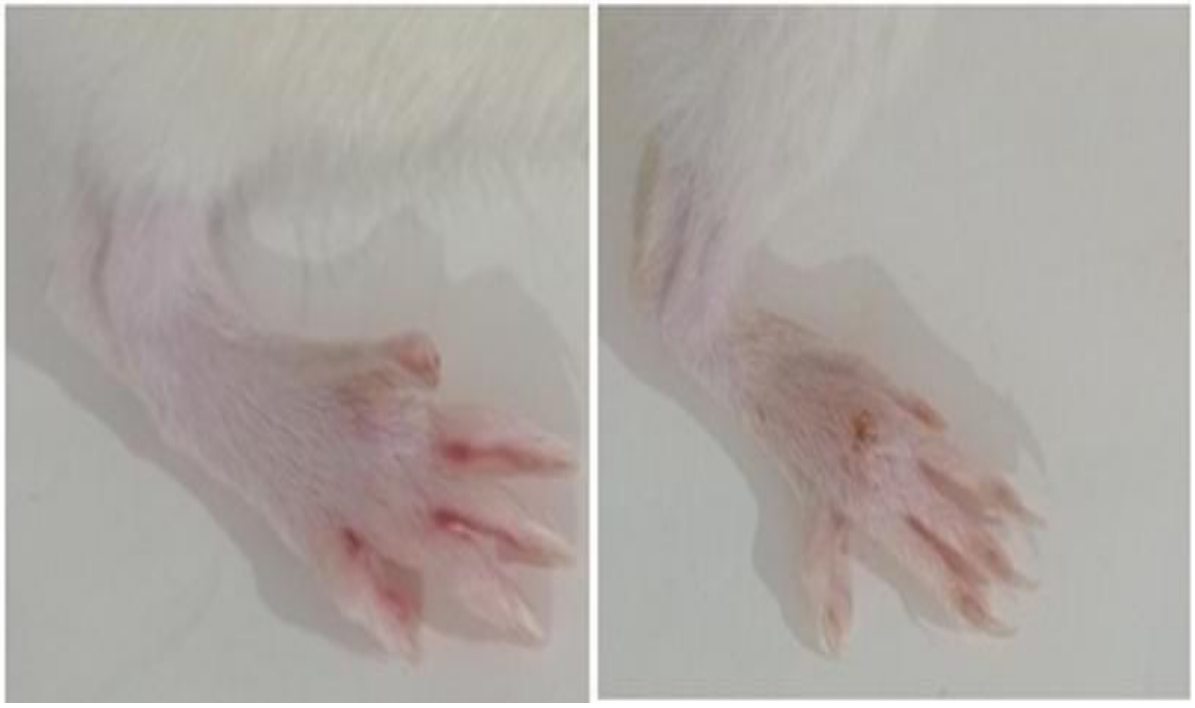

Fig 5.21 Group IV: Sample A 500 mg/kg at 1hr and 4 hr

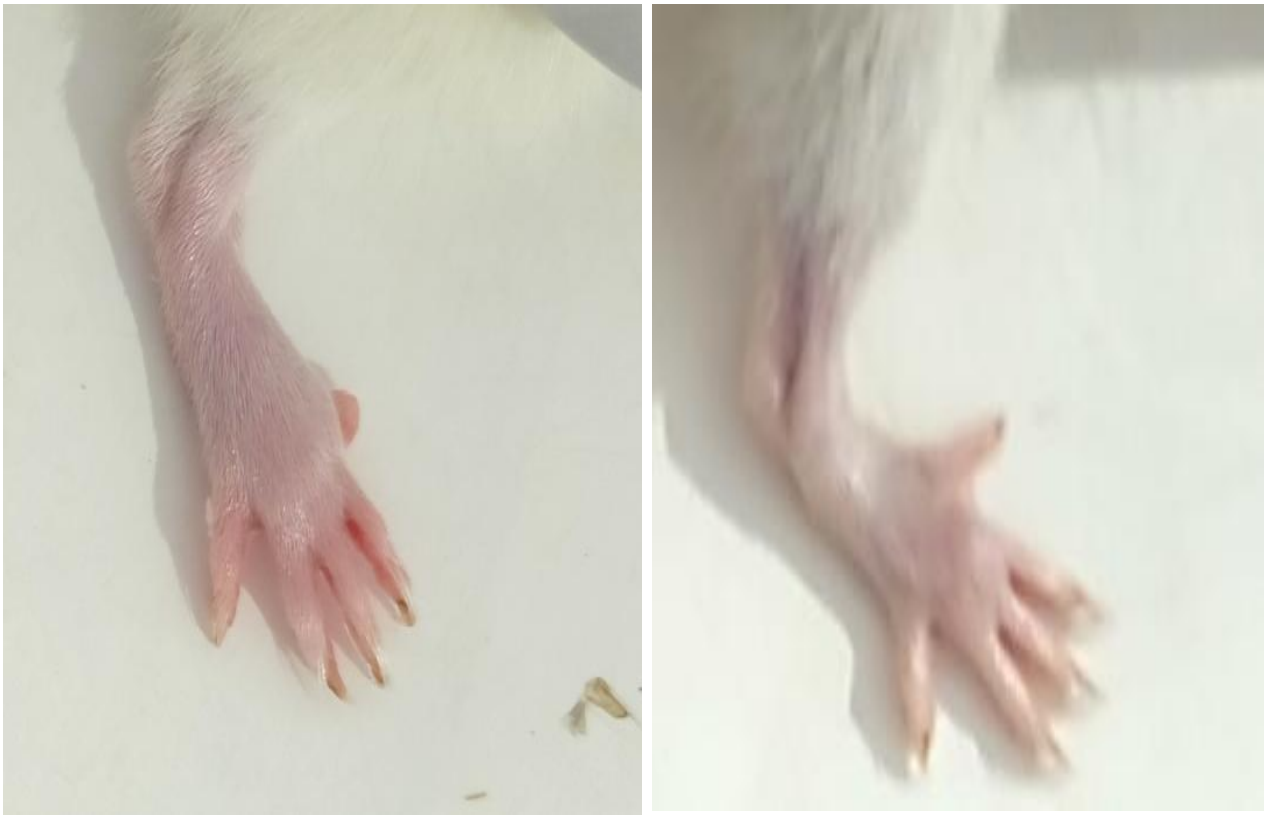

Fig 5.22: Group V: Sample B 500 mg/kg at 1hr and 4 hr.

Both Sample A and Sample B demonstrated anti-inflammatory activities in the carrageenan-induced paw edema model, with Sample B showing a relatively stronger effect.

Chapter 6

Conclusion and Future Perspective

"Based on the findings of this study, it can be concluded that *Guduchi* (*Tinospora cordifolia*) exhibits dioecious traits, with male and female reproductive structures being present on separate individuals. The study comparing male and female *Guduchi* plants revealed both shared morphological characteristics and distinct differences between the genders. While both genders exhibit similar stem appearance and growth patterns, subtle variations were observed in colour, Odor, and taste. Female *Guduchi* plants displayed a darker green colour with a creamish hue, contrasting with the lighter green appearance of male *Guduchi*. Female *Guduchi* emitted a stronger smell compared to males. Female *Guduchi* was reported to be more bitter than male *Guduchi*. The analysis of various parameters for male and female *Guduchi* samples, along with the standard requirements, Loss on Drying, Total Ash, Acid Insoluble Ash, Alcohol Soluble Extractive, Water Soluble Extractive, Foreign Matter, Sulphated Ash, Berberine Assay by HPTLC: Both samples show the presence of berberine, with the female *Guduchi* sample exhibiting a higher concentration compared to the male sample. Overall, the results suggest that both male and female *Guduchi* samples largely meet the standard requirements, with minor differences observed between the two genders in certain parameters.

Impact of Dioecism on Quality and Efficacy of *Sanshamni Vati*. Dioecism, the characteristic of certain plant species where individual plants are either male or female, can indeed have implications for the quality and efficacy of herbal remedies like *Sanshamni Vati* in several ways. The variability in chemical composition between male and female plants, plays an important role in its therapeutic effects. This study investigating the anti-inflammatory effect of *Sanshamni Vati* on the basis of dioecism using the carrageenan-induced paw edema test in rats have shown little better promising results.

Compared to the Carrageenan control group, Sample A exhibits modest

improvement in Hb and WBC counts, but the gains are not statistically significant. It does, however, show a statistically significant decrease in WBC counts, suggesting that it has some anti-inflammatory qualities. When compared to the Carrageenan control group, Sample B shows more encouraging outcomes, including statistically significant increases in Hb, WBC, and ESR levels. This implies that *Sanshamni Vati* Sample B has strong anti-inflammatory properties. Based on the carrageenan- induced paw edema test, WBC count, HB, and ESR levels. In comparison to the male sample (Sample A), the female sample (sample B) of *Sanshamni Vati* verifies more significant anti-inflammatory action by lowering paw edema and increasing HB, WBC, and ESR levels. Therefore, the outcomes of this study demonstrate that dioecy influences *Sanshamni Vati's* effect. However, more researches in future, to confirm these results and establish the ideal dosage, safety profile, and mechanism of action of *Sanshamni Vati* for clinical application based on dioecism, human clinical trials are required. The plant should be used by the manufacturer based on dioecy for best results. Standardization and quality control of herbal formulations are also crucial for ensuring reproducibility and reliability of study outcomes.

Chapter 7
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HERBHEAL
CARE BY NATURE

CIN No.- U24233PB2008PTC032243

ISO 9001

Herbal Health Research Consortium Pvt. Ltd.

(A GOVT. OF INDIA SPONSORED & PB. GOVT. ASSISTED AYUSH CLUSTER PROJECT)
Works at : Village Khyala Khurd, Ram Tirth Road, Amritsar
Ph.: 01858-262025, Fax.: 01858- 262026 E-mail: herbheal@ymail.com
Corrs. Add: 277, East Mohan Nagar, Amritsar - 143006

Date: 23/09/2021

PLANT AUTHENTIFICATION CERTIFICATE

This is to certify that the following plant sample is well authenticated and verified. These studies have been accomplished in consultation with literature in The Wealth of India (Volume X, Page No.251) and with herbarium specimen no. R.S.-028 in Herbal Health Research Consortium Pvt. Ltd. Amritsar.

Sr. No.	Common Name	Botanical Name
1.	Guduchi Male	*Tinospora Cordifolia*

We, here by confirm the authenticity of above mentioned plant.

This certificate issued to Dr.Madhurima, Phd scholar, Department of Ayurvedic Pharmaceutical Sciences, LPU, Phagwara (Punjab) Jalandhar .Registration no: 41900801

For Herbal Health Research Consortium Pvt. Ltd.

Authorized Signatory

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Sr. No.	Common Name	Botanical Name
1.	Guduchi Female	*Tinospora Cordifolia*

We, here by confirm the authenticity of above mentioned plant.

This certificate issued to Dr.Madhurima, Phd scholar, Department of Ayurvedic Pharmaceutical Sciences, LPU, Phagwara (Punjab) Jalandhar .Registration no: 41900801

For Herbal Health Research Consortium Pvt. Ltd.

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A Government Approved Testing Laboratory
Lic no. 1/ASU/TESTING LAB./PB./2014 **Date:30/09/2021**

LAB REPORT
(The Drugs and Cosmetics Act 1940 and the Rules Thereunder)

Sample	***Guduchi Male (Tinospora Cordifolia)***	**A.R. No.**	09/2021/TP/035
Supplied by	Dr. Madhurima Bhargava	**Sample Quantity**	150gm
Batch No.	-	**D/M**	-
Date of receipt	19/09/2021	**D/E**	-

Sr.No.	TESTS	RESULTS	SPECIFIC REQUIREMENTS
1.	**Berberine Assay by HPLC**	0.03%	-
2.	**Microbial test**		
	a) Total Microbial Plate Count	500 cfu/g	1×10^5 cfu/g
	b) Total Yeast and Moulds	90 cfu/g	1×10^3 cfu/g
3.	**Pathogen Tests**		
	a) Escherichia coli	Absent	Absent
	b) Staphylococcus aureus	Absent	Absent
	c) Pseudomonas aeruginosa	Absent	Absent
	d) Salmonella Typhii	Absent	Absent
4.	**Heavy Metals Test**		
	a) Arsenic	Not Detected	3ppm
	b) Cadmium	Not Detected	0.3 ppm
	c) Mercury	Not Detected	1 ppm
	d) Lead	Not Detected	10 ppm
5.	**Test For Aflatoxins (by TLC)** (B1, B2 and G1,G2)	Not Detected	B1 & G1- NMT 0.5 ppm B2 & G2- NMT 0.1 ppm

qc_herbheal@ymail.com

Note:

1. Sample(s) not drawn by us unless otherwise stated.
2. Total liability of this laboratory is limited to the invoiced amount.
3. Test certificate in full or parts shall not be used for promotional or publicity purpose.
4. The sample will be destroyed after one month of the date of issued test report, unless otherwise specified.
5. All the above given specifications are provided by the supplier.

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Lic no. 1/ASU/TESTING LAB./PB./2014 **Date:30/09/2021**

LAB REPORT
(The Drugs and Cosmetics Act 1940 and the Rules Thereunder)

6.	Pesticide Residue(mg/Kg)	RESULTS	Pesticide Residue(mg/Kg)	RESULTS
	Endosulfan	Not detected	Dicholrvos	Not detected
	2-4' -DDT	Not detected	Malathion	Not detected
	Permethrin	Not detected	Parthion Ethyl	Not detected
	4-4' DDt	Not detected	Parthion Methyl	Not detected
	Gamma HCH	Not detected	2-4' DDE	Not detected
	Beta HCH	Not detected	2-4' DDD	Not detected
	Chlorpyrifos	Not detected	Alpha HCH	Not detected
	HeptaChlor	Not detected	Dieldrin	Not detected
	Aldrin	Not detected	Deltamethrin	Not detected
	4-4' DDD	Not detected	4--4' DDE	Not detected

Pooja 30/09/2021 — **Prepared By**

Manbeer 30/09/2021 — **Checked By**

Vinay 30/09/2021 — **Approved By**

"End of report"

qc_herbheal@ymail.com

Note:

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2. Total liability of this laboratory is limited to the invoiced amount. Sample consumed during test.
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A Government Approved Testing Laboratory
Lic no. 1/ASU/TESTING LAB./PB./2014 **Date:30/09/2021**

LAB REPORT
(The Drugs and Cosmetics Act 1940 and the Rules Thereunder)

Sample	***Guduchi female (Tinospora Cordifolia)***	**A.R. No.**	09/2021/TP/036
Supplied by	Dr. Madhurima Bhargava	**Sample Quantity**	80gm
Batch No.	-	**D/M**	-
Date of receipt	19/09/2021	**D/E**	-

Sr.No.	TEST	RESULTS	SPECIFIC REQUIREMENTS
1.	**Berberine Assay by HPTLC**	0.08%	-
2.	**Microbial test**		
	a) Total Microbial Plate Count	400 cfu/g	1×10^5 cfu/g
	b) Total Yeast and Moulds	60 cfu/g	1×10^3 cfu/g
3.	**Pathogen Tests**		
	a) Escherichia coli	Absent	Absent
	b) Staphylococcus aureus	Absent	Absent
	c) Pseudomonas aeruginosa	Absent	Absent
	d) Salmonella Typhii	Absent	Absent
4.	**Heavy Metals Test**		
	a) Arsenic	Not Detected	3ppm
	b) Cadmium	Not Detected	0.3 ppm
	c) Mercury	Not Detected	1 ppm
	d) Lead	Not Detected	10 ppm
5.	**Test For Aflatoxins (by TLC)** (B1, B2 and G1,G2)	Not Detected	B1 & G1- NMT 0.5 ppm B2 & G2- NMT 0.1 ppm

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CIN No.- U24233PB2008PTC032243

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Form No. 50 **Lic no. 1/ASU/TESTING LAB./PB./2014** **Date:30/09/2021**

LAB REPORT
(The Drugs and Cosmetics Act 1940 and the Rules Thereunder)

6.	Pesticide Residue(mg/Kg)	RESULTS	Pesticide Residue(mg/Kg)	RESULTS
	Endosulfan	Not detected	Dicholrvos	Not detected
	2-4' -DDT	Not detected	Malathion	Not detected
	Permethrin	Not detected	Parthion Ethyl	Not detected
	4-4' DDt	Not detected	Parthion Methyl	Not detected
	Gamma HCH	Not detected	2-4' DDE	Not detected
	Beta HCH	Not detected	2-4' DDD	Not detected
	Chlorpyrifos	Not detected	Alpha HCH	Not detected
	HeptaChlor	Not detected	Dieldrin	Not detected
	Aldrin	Not detected	Deltamethrin	Not detected
	4-4' DDD	Not detected	4--4' DDE	Not detected

Pooja 30/09/2021
Prepared By

Manbir 30/09/2021
Checked By

Viney 30/09/2021
Approved By

"End of report"

qc_herbheal@ymail.com

Note:

1. Sample(s) not drawn by us unless otherwise stated.
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3. Test certificate in full or parts shall not be used for promotional or publicity purpose.
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winCATS Planar Chromatography Manager

Herbal Health Research Consortium
Amritsar 143 001
Punjab

Analysis Report

Method	E:\JAN 2021 - DEC 2021\sept-2021\guduchi friday.cme	
Created by	Admin	Saturday, September 25, 2021 12:19:43 AM
Last modified by	Admin	Saturday, September 25, 2021 12:26:30 AM
SOP document		
Validated	Design	
Description :		
Analysis	E:\JAN 2021 - DEC 2021\sept-2021\guduchi friday 254 madhurima.cna	
Created/used by	Admin	Saturday, September 25, 2021 3:33:24 AM
Current user	Admin	

Stationary phase

Executed by	Admin	Saturday, September 25, 2021 3:33:07 AM
Plate size (X x Y)	5.0 x 10.0 cm	
Material		
Manufacturer		
Batch		
GLP code		
Pre-washing	No	
Modification	No	

Definitions - Quantification

Executed by	Admin	Saturday, September 25, 2021 3:33:07 AM

Calibration parameters

Calibration mode	Single level
Statistics mode	CV
Evaluation mode	Peak height

Samples

Sample ID: guduchi stem

Sample ID: standard

Sample ID: guduchi male

Sample ID: guduchi female

Substance name	Rf	Window size	Deviation	Purity	Manufacturer	Batch number	Expiry date	Product number
Berberine	0.59	4.4 mm	10.0 %	1.0000				

Standards absolute

Standard level1

Substance	Amount/fraction
Berberine	0.0100 mg

Sample application - CAMAG Linomat 5

Instrument CAMAG Linomat 5 "Linomat5_180745" S/N 180745 (1.00.12)
Executed by Admin Saturday, September 25, 2021 12:35:28 AM

Linomat 5 application parameters
Spray gas : Inert gas
Sample solvent type : Methanol
Dosage speed : 150 nl/s
Predosage volume : 0.2 ul

Sequence
Syringe size: 100 µl
Number of tracks: 4
Application position Y : 10.0 mm
Band length : 6.0 mm

No.	Appl. position	Appl. volume	Vial #	Sample ID	Active
>1	10.0 mm	5.0 µl	1	guduchi stem	Yes
>2	20.0 mm	5.0 µl	2	standard	Yes
>3	30.0 mm	5.0 µl	3	guduchi male	Yes
>4	40.0 mm	5.0 µl	4	guduchi female	Yes

Detection - CAMAG TLC Scanner

Information
Application position 10.0 mm
Solvent front position 75.0 mm

Instrument CAMAG TLC Scanner "Scanner_180710" S/N 180710 (2.01.02)
Executed by Admin Saturday, September 25, 2021 3:33:07 AM
Number of tracks 4
Position of first track X 10.0 mm
Distance between tracks 10.0 mm
Scan start pos. Y 5.0 mm
Scan end pos. Y 75.0 mm
Slit dimensions 4.00 x 0.30 mm, Micro
Optimize optical system Light
Scanning speed: 20 mm/s
Data resolution: 100 µm/step

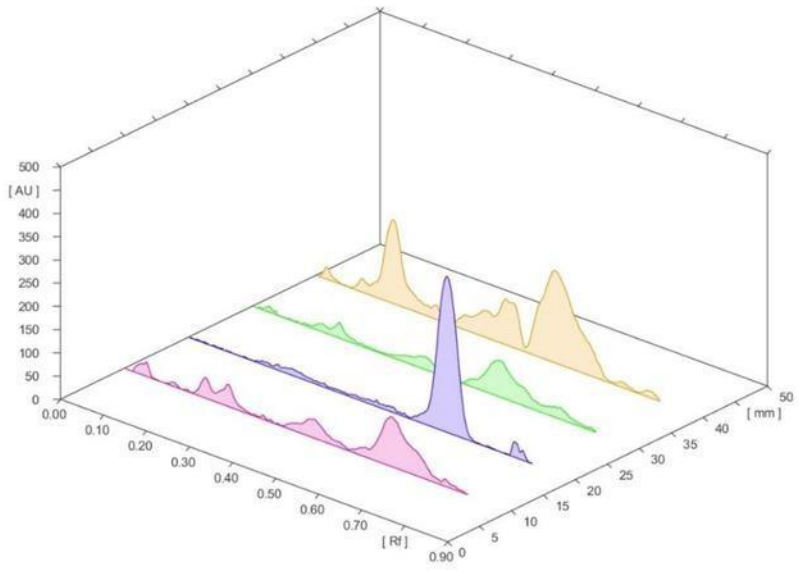
Measurement Table
Wavelength 254
Lamp D2 & W
Measurement Type Remission
Measurement Mode Absorption
Optical filter Second order
Detector mode Automatic
PM high voltage 317 V

Integration

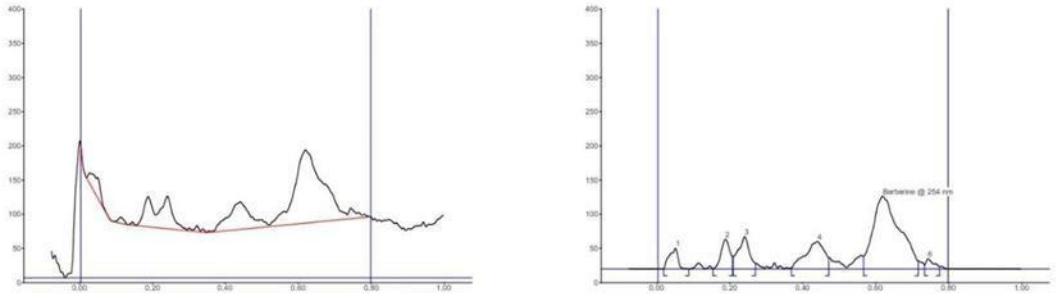
Properties
Data filtering Savitsky-Golay 7
Baseline correction Lowest Slope
Peak threshold min. slope 5
Peak threshold min. height 10 AU
Peak threshold min. area 50
Peak threshold max. height 990 AU
Track start position 10.2 mm
Track end position 62.1 mm
Display scaling Automatic

winCATS Planar Chromatography Manager

All tracks at WavelengthSc4

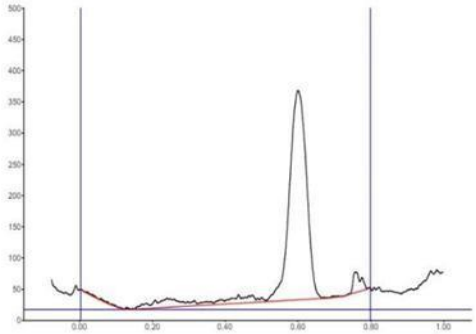


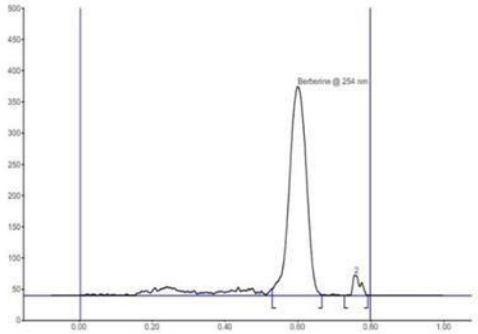
Track 1, ID: guduchi stem



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0.02	0.2	0.05	30.5	10.76	0.09	0.3	548.2	5.40	unknown *
2	0.15	0.6	0.19	43.7	15.41	0.21	18.5	799.6	7.87	unknown *
3	0.21	18.5	0.24	47.3	16.65	0.27	8.3	1114.0	10.97	unknown *
4	0.37	2.2	0.44	40.3	14.19	0.47	16.2	1597.4	15.73	unknown *
5	0.57	18.3	0.62	106.8	37.62	0.72	12.0	5845.1	57.55	Berberine
6	0.74	6.2	0.74	15.2	5.37	0.78	3.8	251.8	2.48	unknown *

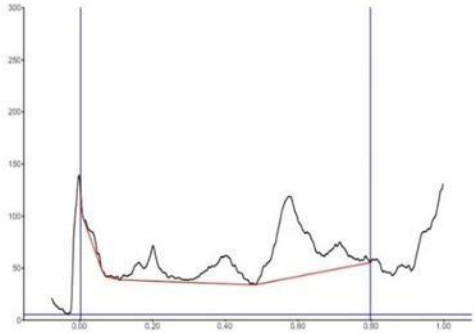
Track 2, ID: standard

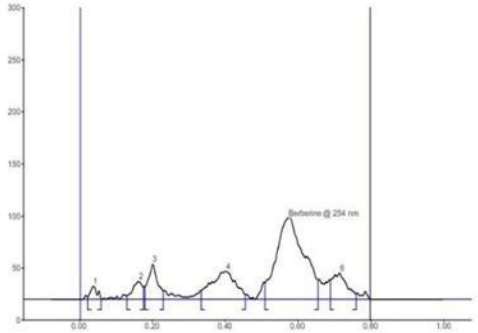




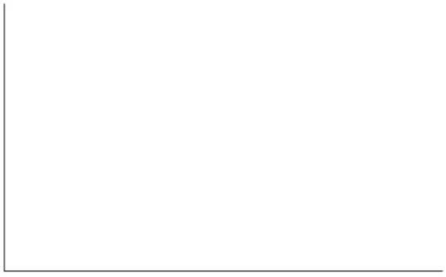
Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0.53	10.5	0.60	335.0	91.08	0.67	1.1	11739.1	95.67	Berberine
2	0.73	0.1	0.76	32.8	8.92	0.79	0.6	531.8	4.33	unknown *

Track 3, ID: guduchi male





Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0.02	2.6	0.04	12.6	6.49	0.06	0.3	175.5	2.27	unknown *
2	0.13	4.0	0.16	17.4	8.96	0.18	11.8	348.7	4.52	unknown *
3	0.18	12.5	0.20	33.9	17.43	0.23	8.3	650.4	8.43	unknown *
4	0.34	8.7	0.40	27.0	13.90	0.46	5.0	1345.3	17.43	unknown *
5	0.51	16.1	0.58	78.3	40.29	0.66	19.0	4429.2	57.38	Berberine
6	0.69	18.6	0.72	25.2	12.94	0.76	5.4	769.3	9.97	unknown *



winCATS summary report

Calibration results per Analysis

No results can be calculated due to the following error(s):
Berberine: Error found in single level calibration height

Herbal Health Research Consortium

Amritsar 143 001
Punjab

Analysis Report

Method	E:\JAN 2021 - DEC 2021\sept-2021\Gilov Samples.cme	
Created by	Admin	Wednesday, October 19, 2022 11:42:51 AM
Last modified by	Admin	Wednesday, October 19, 2022 11:44:51 AM
SOP document		
Validated	Design	
Description :		
Analysis	E:\JAN 2021 - DEC 2021\sept-2021\Gilov Samples_BS.cna	
Created/used by	Admin	Saturday, October 29, 2022 10:53:27 AM
Current user	Admin	

Stationary phase

Executed by	Admin	Saturday, October 29, 2022 9:58:44 AM
Plate size (X x Y)	5.0 x 10.0 cm	
Material	TLC Silica gel 60 F254	
Manufacturer	COM 6	
Batch		
GLP code		
Pre-washing	No	
Modification	No	

Definitions - Quantification

Executed by	Admin	Wednesday, October 19, 2022 11:45:12 AM

Calibration parameters

Calibration mode	Single level
Statistics mode	CV
Evaluation mode	Peak height

Samples

Sample ID: M I

Sample ID: F II

Sample ID: Standard

Sample ID: B-sitosterol

Sample application - CAMAG Linomat 5

Instrument	CAMAG Linomat 5 "Linomat5_180745" S/N 180745 (1.00.12)	
Executed by	Admin	Saturday, October 29, 2022 10:06:42 AM

Linomat 5 application parameters

Spray gas :	Inert gas
Sample solvent type :	Methanol
Dosage speed :	150 nl/s
Predosage volume :	0.2 µl

Sequence

Syringe size:	100 µl
Number of tracks:	3

Application position Y:	8.0 mm
Band length:	6.0 mm

No.	Appl. position	Appl. volume	Vial #	Sample ID	Active
>1	15.0 mm	15.0 µl	1	M I	Yes
>2	25.0 mm	15.0 µl	2	F II	Yes
>3	35.0 mm	5.0 µl	3	B-sitosterol	Yes

Detection - CAMAG TLC Scanner

Information

Application position	8.0 mm
Solvent front position	75.0 mm

Instrument

	CAMAG TLC Scanner "Scanner_180710" S/N 180710 (2.01.02)
Executed by	Admin Saturday, October 29, 2022 10:50:16 AM
Number of tracks	3
Position of first track X	15.0 mm
Distance between tracks	10.0 mm
Scan start pos. Y	5.0 mm
Scan end pos. Y	75.0 mm
Slit dimensions	4.00 x 0.30 mm, Micro
Optimize optical system	Light
Scanning speed:	20 mm/s
Data resolution:	100 µm/step

Measurement Table

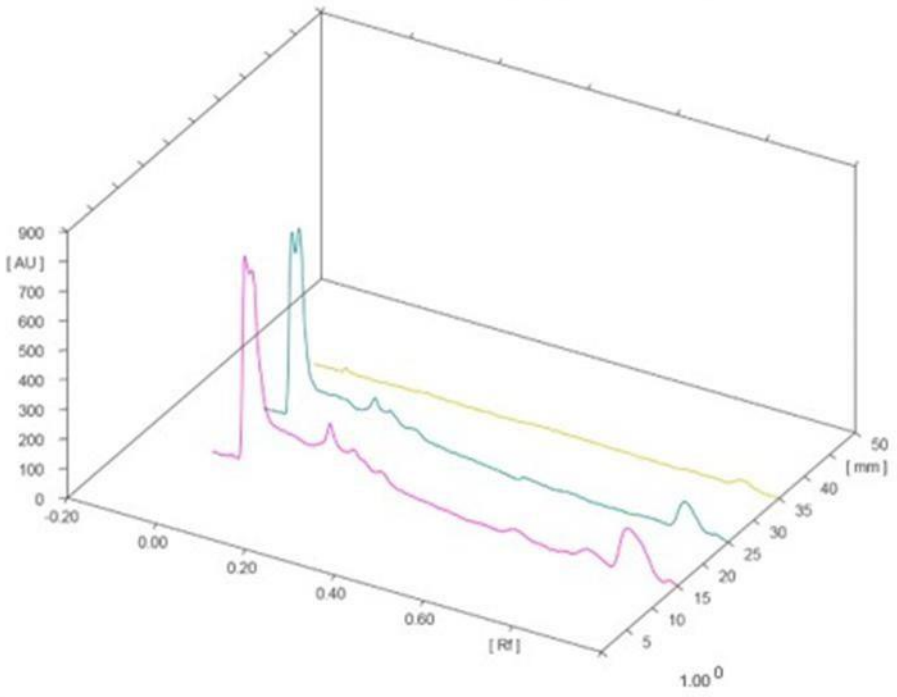
Wavelength	366
Lamp	D2 & W
Measurement Type	Remission
Measurement Mode	Absorption
Optical filter	Second order
Detector mode	Automatic
PM high voltage	372 V

Integration

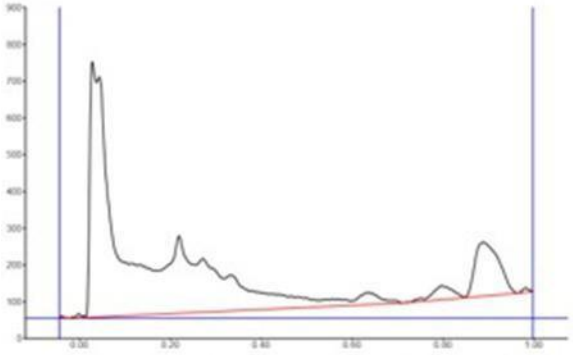
Properties

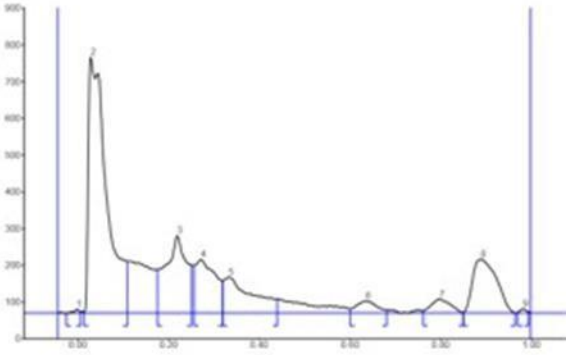
Data filtering	Savitsky-Golay 7
Baseline correction	Lowest Slope
Peak threshold min. slope	5
Peak threshold min. height	10 AU
Peak threshold min. area	50
Peak threshold max. height	990 AU
Track start position	5.0 mm
Track end position	75.0 mm
Display scaling	Automatic

All tracks at WavelengthSc4



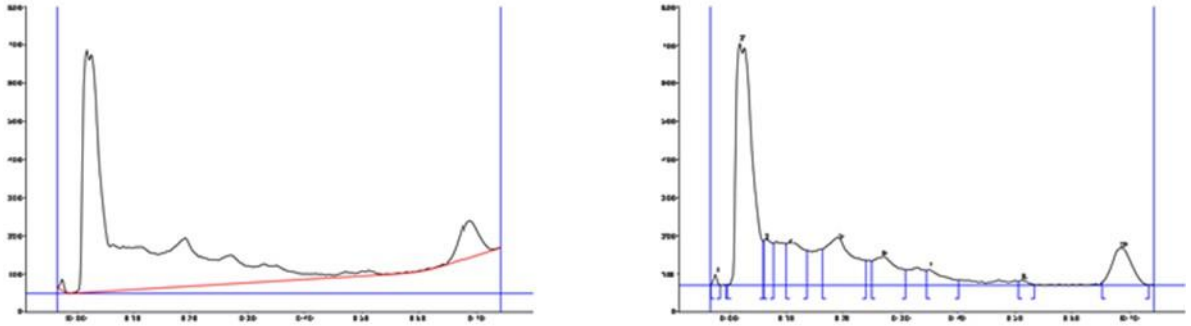
Track 1, ID: M I





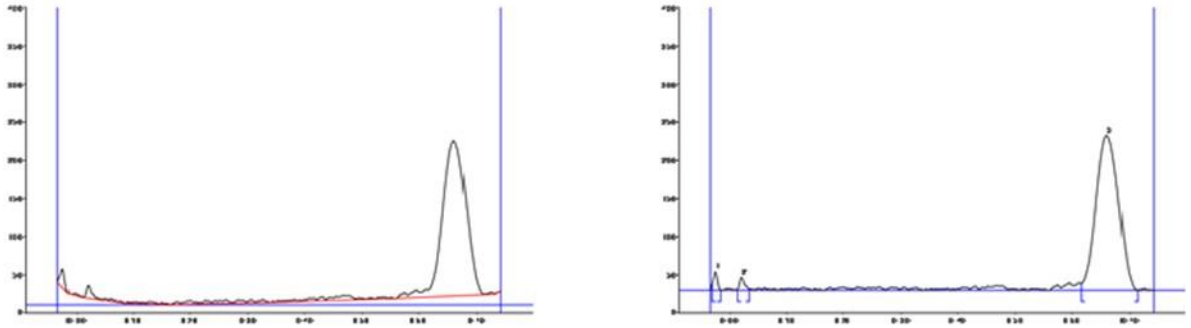
Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	-0.03	0.5	-0.00	10.6	0.76	0.01	4.6	95.5	0.20	unknown *
2	0.01	3.8	0.03	695.7	50.07	0.11	140.7	21810.5	45.41	unknown *
3	0.18	119.0	0.22	210.8	15.17	0.25	130.9	7588.6	15.80	unknown *
4	0.26	129.9	0.27	145.6	10.48	0.32	89.1	5147.4	10.72	unknown *
5	0.32	89.2	0.33	97.4	7.01	0.44	38.3	4816.9	10.03	unknown *
6	0.60	12.3	0.64	33.1	2.38	0.68	8.4	1161.4	2.42	unknown *
7	0.76	6.1	0.80	38.2	2.75	0.85	1.5	1263.0	2.63	unknown *
8	0.85	2.5	0.89	146.4	10.53	0.97	0.1	6018.2	12.53	unknown *
9	0.97	2.9	0.98	11.8	0.85	1.00	3.5	130.7	0.27	unknown *

Track 2, ID: MII



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	-0.03	2.6	-0.02	28.6	2.29	-0.02	0.4	195.2	0.50	unknown *
2	-0.01	2.0	0.02	633.1	50.77	0.06	116.5	18958.6	48.25	unknown *
3	0.06	117.1	0.06	120.8	9.69	0.08	110.7	1971.8	5.02	unknown *
4	0.10	109.7	0.10	110.9	8.90	0.14	91.4	3541.1	9.01	unknown *
5	0.16	95.2	0.19	126.7	10.16	0.24	65.6	6434.6	16.38	unknown *
6	0.25	64.1	0.27	74.7	5.99	0.31	41.5	3314.5	8.44	unknown *
7	0.35	39.4	0.35	41.4	3.32	0.40	13.5	1335.9	3.40	unknown *
8	0.51	10.7	0.51	13.5	1.08	0.53	1.9	223.2	0.57	unknown *
9	0.65	8.5	0.69	97.2	7.79	0.73	1.6	3315.0	8.44	unknown *

Track 3, ID: Berberine standard



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	-0.03	8.8	-0.02	24.1	9.86	-0.01	0.2	186.4	2.17	unknown *
2	0.01	0.7	0.02	16.9	6.95	0.03	1.9	162.2	1.88	unknown *
3	0.62	9.2	0.66	202.9	83.19	0.72	0.3	8262.0	95.95	unknown *

Results per track

winCATS summary report

Calibration results per Analysis

No results can be calculated due to the following error(s):
No substances assigned

LIST OF PUBLICATIONS:

1. Research paper on Comparative Study Between *Tinospora cordifolia* Male and Female Species with Physico-Chemical Analysis published In African Journal of Biological Sciences 10.33472/AFJBS.6. Si2.2024.3233-3241
2. Insights into Anti-Inflammatory Activity of Tinospora cordifolia: A Comparative Docking Study of Natural and Synthetic Compounds
3. Publication Pharmacovigilance programme for Ayush Drugs. A Nation level competition on Pharmakos vigilance for Ayush drugs at present scenario and future prospects on 6th ayurveda day of 75th Independence Day celebration.
4. Paper presented on topic Guduchi "A MIRACLE PLANT ' in 3rd international conference on functional material manufacturing performance ICFMMP-29,30 July 2022 at Lovely Professional University pb.
5. Paper Presentation on GUDUCHI PLANT in Ayur Expo and Ayur Ayog fair 2021with Ayush Mantralya.
6. Paper presentation on competitive evaluation of physico chemical para meter of Guduchi Ghana based on dioecism in Guduchi Plant by ICP (International conference of Pharmacy 2022 by School of Pharmacy and IPA held on 9,10 November 2022 in Lovely Professional University.
7. Participated in Awareness programme to introduce Giloye in Routine Life for the promotion of Health on 26 March 2022 at Lovely Profession University pb.

Ref. PBRI/IAEC/2024137

Date 17-01-2024

CERTIFICATE

This is to certify that the following project has been approved by the Institutional Animal Ethical Committee, Pinnacle Biomedical Research Institute (PBRI), Bhopal (Reg. No. 1824/PO/RcBi/S/15/CPCSEA).

Project Title	Impact of Dioecism On Quality and Efficacy of Sanshamni Vati
Protocol Approval Number	PBRI/IAEC/16-01-23/025
Investigator Name	Madhurima
Approval Date	16-01-2023

Pbri
Pinnacle Biomedical Research Institute

Dr. Mohd. Azaz Khan
Chairperson, IAEC
PBRI

IAEC
Pinnacle Biomedical Research Institute
BHOPAL (M.P.)

https://africanjournalofbiomedicalresearch.com/index.php/AJBR
Afr. J. Biomed. Res. Vol. 28(1s) (January 2025); 793-799
Research Article

Insights into Anti-Inflammatory Activity of *Tinospora cordifolia*: A Comparative Docking Study of Natural and Synthetic Compounds

Madhurima[1], Pankaj[2], Manish Vyas*, Sanjeev Kumar Sahu

School of Pharmaceutical Sciences, Lovely Professional University, Phagwara, Punjab, India-144411,

***Corresponding Author:** Dr. Manish Vyas
manish.17410@lpu.co.in

Abstract:
Inflammation is an essential immune response, but its dysregulation can result in chronic diseases. The quest for effective anti-inflammatory therapies has led to significant research into both synthetic drugs and natural bioactive compounds. This study explores the comparative anti-inflammatory potential of berberine, β-sitosterol, and celecoxib through in silico molecular docking techniques.
The docking analysis focused on evaluating the binding affinities of these compounds to key pro-inflammatory targets, including cyclooxygenase-2 (COX-2), which are critical regulators of inflammation. Using AutoDock and Discovery Studio software, ligand-receptor interactions, binding energies, and interaction patterns were systematically analysed.
This study highlights the potential of natural compounds in anti-inflammatory drug discovery and demonstrates the

AFJBS African Journal of Biological Sciences

Article ID: AP-1705

Date: 30-05-2024

Dear Author(s)

Madhurima[1], Manish Vyas[2*], Isha Agrawal[3], Pankaj[4]

Lovely school of pharmaceutical science, Lovely Professional University, India

We would like to inform you that your manuscript has been accepted for publication in African Journal of Biological Sciences ISSN: 2663-2187

Manuscript Title: "Comparative Study Between Tinospora cordifolia Male and Female Species with Physico-Chemical Analysis".

Thanks for submission of your work with us.

Regards,

Eugene A Silow

Editor in Chief
African Journal of Biological Sciences

ISSN: 2663-2187

https://www.afjbs.com

https://doi.org/10.33472/AFJBS.6.Si2.2024.3233-3241

African Journal of Biological Sciences

Comparative Study Between *Tinospora cordifolia* Male and Female Species with Physico-Chemical Analysis

Madhurima[1], Manish Vyas[2*], Isha Agrawal[3], Pankaj[4], Navneet Khurana[5], Neha sharma[6], Pramod Yadav[7]

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Article History
Volume 6,Issue Si2, 2024
Received:13 Mar 2024
Accepted : 16 Apr 2024
doi: 10.33472/AFJBS.6.Si2.2024.3233-3241

ABSTRACT
Guduchi, commonly known as *Tinospora cordifolia* or *Giloy*, is a highly esteemed medicinal plant in traditional Indian medicine, known for its many therapeutic properties. Although it is frequently exploited, detailed comparative studies of the male and female plants of this species are lacking. This study aims to reveal the differences between male and female *T. cordifolia* plants by a comprehensive physiochemical analysis. Also, a comparative study between the *Kwatha* prepared by both samples were analyzed. Physicochemical analysis revealed that while both genders contain similar types of bioactive compounds, there are notable quantitative differences between them. Both male and female *T. cordifolia* plants exhibit nearly identical results, showing minimal discernible differences like pH values, alcohol soluble extractive values and ash values in which female variety showing slightly better values than male plant of *Guduchi*. These findings emphasize the need to consider plant gender when using *T. cordifolia* medicinally, suggesting that targeting the use of male or female plants could improve therapeutic effects for specific health conditions.

Keyword: Guduchi, Giloy, Physico-chemical

Introduction:

Tinospora cordifolia (T.cordifolia), known as *Guduchi*, is a medicinal plant that has been used in traditional Ayurvedic medicine for centuries. *T. cordifolia* is known for its immunomodulatory, antioxidant, and anti-inflammatory properties. It is also believed to be effective in treating various conditions such as fever, diabetes, and skin diseases. In this comparative study, we will analyze the physico-chemical differences between *T. cordifolia* male and female plants. This study aims to provide a better understanding of any variations in the chemical composition of the two genders of

PHARMACOVIGILANCE PROGRAMME FOR ASU&H DRUGS
INTERMEDIARY PHARMACOVIGILANCE CENTRE FOR AYURVEDA
Institute of Teaching and Research in Ayurveda, Jamnagar
(Institute of National Importance)
Ministry of AYUSH, Govt. of India

CERTIFICATE OF PARTICIPATION

आज़ादी का अमृत महोत्सव

This is to certify that

Dr. Madhurima Ph.D Scholar.

Lovely Professional University.

Has participated in "National Level Essay Writing Competition" Organized by IPvC, ITRA, Jamnagar, Ministry of AYUSH, Government of India on the occasion of 6th AYURVEDA DAY & 75th INDEPENDENCE DAY CELEBRATION

Director
ITRA, Jamnagar

Coordinator
IPvC, ITRA, Jamnagar

Certificate No.246137

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NATIONAL MEDICINAL PLANTS BOARD

Certificate of Participation

This is to certify that **Dr. Madhurima** of **Lovely Professional University** has participated in the **"Awareness Program to Introduce Giloy in Routine Life for The Promotion of Health"** held on **26th March 2022**, organized under the funded project entitled ***"National Campaign on Amrita for Life (Tinospora cordifolia)"*** at Lovely Professional University, Punjab.

Date of Issue: 18-05-2022
Place: Phagwara (Punjab), India

Prepared by
(Administrative Officer-Records)

Dr. Manish Vyas
Convener

Dr. Sorabh Lakhanpal
Convener

Serial No. 31CP2022-1525

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Certificate of Participation

This is to certify that Prof./Dr./Mr./Ms. Madhurima has successfully participated as Delegate & Presented Poster/ Oral Presentation on Comparative Evaluation of Physico-Chemical Parameters of Guduchi Ghana based on Dioecism in Guduchi Plant in the 3rd International Conference of Pharmacy (ICP-2022) on the Theme of "Practice, Promotion & Publication of Innovation : A Way of Transforming Health" held on 09th & 10th November 2022 organized by School of Pharmaceutical Sciences in a collaboration with Indian Pharmaceutical Association (IPA) at Lovely Professional University, Punjab.

Mr. Suresh Khanna, National Hon. Gen. Secretary, IPA
Dr. Bimlesh Kumar, Organizing Secretary
Dr. T.V. Naryana, National President IPA
Dr. Monica Gulati, LOC Chairperson

Sri Herbasia Biotech | Tishk International University, Iraq

Pharmacovigilance for ASU& H Drugs – Present Scenario and Future Prospects

- **Dr. Madhurima,** Ph. D Scholar
Lovely Professional University, Phagwara, Punjab
Email id: drmbdav@gmail.com

Pharmacovigilance is the study and observation of pharmaceutical drugs. Pharmacovigilance consists of two words, 'pharmakin' from Greek meaning drug and 'vigilore' from Latin meaning to be awake and monitor. Pharmacovigilance is a discipline of science that concentrates to encounter, explore, evaluate, perception, and prevention of inauspicious reactions of medicine. In nutshell, we can say this term describes drug safety, collection analysis, monitoring, and prevention of adverse effects and therapies. It's for doctors to do good and enough for the patient's care. Monitoring and testing should be the parameter for daily use. Pharmacovigilance aims to improve patient safety by determining the use of medicines and all medical and paramedical arbitration; In case of any medical difficulties, providing solutions in a timely manner; providing drug safety and promoting knowledge access of pharmacovigilance for its effective communication to the general public. Nowadays Ayush medicines have an identity across the world that it is a widely practical system of medicine. In India, Ayurveda is considered the safest medical system as ayurvedic physicians use herbal minerals, metals, animal originated drugs for different diseases. Many people have faith in Ayush but several adverse effects are also reported because Ayush drugs are also used by non-qualified physicians without any guidance. The spirit of Pharmacovigilance is alive in the ayurvedic text but it is not directly mentioned in it. Pharmacovigilance is referred to as 'the detection, assessment, understanding, and prevention of adverse effects of drugs or any other possible drug-related problems.

Holistic approach of this science visualize:

स्वस्थस्य स्वास्थ्य रक्षणं।
आतुरस्य विकार प्रशमनं च: ॥.

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Certificate No.253394

Certificate of Presentation

This is to certify that **Dr./Mr./Ms. Madhurima Bhargava** of **School of pharmaceutical Sciences, Lovely Professional University, Phagwara** has presented a paper on **Guduchi - A miracle plant** in the **"3rd International Conference on Functional Materials, Manufacturing and Performances (ICFMMP-2022)"** held on July29-30th, 2022, organized by **Division of Research and Development, Lovely Professional University, Punjab**.

Date of Issue: 30-08-2022
Place: Phagwara (Punjab), India

Prepared by
(Administrative Officer-Records)

Dr. Hitesh Vasudev
Convenor

Dr. Pranav Kumar Prabhakar
Organizing Secretary

Dr. Chander Prakash
Conference Secretary

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MINISTRY OF SCIENCE AND TECHNOLOGY