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Pharmaceutico- Analytical Study of Nimbadi Ubatan and It's **Antimicrobial Activity.**

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ABSTRACT:

Aim: To Study Physico chemical analysis of Nimbadi ubatan and it's antimicrobial activity.

Objectives: 1) Preparation of *Nimbadi* ubatan. 2) To Study literature of dravyas used in preparation of *Nimbadi ubatan*. 3) Study of Microbes used to antimicrobial activity of Nimbadi ubatan. Materials and Methods: Preparation of Nimbadi ubatan according to the reference Astang hrudaya chikitsa sthana kusthchikista adhyay 19/65-66. Analytical Nimbadi study of ubatan and antimicrobial activity of Nimbadi ubatan was performed by time kill test method. **Discussion:** As per reference of Astang hrudaya, Nimbadi ubatan was prepared and was analysed. Physico chemical parameters of *Nimbadi ubatan* was also done.

Antimicrobial activity of Nimbadi ubatan shown 2.25_4 log reduction / 99% reduction of S. aureus, S. Pyogenes, p. aeruginosa and Candida albicans in 1 minute. Conclusion: The present Study entitled "pharmaceutico analytical study of Nimbadi ubatan and it's antimicrobial activity" was aimed to prepare perform all analytical test and antimicrobial analysis of Nimbadi ubatan.

KEYWORDS: Nimbadi ubatan, Astang hrudaya, Antimicrobial activity,

INTRODUCTION:

Shastra Bhaishajya Rasa and Kalpana is one of the branch of Ayurveda in which all methods of ayurvedic medicine purification, formation, dose and indication are available. Ashtang hrudaya is a very famous samhita which has many kalpas for various diseases.

Now a days with increasing competition in every field, the fast life with unhealthy eating habits like fast food, Milk shakes (*Virudha aahar*) and lack of exercise, lack of sleep, stress, more use of electronic gadgets (*Mithya Vihar*) add to the cause of skin diseases.

All above mentioned hetus can be correlated with hetus described in *Kushtha vyadhi*. In *Astang Hrudaya*, *Nimbadi ubatan* is a formulation mentioned in Kushtha adhikar, it can be used as varnya and to treat skin disorders in the form of udvartan, churna or as lepa.

In ayurvedic classics, according to acharya vaghbhata, the word kustha is ''Kusnati vapu'' which means the disease that makes the body disfigured and ugly. Kustha is a condition in which different body parts dhatu, upadhatu are destroyed. Even though the term kustha is used in ayurveda for entire dermatological problem, all the acharyas of bruhatrayee unanimously have accepted the numerology of kushtha as eighteen.

Giving friction to the body by kashayadi choorna is called as *udvartana*. The concept of *udvartana* is explained right from the oldest textbook charaka Samhita

and most of the other authors under *rookshana* karma which is one among the shadvidhopakrama. The word meaning of *rookshana* is making thin or inducing dryness^{.[1,2,3]}

AIM: To Study *Physico* chemical analysis of *Nimbadi ubatan* and it's antimicrobial activity.

OBJECTIVES:

- 1) Preparation of Nimbadi ubatan.
- 2) Study of Microbes used to check antimicrobial activity of *Nimbadi ubatan*.

MATERIALS AND METHODS:

Preparation of Nimbadi ubatan:[4]

Reference:

N J-R A S

निम्बादि उबटन –

निम्बंहरिद्रेसुरसंपटोलंकुष्ठाश्चगंधेसुरदारुशिगुः।

ससर्षपंतुम्बरुधान्यवन्यंचण्डाचचूर्णानिसमानिकुर्यात।

तैस्तक्रपिष्टैः प्रथमंशरीरं तैलक्तमुद्धर्तयितुं यतेत।

तथास्यकण्डूःपिटिकाःसकोठाःकुष्ठानिशोफाश्चशमंबजन्ति॥

(अ.ह.चि.१९/६५–६६)

Table No. 1: Herbal raw materials for Preparation of Nimbadi Ubatan: [5,6]

Coarse powder (Pata churna) form with mesh no.60.

Sr. No.	Name	Latin Name	Part Used	Quanity
1	Nimb	Azadirachta indica	Patra	50 gms
2	Haridra	Curcuma longa	Kand	50 gms
3	Daruharidra	Berberis aristata	Twak	50 gms

4	Surasa	Ocimum sanctum	Вееј	50 gms
5	Patol	Tricosanthes dioica	panchang	50 gms
6	Kushtha	Saussurea lappa	Mool	50 gms
7	Ashwagandha	Withania somniferra	Mool	50 gms
8	Surdaaru	Cedrus deodaru	Saar	50 gms
9	Shigru	Moringa pterygospermum	Twak	50 gms
10	Sarshap	Brassica campestris	Вееј	50 gms
11	Tumbaru	Zanthoxylum alatum	Phal	50 gms
12	Musta	Cyperus rotundus	Kand	50 gms
13	Chanda	Angelica glauca	Mool	50 gms

Procedure of Preparation of *Nimbadi Ubatan*: [4,7]

- Coarse powder (pata churna) of the ingredients of Nimbadi Ubatan is required for this Kalpana.
- Coarse powder (*pata churna*) of all the ingredients of *Nimbadi Ubatan* was purchased from local market.
- ➤ All the ingredients of *Nimbadi Ubatan* were authentified from certified Lab.
- All the ingredients of *Nimbadi Ubatan* were weighed on a weighing scale.
- ➤ 50 gms of each of the raw material were taken.
- Coarse powder of all the ingredients of *Nimbadi Ubtan* was taken in sieve no. 60 according to the sequence explained in the shloka.
- Fequal quantity of *Nimba* Powder, *Haridra* powder, *Daruharidra* powder, *Surasa* powder, *Patol*

Powder, Kushtha powder, Ashwagandha powder, Shurdaru powder, Shigru Powder, Sharshap Powder, Tumbaru Powder, Musta Powder, Chanda Powder were taken into sieve respectively.

- J-D-After sieving all the above ingredients, the mixture was taken into *khalvayantra* for *mardan* process.
 - ➤ 80 ml of Takra was added to the mixture and it was triturated in *khalvayantra* for 30 minutes.
 - Another 50 ml of *Takra* was added to the mixture and triturated in *khalvayantra* for 40 minutes.
 - After trituration, mixture was taken out of the *khalvayantra*.
 - ➤ The final product obtained was then weighted on a weighing scale which was 650 gms of *Nimbadi Ubatan* and kept in an air tight container.







PREPARATION OF NIMBADI UBATAN:



TRITURATION OF NIMBADI UBATAN WITH TAKRA



Antimicrobial activity of Nimbadi ubatan by Time kill test method: [8,9]

The Time Kill Analysis measures the change in a population microorganisms within a specified sampling time after exposure to an antimicrobial test material in vitro. The test article or its dilution is brought into contact with a known population of microorganisms for a specified period of time at a specified temperature. The test article is then neutralized at the target sampling time and the surviving organisms are enumerated. The percent and/or log10 reduction from either an initial microbial population or test blank is calculated. The four organism which were taken to perform antimicrobial activities Staphylococcus those Aureus.

Streptococcus *pyogenes*, Pseudomonas *aeruginosa*, Candida *albicans* which causes skin infections like boils, *folliculitis*, impetigo, *cellulitis* etc.

Procedure of Antimicrobial activity of *Nimbadi ubatan* by Time kill test method:

Test product was inoculated with test inoculum individually (approximately 10⁸ CFU/ ml). After the specified exposure time of 1 minute, surviving microorganisms were recovered by drawing an aliquot, neutralizing it and

- performing Standard Pour Plate Technique.
- ➤ Culture count was ascertained by dilution Blank. Adequate Validation of Neutralizing agent was also carried.

> Test was carried out in duplicate and average count was taken as CFU/ ml.

OBSERVATION AND RESULTS:[10]

Table no. 2 Observations and Result of Analytical process of Nimbadi Ubatan:

Sr. No.	Test	Result
1	Appearance	Fine powder
2	Colour	Faint yellowish green
3	Odour	Herbaceous
4	Taste	Bitter
5	Moisture Content	4.0%
6	Toal Ash Value	8.71%
7	Acid Insoluble Ash	3.30%
8	Alcohol Soluble Extractive	12.96%
9	Water Soluble Extractive	25.03%
10	pH	4.9

Table no. 3 Observations and Result of Antimicrobial process of *Nimbadi Ubatan*: By Time Kill Test method:

Sample Identification	Test Organism	Exposure . Time	Count of Test Organism			Antimicrobial activity	
			Initial Count	After Exposure		Log	Percentage
				CFU/ml	Log	Reduction	Reduction
Nimbadi Ubtan	Staph. aureus	1 minute	1.80 x 10 ⁵ CFU/ ml L = 5.25	1.00 x 10 ³	3.00	2.25	99.44
	St. pyogenes		1.53 x 10 ⁵ CFU/ ml L = 5.18	< 10	<1	>4.18	>99.99
	Ps. aeruginosa		2.30 x 10 ⁵ QFU/ ml L = 5.36	2.40 x 10 ²	2.38	2.98	99.89
	Candida albicans		1.50 x 10 ⁵ CFU/ ml L = 5.17	7.40 x 10 ²	2.86	2.31	99.50

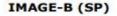
RESULT:

Test Product labeled as *Nimbadi Ubtan* has shown 2 – 4 log reduction/
>99% reduction of *Staphylococcus aureus*,

Streptococcus pyogenes, Pseudomonas aeruginosa, and Candida albicans in 1 minute when analyzed as per ASTM E 2315 - 16 Method.

1) RESULT OF ANTIMOCROBIAL ANALYSIS OF NIMBADI UBATAN

IMAGE-A (SA)





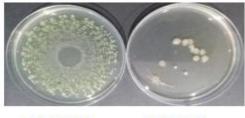




AT ZERO MIN.

AT ONE MIN.

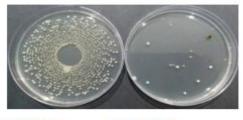
IMAGE-C (PS)



AT ZERO MIN.

AT ONE MIN.

IMAGE-D (CA)



AT ZERO MIN.

AT ONE MIN.

ALL FOUR IMAGES OF NIMBADI UBATAN HAS SHOWN 2-4 LOG REDUCTION/ >99% REDUCTION OF STAPHYLOCOCCUS AUREUS, STREPTOCOCCUS PYOGENES, PSEUDOMONAS AERUGINOSA, CANDIDA ALBICANS IN ONE MINUTE.

DISCUSSION:

Now a days society is more conscious about external beauty compare to internal beauty and also some professional fields they require good looking and charming beauty. Usually 10-15% cases present before general practitioners are pertaining to skin diseases. Due to altered life style, lack of physical exercise, unhygienic, mental stress, over eating, nutrition deficiency skin diseases are commonly observed. In modern medicine there is a treatment for skin disease, but there is no permanent solution. So to avoid recurrence of skin disease this topic had been chosen. The study entitled Analytical "Pharmaceutico-Study

Nimbadi ubatan and It's Antimicrobial activity". As per reference of Astang Hrudaya, Nimbadi ubatan was prepared and was analysed. According to classical text Nimbadi ubatan can be used in the form of lepa in skin disorder. Nimbadi ubatan was analysed for physico chemical parameters. It was in a fine powder form with faint yellowish green in colour, bitter in taste.

Analytical testing of *Nimbadi ubatan*:

Moisture Content:

This method is used to measure the amount of water content and other volatile material in a sample upon drying or heat treatment.

Moisture Content of *Nimbadi ubatan* was found 4.0% which is within the limit.

Total ash value:

The residue remaining of incineration is the ash content of the drug. It Measures the amount of carbon-free ash present in a prepared sample which represents the inorganic salts naturally occurring in drug or adhering to it or deliberately added to it as a form of adulteration.

Total ash value of *Nimbadi ubatan* was 8.71% which is within the limit.

Acid insoluble ash:

A high value of acid insoluble ash suggests the presence of sand, dust, dirt, stones, etc. that get mixed during processing or are present in the parent material as contamination. The higher value indicates inferior quality and low hygiene standards in the production process.

Acid insoluble ash of *Nimbadi* ubatan was 3.30% which is within the limit.

$\mathbf{p}^{\mathbf{H}}$.

The pH value of an aqueous solution may be defined as the logarithm of the reciprocal of the hydrogen ion concentration. Drug gets easily absorbable in acidic media and hence enters into the system and produces quick results.

The pH of *Nimbadi ubatan* was 4.9 (acidic).

Extractive values:

These are useful for the evaluation of a crude drug. Gives an idea about the nature of the chemical constituents present in the crude drug. Useful for the estimation of constituents extracted with the solvent used for extraction. Employed for material for which as yet no suitable chemical or biological assay exists.

Alcohol soluble extractive of *Nimbadi ubatan* was 12.96% and water soluble extractive of *Nimbadi ubatan* was 25.03%.

Thin Layer Chromatography:

TLC can be used to help determine the number of components in a mixture, the identity of compounds, and the purity of a compound.

TLC of *Nimbadi Ubatan* was done using stationary phase of Aluminum coated silica plates 60F254 and mobile phase of Toluene: Ethyl acetate: Formic acid in a ratio of (9.7: 0.3: 0.5) (v/v/v) with saturation time of 20 minutes.

After the whole process TLC plate was observed under different wavelengths and under different wavelengths different colour and different number of bands were noted.

Antimicrobial activity of *Nimbadi ubatan* by Time kill test method:

According to this method *Nimbadi* ubatan shown 2-4 log reduction/>99% reduction of S. *aureus*, S. *Pyogenes*, P. *aeruginosa*, and Candida albicans in 1 minute, So from above results we can say that *Nimbadi ubatan* shows good inhibitory action against

S.aureus, S.pyogenes, Pseudomonas aeruginosa, candida albicans.

CONCLUSION:

The study entitled present "Pharmaceutico-Analytical Study Nimbadi Ubatan and It's Antimicrobial activity" was aimed to prepare and perform all Analytical tests and Antimicrobial analysis of Nimbadi ubatan. Nimbadi ubatan for physico was analysed chemical parameters. It was in a fine powder form with faint yellowish green in colour, bitter in taste. It's Moisture Content was found 4%, Total ash value was 8.71%, acid insoluble ash was 3.30%. The pH was 4.9(acidic), alcohol soluble extractive was 12.96% and water soluble extractive was 25.03%. Nimbadi Ubatan shows good inhibitory action against S. aureus, S. pyogenes, Pseudomonas aeruginosa, candida albicans.

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Conflict of Interest: Non

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