The Study Protocol of "Pharmaceutico Analytical and in Vitro Antimicrobial Activity of *Gunjadya Taila* Prepared by Using *Murchchhita* and *Amurchchhita Tila Taila*"

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Abstract

Background: In this modern era life is full of glamour and faced many health related problem. In modern science there are huge numbers of drugs said for treatment of diseases but these drugs get resistance to that particular disease. Traditionally medicinal plants have been used for many years as topical and internal administration in the treatment of fungal and bacterial diseases. In *Rastarangini Gunjadya Taila* mentioned in *Kushta and Kandu*. In Ayurveda all skin diseases are involved in *Kushta Roga*. Skin diseases mainly caused by microorganism as per modern science such as *Staphylococcus* aureus, streptococci and Candidiaalbican etc. Hence to provide scientific data and statistical validation the study on pharmaceutical analysis and in vitro antimicrobial activity of *Gunjadya Taila* prepared by *Murchchhita* and *Amurchchhita Tila Taila* is undertaken.

Objective: To study antimicrobial activity of Gunjadya Taila prepared by using *Murchchhita* and *Amurchchhita Tila Taila*.

Material and Method: *Gunjadya Taila* will be prepared in *Rasashastra* department by two method one with *Murchchhita Tila Taila* and other with by using *Amurchchhita Tila Taila*. The prepared drug *Gunjadya Taila* will be evaluated for organoleptic characters and physicochemical parameters. *Gunjadya Taila* will be assessed for antimicrobial effect on gram positive bacteria,gram negative bacteria and fungal group. Inhibition of microbial growth will be investigated by diffusion method.

Observation and Results: The data will be assessed for its antimicrobial and antifungal effects by using ANOVA test & unpaired 't' test.

Conclusion: This research work on *Gungadyataila* will show all analytical parameters within range and significant effects on gram positive and gram negative bacteria.

Keywords: *Gunjadya Taila, Kushta, Skin Disease, Antimicrobial activity.*

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Introduction

Traditionally medicinal plants have been used for many years as topical and internal administration in the treatment of fungal and bacterial diseases. Medicinal plants are considered as new resources for producing agents that could act as alternative to antibiotics in the treatment of fungal and bacterial diseases. Skin disease is a common ailment affecting all age groups[1]. Skin conditions such as purities, scabies impetigo, eczema

and molluscumcontagiosum ranged from the 2nd to 11th leading cause of disability in India. Skin conditions were account the fourth leading cause of nonfatal disease burden at global level and affect nearly 900 million population throughout world[2].Skin infections categorized as primary or secondary. Primary infections have specific characteristic morphologies, are originated by lone organisms and routinely occur in normal skin. They are often caused by *Staphylococcus aureus*, *Streptococcus pyrogenes* and coryne form bacteria and responsible to develop erythrema, impetigo, boils and folliculitis. Secondary infections initiate in diseased skin as a superimposed form. e.g. Impetigo and toe web infections[3].There are various topical and systemic synthetic drug available in the market but they have various adverse effect.

Ayurveda is a complete and holistic traditional health-care system of India that contains both preventive and therapeutic aspects. Bhaishajya (drug) is the pride and part of Ayurveda and recognized as one of the piller of therapy used to conquer the deadly diseases[4]. In modern science there are huge number of drugs said for the treatment of diseases but these drug get resistance to that particular diseases. There are large number of drugs said in Ayurvedic literature.In *RastaranginiGunjadya Tail*is mentioned for the treatment of*Kushta and Kandu*[5].In ayurveda all skin diseases are involveed in *KusthaRoga*. As per modern science skin disease caused by micro-organisms such as stapphlococcus aureus, strepto- cocci and candidiaalbican. Hence to provide scientific data and statistical validation the study on pharmaceutical analysis and antimicrobial activity of *Gunjadya Taila* prepared by *Murchchhit*and *Amurchchhittilataila* is undertaken.

Rationale of the study: Skin diseases commonly occur in children and adult scan direct to disability. At global level, it accounts for the 4th leading source of nonfatal disease burden. Various research studies conducted proved that skin diseases can have a major impact on the quality of life of those suffered. Skin cancer and infections are some of the potentially life-threatening skin diseases and involve high costs for the individual and society. In Ayurveda and in modern science,several theories have been put forward describing this disorder. As in modern medical science its treatment is symptomatic with bothersome side effects, there is an ample possibility to work out on its etio-pathology and management aspect of the skin diseases. In Ayurveda, *Gunjadya Tail* is described in skin diseases but its preparative and analytical standards are not established yet .This study will fulfill this gap. This study is an attempt to prepare and standardise *Gunjadya Taila*with *Murchchhita* and *Amurchchhita Tila Taila* and to test its efficacy through antimicrobial activity.

Material and Method

A Pharmaceutical study: This study is related to drug preparation in which three different batches of *Gunjadya Taila* will be prepared to establish pharmaceutical standardization. Pharmaceutical study will be done in following steps;

I. **Procurement of Raw materials:** All required raw materials will be procured from field and authentic reliable sources.

II. **Authentication of Raw materials:** Raw drugs will be indentified and authenticated by Taxonomist. Raw drug will be standardized as per A.P.I.

III. **Process of Taila Murchchhana[6]:** *TailaMurchchhana* will be done as per SOP . Ingredients required for *Taila Murchchhana* mentioned in Table No. 1.

Standard operative procedure of *Murchchhana* of *Tila Taila:* All the collected and authenticated drugs will be cleaned (to remove foreign matter).

Individual

↓

The individual drugs will be then powdered using pulvelizer.

↓

All individual drugs will be sieved through mesh no. 80.

↓

The required quantity of powder will be taken.

↓

Powder of drugs will be mixed with sufficient quantity of distilled water to prepare *Kalka*.

↓

Kalka (1/4th part), *Tila Taila*(1part) and distilled water (4 parts) will be taken in clean and wide mouth steel vessel.

↓

The above mixture will be heated on *Mandagni*(low flame - gently boiling) with frequent

Stirring till *SnehasiddhiLakshanas*appears.

⬇

*Taila*will be allowed to get *SwangSheeta.*

⬇

After cooling *Taila*will be filtered through doubled muslin cloth and stored in air tight glass

Bottle.

⬇

Murchchhita Tila Taila

IV. Purification of GunjaBeejain Godugdha[7]: *Gunjabeeja* (seeds of *Abrusprecatorius*) will be purified as per reference with the help of *Dolayantra* in *Godugdha***.**

V. Preparation of *Bhrungaraja Swaras*[8]: Fresh *Bhrungaraja* leaves will be taken and will be crushed to a paste and finally expressed through white cloth to obtained clear liquid.

VI. Preparation of *Gunjadya Taila*[5]:

Ingredients required for preparation of *Gunjadya Taila* mentioned in Table no 2.

Preparation of *Gunjadya Tail* by using *Murchchhita Tila Tail*

Kalka from *Gunja Beeja & Bhrungaraja Swaras* will be prepared from fresh plant

⬇

Obtained *Gunja Beea Kalka 1part+Murchchhita Tila Taila* (4 part) + *Bhrungaraja Swaras* 16 part

⬇

Will be taken in a wide mouth steel vessel.

⬇

Above mixture will be heated on *Mandagni* (low flame-gently boiling) with frequent stirring till the *Snehasiddhi Lakshans* appears.

⬇

Taila will be allowed to get *Swang Sheeta* (self cooled)

⬇

After cooling *Taila* will be filtered through doubled muslin cloth and stored in air tight glass

Bottles

⬇

Gunjadya *Taila* prepared by *Murchchhita Tila Taila*

Preparation of *Gunjadya Taila*by using A*murchchhita Tila Taila:*

Kalka from *Gunja Beeja & Bhrungaraja Swaras* will be prepared from fresh plant

⬇

GunjaBeeja Kalka 1part + *Amurchhita Tila Taila* (4 part) + *Bhrungaraja Swaras* 16 part will be taken in a wide mouth steel vessel.

⬇

Above mixture will be heated on *Mandagni* (low flame-gently boiling) with frequent stirring till the *Snehasiddhi Lakshans* appears.

⬇

Taila will be allowed to get *Swang Sheeta* (Self cooled)

⬇

After cooling *Taila* will be filtered through doubled muslin cloth and stored in air tight glass Bottles

⬇

Gunjadya *Taila* prepared by *Amurchchhita Tila Taila*

The research drug will be prepared in 3 batches with same quantity of ingredients and same condition one with *MurchchhitTila Taila*and other with *AmurchchhitTila Taila.*

B. Analytical study

a. Subjective Parameters of finished drugs:

Description (Organoleptic characters)[9]: The prepared drug Gunjadya Taila will be evaluated by organoleptic characters.

1. *Sparsha.*(Sensation)
2. *Rupa*.(Colour)
3. *Gandha.*(Smell**)**

b. Objective Parameters of finished drugs:

Description (Physicochemical parameters)[10]

1. Specific Gravity
2. Refractive index at 25^0C
3. Viscosity
4. Acid value
5. Saponification value
6. Iodine value
7. Peroxide value
8. Unsaponificable matter
9. HPTLC or GC-MS (Quantitative)

C. Antimicrobial study[11-13]: Antimicrobial activity is tested byvarious method but in present study Agar well diffusion method will be used.

Principle: The antimicrobial present in the plant extract are allowed to diffuse out into the medium and interact in a plate freshly seeded with the test organism. The resulting zone of inhibition will be uniformly circular as there will be a confluent lawn of growth . The diameter of zone of inhibition can be measured in millimeter as shown in Table no 3,4,5.

C1: Selection of test microorganism: The bacterial strains used for the experiment will be procured from dept. of microbiology JNMC, Wardha

Selection of test microorganism

(a) Gram Positive bacteria:

1. Staphylococcus aureus
2. Streptococcus pyrogen

(b) Gram Negative bacteria:

1. Escheria coli
2. Klebsiellapneumoni

(c) Fungal group: 1 Candida albicans

C2: Positive control group drugs[14]

For gram positive bacteria - Azithromycin 50ug/6ul

For gram negative bacteria - Ciprofloxacin 30ug/6ul

For candida albicans - Clotrimazole 50ul/6ul

Sample Size:

1. *Tila Taila*-1sample
2. *Gunjadya Tail* aprepared by *Murchchhit Tila Taila*-1 sample
3. *Gunjadya Taila* prepared by *Amurchchhit Tila Taila*-1 sample
4. Gram positive bacteria-2 samples
5. Gram negative bacteria-2 samples
6. Fungal group -1 sample
7. Doses of oil -3 samples
8. Positive control group – 3 samples

Method of data analysis: Statistical analysis will be carried out by institutional statistician. Statistical assessment will be done throughunpaired 't' test and ANOVA test.

Results

Gunjadya Taila will show antimicrobial activity against gram +ve, gram -vebacteriaand fungal group

Table 1: List of ingredients used for *Tila Taila Murchchhana*

Sr No	Dravya	Part to be used	Proportion
1	*Manjishtha (Rubiacordifolia* Linn*)*	Stem	¼ part
2	*Haridra (Curcuma longa.*Linn*.)*	Rhizome	1/4 part
3	*Lodhra (Symplocosracemosus*.Roxb*)*	Stem bark	1/4 part
4	*Musta (Cyperusrotundus* Linn)	Tuber	1/4 part
5	*Amalki* (*Emblica officinalis* Gaertn)	Pericarp	1/4 part
6	*Bibhitaki* (*Terminalia bellirica* Roxb)	Pericarp	1/4 part
7	*Haritaki* (*Terminalia chebula* Retz)	Pericarp	1/4 part
8	*Vatankur (Ficusbengalensis* Linn*)*	Leaf bud	1/4 part
9	Tila Taila		1 part
10	Water		4part

Table 2: Contents of *Gunjadya Taila*

Sr No	Contents	Proportion
1	Gunja Beeja Kalka	1 part
2	Tila Taila	4 part
3	Bhrungaraja Swaras	16 part

Discussion

To treat infections with plants and their extracts is known for centuries and has been practiced all overthe world. As modern medicines are costlier and associated with side effects, herbal medicines are gaining growing interest among the people due to its cost effective and all time availability. In the last three decades, even though pharmacological industries have manufactured number of new antibiotics, resistance to these drugs by microorganisms has been increased. Hence, more and more studies with respect to the use of herbal drugs as therapeutic agents is the need of the hour, especially those related to the control of antibiotic resistant microbes. Hence, this study is proposed to asses pharmaceutico analytical and in vitro antimicrobial activity of *'Gunjadya Taila'* prepared by using *Murchchhita* and *Amurchchhita Tila Taila.*

Conclusion

In vitro study on *Gunjadya Taila* will show significant antimicrobial effect on gram positive and gram negative bacteria.

Ethical Clearance: Taken from institutional ethics committee.

Source of Funding: Self.

Conflict of Interest: Nil.

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