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PREPARATION AND STANDARDIZATION OF NISHALOUHAM VATI- A RANDOMLY PRESCRIBED CLASSICAL YOGA

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Abstract:

Background: *Nishalouham vati is an effective ayurvedic classical formulation frequently prescribed for mainly jaundice, anaemia, and diabetes.* ***Aim:*** *In the present study, Nishalouham vati was formulated in laboratory by the formula given in an official ayurvedic treatise Bhaisajyaratnavali written by Acharya Govinda Das and the prepared vati or pill is subjected to standardize.* ***Materials and Method:*** *Formulation was prepared by maintaining the aseptic conditions. Following the testing protocol of ASU drugs, (Ayurveda, Siddha and Unani) various testing parameters including organoleptic and physicochemical tests for vati like: friability, disintegration, dissolution, TLC, HPTLC etc. had been performed to fix the quality standards of the drug. Among the critical analytical study atomic absorption spectroscopy (AAS) was attempted over the sample for the detection of the limit of arsenic, lead etc. heavy metals in the sample. Microbial assay was also performed.* ***Result:*** *This study results a set of standard data, essential for its standardization. The values obtained after physicochemical study showed that these values were all within the limit.* ***Conclusion:*** *The data maybe further helpful to develop a standard operating procedure and a quality standard limit for this specific formulation.*

Keywords: *Nishalouhamvati, Preparation, HPTLC, AAS, Microbial assay*.

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INTRODUCTION:

Ayurveda is considered by many scholars to be the oldest healing science. In Sanskrit, Ayurveda means "The Science of Life." Ayurvedic knowledge was originated in India more than 5,000 years ago and is often called the "Mother of All Healing." It stems from the ancient Vedic culture and was taught for many thousands of years in an oral tradition from accomplished masters to their disciples. Some of this knowledge was set to print a few thousand years ago, but much of it is inaccessible. The principles of many of the natural healing systems now familiar in the West had their roots in Ayurveda. [1]

Medicines prepared in the form of tablet or pills are known as vati. According to the Acharya Sarangahadhara, the information about Vati kalpana is mentioned like this. Gutika, vati, modaka, vatika, pindi, guda, varti (tablets, bolus, pills, are synonyms of vati kalpana (these are the names given for vati kalpana on the basis of shape, dose, root of administration e.g. vati etc.). It is prepared either by cooking the powder of drugs with jiggery, sugar, or guggulu or without cooking, by macerating the powder with any liquid like honey and guggulu then rolling into pills.[2,3]

Nishaloham vati is a frequently prescribed ayurvedic classical formulation mainly used in jaundice, anaemia, and diabetes. The main aim and objective of the present study is to prepare the Nishalouham vati by following the ayurvedic guidelines.

Haridra (*Curcuma longa*) [4] is an effective remedy for jaundice, among other ingredients Daruharidra [5] (*Berberis aristata*), Amlaki (*Emblica officinalis*)[6], Haritaki (*Terminalia chebula*)[7], Vibhitaki (*Terminalia bellerica*)[8], Katuki (*Picrorhiza kurroa*)[9] etc. having the potent hepatoprotective activity and those activities are also proved by recent researches. All the data claimed the hepatoprotective, anti-diabetic and anti-emetic condition of Nishalouham vati. Louha vashma is another important ingredient of this preparation which is added in maximum amount in comparison with the other ingredients. Honey and ghee were used as natural binder.

MATERIALS AND METHOD:

Collection of Raw Materials:

All the raw materials required for the preparation of Nishalouham vati were collected from the authentic suppliers and further tested in laboratory to ensure the optimum quality of the samples. The raw material of nishalouham vati is enlisted under table 1.

Table 1: raw materials required for the preparation

Nameof the Ingredients	Required Amount
Haridra (*Curcuma longa*)	50 gm
Daruharidra (*Berberisaristata*)	50 gm
Amlaki (*Emblicaofficinalis*)	50 gm
Haritaki (*Terminaliachebula*)	50 gm
Vibhitaki(*Terminaliabellerica*)	50 gm
Katuki (*Picrorhizakurroa*)	50 gm
Louhabhasma	300 gm

Preparation:[10]

The ingredients are shade dried and crushed drugs in mortar pestle or grinder

↓

The powdered drugs were passedthrough 80 meshes

↓

They were mixed thoroughly

↓

Then required amount of honey and ghee in unequal amount was mixed with it to form a soft paste like appearance

↓

Then pills of 250 mg and 500 mg was prepared from it

↓

Finally it was preserved in a tight closed container

Standardization of the Formulation:[11]

The prepared formulation was further studied for standardization. The basic parameters which were followed for testing the sample as below:

(a) Organoleptic Properties: 1) colour, 2) odour, 3) taste.

(b) Phytochemical Tests:1) Phytochemical screening (Qualitative tests for the presence of alkaloid, flavonoid, carbohydrate, steroid, terpenoid etc.), 2) Thin Layer Chromatographic (TLC) Technique.

(c) Instrumental analysis:1) P^H, 2) Hardness tester, 3) Friability test, 4) Weight variation, 5) Disintegration test,6)Dissolution test.

(d) Advanced Instrumental Analysis:1) Identification of the constituents by High Performance Thin Layer Chromatography (HPTLC) technique. 2) Determination of the heavy metals with the help of Atomic absorption spectroscopy (AAS).

Chromatographic Conditions:[12]Application mode: Camaglinomate V; Development chamber : Camag Twin through chamber; Plates: Precoated silica gel $^{60}F_{254;}$ Chamber saturation 30 min; Development time 30 min; Development distance: 7 cm; Scanner: Camag scanner II; Scanner: Deuterium lamp and mercury lamp; Photo documentation: Camagreproster; Data system: Win cats software; Drying device: Oven and was visualized under 254 nm and 366 nm; Sample Preparation:1g of sample was taken and dissolved in methanol: water= 1:1; Solvent system:Ethyl Acetate: Methanol = 3:7.

(e) Biological Analysis: 1) Total bacterial count (TBC), 2) Total fungal count (TFC), 3) Tests for specific pathogen: *E.coli, Salmonella sp, S. aureus, Pseudomonas aeruginosa.*[13]

RESULT AND DISCUSSION:

(a) Organoleptic Properties:

The organoleptic observations are summarized under table 2

Table 2: Organoleptic Observations

Parameter	Observation
Colour	Deep brown or blackish
Odour	Vegetative
Test	Bitter, sweet, astringent

(b)Phytochemical Tests:

The results of phytochemical screening is summarized under table 3

Table 3: Phytochemical screening

Parameter	Observation
Alkaloid	Present
Phenolic compound	Absent
Steroid	Present
Terpenoid	Absent
Glycoside	Present
Saponin	Absent
Carbohydrate	Present
Protein	Present
Tanin	Present
Fixed oil	Absent

Thin Layer Chromatography (TLC):

Two types of solvent systems were selected for the TLC study were: (a) Ethyl acetate: Methanol = 6:4(b) Chloroform: Ethyl acetate = 2:8

i)The R_f value of the sample was found to be 0.816, 0.766, 0.733, 0.55, 0.516 [In the solvent system(Ethyl acetate: Methanol = 6:4)]

ii) The R_f values of the sample were found to be 0.966, 0.186 [In the solvent system ($CHCl_3$:Ethyl acetate= 2:8)].

c) Instrumental Analysis:

The tests results are summarized under table 4.

Table 4: Instrumental Analysis

Test parameters	Results Value
pH Value	3.5-4
Hardness	6.5 kg
Loss on drying	84.733 % w/w
Disintegration time	30 minutes
Disslution test	Maximum range found to be 99.27% and minimum range 94.67 %
Friability test	0.11% (Pass the limit)
Weight variation	For 250 mg tablet ±7.25 % (Passed the limit)

c) Advanced Instrumental Analysis:

Higher Performance Thin Layer Chromatography (HPTLC):

The solvent system used for HPTLC was Ethyl acetate: Methanol = 6:4. The R_f values were found to be 0.20, 0.47, 0.76, 0.87 after derivatization with Anisaldehyde-H_2SO_4. The chromatographic pictures as observed under UV chamber at 254 nm, 366 nm, after derivatization with the indicator anisaldehyde-H_2SO_4 at naked eye and at 366 nm were depicted gradually under figure 1, figure 2, figure 3 and figure 4.

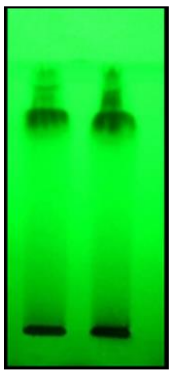

Fig. 1: At 254 nm

Fig. 2: At 366 nm

Fig. 3: After Derivatization

Fig. 4: After derivitization with anisaldehyde-H_2SO_4 at 366 nm

Table 5: Results of Total Microbial Count

	Media used	Report
Total aerobic count	Nutrient broth agar medium	Growth was not found
Total fungal count	Czapekdox agar medium	Growth was not found
Name of pathogen	**Media used**	**Report**
Escherichia. coli	EC broth agar medium	Growth was not found
Salmonella sp.	Salmonella differential agar medium	Growth was not found
Pseudomonas aeruginosa	Cetramide agar medium	Growth was not found
Staphylococcus aureus	Mannitol salt agar medium	Growth was not found

Table 6: Results of Tests for the presence of Specific Pathogen

Sample Name	Element	Wave length	Instrument Detection Limit (ppm)	Permissible Limit	Results
Nishalouham vati	Lead (Pb)	217.0	0.001	3 ppm	BDL
	Arsenic (As)	193.7	0.02	1 ppm	BDL

Atomic Absorption Spectroscopy (AAS):
AAS was performed to detect the presence of heavymetals in the sample. The result obtained was summarised under table 5.
ppm- Parts per million, BDL- Below detection limit

e) Microbial Assay:
Microbiological analysis revealed that the prepared sample had passed the limit of total bacterial count and total fungal count. The sample was also free from specific pathogens. The results were summarized under table 5 and table 6.

CONCLUSION:

Nishalouham vati can be concluded that, it is an effective formulation for jaundice, anaemia, diabetes etc. Haridra is an effective remedy for jaundice, among other ingredients Daruharidra, Amlaki, Haritaki, Vibhitaki, Katuki etc. having the potent hepatoprotective activity those activities were also proved by the recent researches[13-17]. All the data claimed that, the hepatoprotective, anti-diabetic and anti-emetic capability of Nishalouham vati. Louha vashma is another important ingredient of this preparation which was added in maximum amount in comparison with the other ingredients.
Honey and ghee were used as natural binder. In this formulation hence, no synthetic or chemical binder was used; however, it showed a good disintegration value (30 minute) with a permeable hardness (6.5 kg) which indicated good dispersible effect of this vati or pills. Phytochemical screenings revealed that alkaloid, steroid, glycoside, carbohydrate, protein, tannin were present, the P^H value was observed acidic that is 3.5-4. The permissible limit of lead was 10 ppm as per recommendation of Department of AYUSH and the vati were tested successfully to pass the result under permeable limit. Microbiological analysis revealed that the vati did not contain any specific pathogen or any bacteria or fungi. So, in a nutshell it can be summarized that the operation or process of Nishalouham vati can be followed as a standard operating procedure and the analytical data may be further useful to prepare a standard monograph of Nishalouham vati.

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