Impact of *Shodhana* (Purificatory Procedures) on *Kupeelu* (*Strychnos Nux- Vomica* Linn.) Seeds: A Pharmaceutico-Analytical Study
Swarnendu Mitra, V J Shukla and Rabinarayan Acharya

Evaluation of Biochemical effects of Homeopathic Preparation of *Chelidonium majus* in Potencies of 30 C and 200 C on Liver Functional Markers
Roopali Gajraj Redkar and Sadhana Sathaye

Status of Implementation of Indian Systems of Medicine and Homoeopathy (ISM & H) Policy, 2002 - A Secondary Data Analysis
Balpreet Singh, Amarjeet Singh and Manoj Kumar

Efficacy and safety of poly herbal formulation WH-1 FAS-3 kit and fluconazole in female patients with vaginosis / vaginitis
Kulkarni Chanda, Deb Jarita, Mishra Devesh, Dias Mary, Mhaskar Rita and A.S. Mohammed

Evaluation of ‘Bhavana Samskara’ with reference to Pippali Churna and Chausasta Pippali
P. B. Pimpalgaonkar Ashlesha Raut and R. S. Sawant

Antimicrobial activity of an Indian Mistletoe, The Hemiparasite *Dendrophthoe Falcata* L. F. (Loranthaceae)
S. H. Patil, V.S.Patil, R. B.Jadhav, G. S. Talele and S. J. Surana

Role of Gokshuradi Yoga in the management of Klaibya (Erectile Dysfunction)
Dhanaraj Nagar, Manjunatha T Sanoor, Baldev Kumar and Hetal Dave

Age Related Macular Degeneration: Therapeutic Targets and Possible Role of Ayurveda
Manoj Kumar, S.C. Varshney and O.P.S. Maurya

Conferences and Forthcoming Events
CONTENTS

Impact of Shodhana (Purificatory Procedures) on Kupeelu (Strychnos Nux-Vomica Linn.) Seeds: A Pharmaceutico-Analytical Study
Swarnendu Mitra, V J Shukla and Rabinarayan Acharya ... 65-71

Evaluation of Biochemical effects of Homeopathic Preparation of Chelidonium majus in Potencies of 30 C and 200 C on Liver Functional Markers
Roopali Gajraj Redkar and Sadhana Sathaye ... 73-80

Status of Implementation of Indian Systems of Medicine and Homoeopathy (ISM & H) Policy, 2002 - A Secondary Data Analysis
Balpreet Singh, Amarjeet Singh and Manoj Kumar ... 81-92

Efficacy and safety of poly herbal formulation WH-1 FAS-3 kit and fluconazole in female patients with vaginosis / vaginitis
Kulkarni Chanda, Deb Jarita, Mishra Devesh, Dias Mary, Mhaskar Rita and A.S. Mohammed ... 93-100

Evaluation of ‘Bhavana Sanskara’ with reference to Pippali Churna and Chausasta Pippali
Dr. P. Pimpalgaonkar Ashlesha Raut and R. S. Sawant ... 101-106

Antimicrobial activity of an Indian Mistletoe, The Hemiparasite Dendrophthoe Falcata L. F. (Loranthaceae)
S. H. Patil, V.S.Patil, R. B.Jadhav, G. S. Talele and S. J. Surana ... 107-111

Role of Gokshuradi Yoga in the management of Klaibya (Erectile Dysfunction).
Dhanaraj Nagar, Manjunatha T Sanoor, Baldev Kumar and Hetal Dave ... 113-120

Age Related Macular Degeneration: Therapeutic Targets and Possible Role of Ayurveda
Manoj Kumar, S.C. Varshney and O.P.S. Maurya ... 121-128

Conferences and Forthcoming Events ... 129
THE JOURNAL OF RESEARCH & EDUCATION IN INDIAN MEDICINE

Journal of Research and Education in Ayurveda, Yoga, Naturopathy, Unani, Siddha, Homeopathy, Complementary and Alternative Medicine, Integrative Medicine, Medicinal and Aromatic Plants, Pharmaceutical Sciences …… An International Quarterly

Call for Authors

The Editorial Board of The Journal of Research and Education in Indian Medicine (JREIM) (Website: www.jreim.com/aboutus.htm) is interested in receiving and reviewing manuscripts in all areas of Research and Education in Ayurveda, Yoga, Naturopathy, Unani, Siddha, Homeopathy, CAM, Integrative Medicine, Medicinal plants and Pharmaceutical Sciences in general and on education of AYUSH sector in particular.

Papers submitted for consideration for publication in The Journal of Research and Education in Indian Medicine (JREIM) will be reviewed by at least two expert members of the Editorial Review Board with appropriate credentials and expertise in the specific topic covered. On our web site click on the links under the “Information for Authors” section for complete guidelines for authors and submission requirements. Manuscripts can be electronically submitted by e-mail to editor@jreim.com or any Subject Editor

The Journal of Research and Education in Indian Medicine (JREIM) invites submission of Review articles and research papers in all areas of Research & Education in Ayurveda, Yoga, Naturopathy, Unani, Siddha, Homeopathy and other Traditional/Alternative/Oriental Systems of Medicine. The sole criterion for publishing is academic and scientific accuracy.

We invite anyone who is involved in research in any relevant area to submit his or her manuscript for consideration of publication. The Journal of Research and Education in Indian Medicine (JREIM) is committed to rapid review and publication. Your manuscript will be published, once accepted, within 90 days of acceptance.

An Invitation to Subscribe the Journal

I hope that after examination of our Journal and other JREIM publications, you will decide that some or all of these publication merit being recommended to your main or departmental library for current subscription and for acquisition of Special Issues of some of the back volumes.

If you have any questions, please submit them through our website: www.jreim.com/aboutus.htm or e-mail to editor@jreim.com

Prof. Suresh Kumar
Founder Editor

Prof. Em. R.H. Singh
Editor-in-Chief

* The manuscripts submitted in proper JREIM format/typesetting/style are only Registered for publication and assigned Manuscript Number and Reviewers
IMPACT OF SHODhana (PURIFICATORY PROCEDURES) ON KUPEelu (STRyCHNOs Nux-VOMICA LINN.) SEEDs: A PHARMACEUTICO-ANALYTICAL STUDY

SWARNENDu MITRA,1 V J SHUKLA2 AND RABINARAYAN ACHARYA3

Department of Dravyaguna,1,3 Pharmaceutical Laboratory,2 Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University, Jamnagar, Gujarat (India)

Abstract: Kupeelu (Strychnos nux-vomica Linn.), a drug mentioned under Upavisha (semi-poisonous) group of Ayurvedic pharmacopoeia, is being practiced widely in Ayurvedic therapeutics since long. Certain compound formulations containing Kupeelu are also well practiced in Homeopathy and Unani System of Medicine. Ayurveda strictly recommend the use of this drug in therapeutics only after proper Shodhana (purificatory procedure) through some specific media like Gomutra, Godugdha, Goghrita, Kanji etc. Though various Shodhana procedures are recommended in Ayurvedic classics for purification of Kupeelu seeds, but updated scientific researches regarding the Shodhana methods are lacking. Keeping this fact in mind, an attempt has been made in the present study to evaluate the impact of Shodhana on Kupeelu seeds while performing the specific Shodhana method, recommended by the Ayurvedic Formulary of India (A.F.I.). This study reveals that the toxic alkaloids Strychnine & Brucine, present in Kupeelu seed, were reduced by 71.49% and 54.02% respectively, in comparison to the raw seed, as determined by H.P.T.L.C. study.

Keywords: Kupeelu, Strychnos nux-vomica, Shodhana, Ayurvedic Formulary of India (A.F.I), Strychnine, Brucine.

Introduction

The uses of poison have been recorded in ancient Ayurvedic classic since long ago. Acharya Charaka has elaborately mentioned various poisoning, symptoms and their management (Charak). Acharya Sushruta has classified categorically the various sources of poison and described them accordingly (Sushruta). The poisonous plants reported in ancient scriptures of Ayurveda are being still practiced widely to combat number of diseases after passing through specific Shodhana (purificatory procedure). The concept of Shodhana was reported in the Charaka Samhita for first time, in the context of Danti Dravanti Kalpadhyaya. Here, to reduce the ‘Vikasi’ property of Danti (Baliospermum montanum) root, Charaka advocated a specific Samaskara by Agni (Charak). Acharya Vagbhata also advocated the Shodhana of Bhallataka fruit (Semicarpus anacardium Linn.) in detail in the context of Bhallataka Rasayana and Amrita Bhallataka (Vagbhata).

Kupeelu (Nux-vomica) has been described as a lethal poison and a cure for demonic possession in the KITAB AL-SUMMAM, an Arabian book of poison, which dates back to the 9th Century (Crozier A, Ashiara H).

Description of Kupeelu could not be traced in the ‘Brihat Trayee’ texts of Ayurveda (Shastry JLN). But this plant was described in different lexicons of Ayurveda by the name of Visatinduka, Kupeelu, Visamusti etc., which indicate toxic nature of this tree. Though the plant is described under the ‘Upavisa Vargas’ (semi-poisonous group) 13, its seeds have been used successfully in different Ayurvedic formulations after proper Samaskara known as Shodhana (Rasa Tarangini, Gogte VM). 16 alkaloids have been separated and identified from

1. Ph.D. Scholar   2. Head   3. Associate Professor
the raw nux-vomica seed and 80% of them are strychnine and brucine, and their derivatives such as isostrychnine and brucine N-oxide (Cai et al., 1990). The seeds also contain chlorogenic acid, a glycoside (loganin), and about 3% of fixed oil. Besides other minor alkaloidal constituents, the major chemical constituents of the seeds i.e. Strychnine (C21H22O2N2; m.p.286 to 288 °C) and brucine (C23H26O4N2; m.p.178 °C) have been reported for their adverse effects (Nadkarni KM). These alkaloids are found not only in the seed but also in the roots, bark, leaves, fruit - pulp, and the hard fruit-shells (Anonymous, 1998). A study reported that twenty-two identified alkaloids were isolated from the root bark and leaves of S. nux vomica (Baser Kemal HC et al., 1982).

The Shodhita (processed) Kupeelu seeds are mainly used as aphrodisiac, appetizer, anti-periodic, digestive, purgative, and stimulant. Further the seeds are also used in anemia, asthma, bronchitis, intermittent & malarial fever and in weakness of extremities (Sabnis M). Shodhita Kupeelu is also claimed to be a potent drug in countering old age problems and specially recommended during senility as Rasayana (antioxidant) (Pandey G). The plant is also found to have analgesic & anti-inflammatory (Yin W et al., 2003), anti-oxidant (Tripathi YB and Chaurasia S, 1996), anti-tumor (Deng XK et al., 2006), anti-snake venom (Chatterjee I et al., 2004), anti-diarrheal (Shoba F et al., 2001), and hepato-protective (Gopalkrishna SV et al., 2010), activities when studied in animal models.

Searching through various research journals, text books of Ayurveda and different search engines reveals that very few works have been reported on the Shodhana aspect of Kupeelu (Mitra S et al., 2012). A few folklore purificatory methods are followed traditionally in some parts of India but these methods are not accepted as the official methods (Mitra S et al., 2011). Though a specific Shodhana method of Kupeelu seed has been recommended by the Ayurvedic Formulary of India but the scientific research work regarding the impact of this Shodhana process is lacking till today. Hence, the present study has been planned to evaluate the impact of Shodhana on Kupeelu seeds by quantifying the toxic alkaloids through HPTLC technique.

Material and Methods

Present study was carried out by adopting the purificatory procedure recommended by A.F.I (Ayurvedic Formulary of India) where the three principles of Shodhaha i.e., Nimajjana (Dipping), Swedana (Boiling) and Bharajana (Frying) were followed consecutively.

Collection of drugs

Fully matured Kupeelu (Strychnos nuxvomica Linn.) fruits were collected from Bankura district, West Bengal, India during the month of December and were botanically authenticated by pharmacognosists and sample specimen were kept in the Institute’s museum for future reference (voucher no. 8009). Seeds were taken out from the fruit pulp, thoroughly washed in tap water, shade dried and then kept in air tight glass container for future study.

Collection of media

Cow urine (Gomutra) and cow milk (Godugdha) were collected from the local cowshed daily in the morning at 6 A.M. and cow ghee (Goghrita) was procured from the local market (Brand name: Gowrdhan; Mfd. By Parag Milk Foods Pvt.Ltd., Pune.) for Shodhana (proper processing/purification) of the seeds.

Equipment for Shodhana

Stainless steel vessel (20 cm × 30 cm); capacity of 7 L (used as Dolayantra), stainless steel rod (28 cm.), stainless steel vessel (48 cm × 30 cm × 7 cm); capacity of 3 L, cotton threads 30 cm in length, measuring mug (capacity of 1 L), muslin cloth (45 cm × 45 cm), digital weighing machine, pyrometer, digital induction cooker, stainless steel knife (blade: 15 cm × 2
Impact of Shodhana on Kupeelu seeds: A Pharmaceutico-analytical study

cm), frying pan (diameter: 20cm), stainless steel spatula (length: 30 cm), and measuring cylinder (10 ml, 25ml).

Procedure
100 g clean and dried raw Kupeelu seeds (KR) were taken in a stainless steel tray. One liter of cow urine was added to it and kept for 7 days. Every day at 7 A.M. the cow urine (1 lit.) was replaced by fresh one. On the eighth day, the seeds were taken out from Gomutra, washed with lukewarm water. The seeds were kept in a muslin cloth and were made into a Pottali. The Pottali was hanged in a stainless steel vessel and cow milk was filled in the vessel up to the complete immersion of the Pottali. It was then boiled on an induction cooker for three hours at 1000C throughout the experiment. After boiling for three hours, the seeds were taken out from Pottali and washed with lukewarm water. The seed coat and embryo were removed by a knife; the cotyledons were fried with cow ghee in mild temperature (temperature was set at 600C) on an induction heater until they became reddish yellow in color. Instantly the fried cotyledons were made into powdered form, dried under the shade, finally kept in an airtight glass container as ‘KGMDG powder’ for further use. The same procedures were repeated for three times to standardize the Shodhana process.

Equipment for HPTLC
A CAMAG (Switzerland) HPTLC system equipped with a sample applicator Linomat V sample applicator was used for application of samples. CAMAG TLC Scanner 3, Reprostar and Wincats 4.02 were used for scanning the plates. CAMAG twin through glass chamber was used for developing the plates.

Chemicals
Pure Strychnine and Brucine were obtained from Sigma Aldrich, U.S.A and precoated silica gel 60 F254 TLC aluminium plates (10×10 cms, 0.2mm thick), AR grade toluene, ethyl acetate, diethyl amine, methanol and chloroform were obtained from M/S Merck Ltd. Mumbai, India.

Preparation of standard strychnine and brucine solution
Strychnine standard (10 mg) and brucine standard (10 mg) were accurately weighed and dissolved in methanol in two standard flasks and final volumes were adjusted to 10 ml with methanol. (1 μg/μl)

Calibration curve for strychnine and brucine
The standard solutions corresponding to 2μg to 6μg of standard strychnine and brucine were applied on TLC plates (10cm× 10cm), precoated with silica gel as 6 mm bands by using CAMAG Linomat V sample applicator. The plate was developed in a solvent system of Toluene: Ethyl acetate: Diethyl amine (7: 2:1, v/v) in a CAMAG twin through chamber up to a distance of 7.5 cm at a temperature of 30 ± 20 C. The plates were air dried and scanned at a wavelength of 254 nm using CAMAG TLC scanner and Wincats 4.02 software. The peak area of strychnine and brucine were recorded for each concentration. The calibration curves of strychnine and brucine were obtained by plotting the graphs of peak areas vs. concentrations of strychnine and brucine.

Preparation of sample solutions for estimation of strychnine and brucine
Both the samples (KR & KGMDG) weighed 2g each, was defatted individually with petroleum ether. Defatted samples were then mixed with 10% ammonia and finally extracted with 25 ml methanol for 1 hr. under reflux. The methanol extracts were filtered and concentrated to 5 ml and used as test solutions. 5μl of each test solution was spotted along with 2 to 6 μl standard solutions of strychnine and brucine. The plates were developed in mobile phase of Toluene: Ethyl acetate: Diethyl amine (7:2:1, v/v) and scanned at 254 nm for strychnine and brucine. Peak areas were noted and quantity of
strychnine and brucine were calculated by comparing the areas of standard solutions from calibration curve.

**Results and Discussion**

The toxic principles present in the drugs are also considered as their active constituents. Therefore, it is not desirable to expel them out completely from the drugs. A study reports that the major toxic alkaloids i.e., strychnine & brucine present in the Kupeelu seed were reduced after Shodhana in Kanji and Adraka swarasa. This study exposes the fact that Shodhana with these two media i.e., Kanji and Adraka swarasa reduce the strychnine content by 39.25% and 67.82% respectively and brucine content by 17.60% and 40.06% respectively in comparison to the raw Kupeelu seeds. (Mitra S et al., 2012).

The main aim of Shodhana process is to reduce the toxic constituents to some extent or by potentiating their chemical transformation to non-toxic or relatively less toxic substances. There may be some new principles added to the drugs which are responsible for enhancing their biological efficacy. Hence, maximum beneficial effect can be obtained by administering the Shodhita drugs within their therapeutic dosage limit. Previous study revealed that crude Vatsanabha which is having cardiac depressant activity, converted into a cardiac stimulant drug after Shodhana in cow urine. (Singh LB).

In this study, Shodhana of Kupeelu seed was carried out by the A.F.I recommended method. Each Shodhana procedure was repeated for three times to establish the validation of the pharmaceutical processing. Shodhana of Kupeelu was performed by the subsequent processing through Nimajjana (dipping) in Gomutra, Swedana (boiling) in Godugdha, Bharjana (frying) in Goghrita for a specific time period.

Principles of Nimajjana & Swedana methods are similar to the common stages of extraction process where the solvent enters through the pores into the cells resulting in the swelling of the tissues and solution of the soluble constituents takes place within the cells followed by escape of dissolved material through the solvent boundary layer by the process of diffusion – finally separation of the solution from the drug occurs. The rate of extraction depends mainly on the temperature gradient and concentration gradient across the cell membrane. The rate of extraction and solubility is increased by elevation of temperature. Rising temperature increases the concentration gradient across the cell membrane thereby increasing mass transfer of active principles from solid material to the solvent (Carter SJ, Cooper and Gunn’s).

In Bharajana method, the seeds were fried with cow’s ghee in mild temperature for a specific period of time. It is reported that during Bharjana process, some physical & chemical changes like reduction in hardness, increase brittleness, formation of new chemical compounds may take place which ultimately make the drug body friendly. (Sarkar PK, 2008)

During Shodhana of Kupeelu in three media, change in color of those media was noticed and it might be due to the removal of color containing materials from the endosperm of the seeds. Taste of every media became bitter after Shodhana due to the extraction of bitter principles like Strychnine, Brucine, etc.

Table 1. Organoleptic characters of raw & purified Kupeelu seeds powder.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Organoleptic characters of raw Kupeelu seeds powder</th>
<th>Organoleptic characters of Kupeelu seeds powder purified by A.F.I. approved method (KGMDG) in three batches</th>
</tr>
</thead>
<tbody>
<tr>
<td>Texture</td>
<td>Smooth</td>
<td>KGMDG-1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>KGMDG-2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>KGMDG-3</td>
</tr>
<tr>
<td>Colour</td>
<td>Greyish white</td>
<td>Smooth</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Smooth</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Smooth</td>
</tr>
<tr>
<td>Odour</td>
<td>Slightly acidic</td>
<td>Reddish brown</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Reddish brown</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Reddish brown</td>
</tr>
<tr>
<td>Taste</td>
<td>Intense bitter</td>
<td>Pungent</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pungent</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pungent</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Bitter</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Bitter</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Bitter</td>
</tr>
</tbody>
</table>
Impact of Shodhana on Kupeelu seeds: A Pharmaceutico-analytical study

Changes in organoleptic characters of Kupeelu seeds were also noticed and the final quantity obtained after the Shodhana procedure was noted accordingly.

The data generated from the study reveals the effect of Shodhana with cow urine, milk & ghee successively on organoleptic characters and yield of final products of Shodhita Kupeelu.

Table 2. Effect of Shodhana on yield of final product (KGMDG powder).

<table>
<thead>
<tr>
<th>Batch</th>
<th>Weight (g) of the seeds</th>
<th>Initial</th>
<th>After frying in Goghrita</th>
<th>After drying</th>
</tr>
</thead>
<tbody>
<tr>
<td>KGMDG-1</td>
<td>100</td>
<td>77.70</td>
<td>63.20</td>
<td></td>
</tr>
<tr>
<td>KGMDG-2</td>
<td>100</td>
<td>74.80</td>
<td>61.80</td>
<td></td>
</tr>
<tr>
<td>KGMDG-3</td>
<td>100</td>
<td>78.10</td>
<td>66.70</td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td>100</td>
<td>76.86</td>
<td>63.90</td>
<td></td>
</tr>
</tbody>
</table>

Table 3. Physicochemical parameters of raw and purified seeds.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>KR</th>
<th>KGMDG</th>
</tr>
</thead>
<tbody>
<tr>
<td>Loss on drying 110°C (% w/w)</td>
<td>3.39</td>
<td>4.13</td>
</tr>
<tr>
<td>Ash Value (% w/w)</td>
<td>1.11</td>
<td>1.01</td>
</tr>
<tr>
<td>Water soluble extractive (% w/w)</td>
<td>37.83</td>
<td>26.93</td>
</tr>
<tr>
<td>Methanol soluble extractive (% w/w)</td>
<td>3.89</td>
<td>1.03</td>
</tr>
<tr>
<td>pH</td>
<td>4.75</td>
<td>5.71</td>
</tr>
</tbody>
</table>

Table 4. Qualitative tests for various functional groups.

<table>
<thead>
<tr>
<th>Functional groups</th>
<th>KR</th>
<th>KGMDG</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carbohydrate</td>
<td>+ ve</td>
<td>+ ve</td>
</tr>
<tr>
<td>Protein</td>
<td>+ ve</td>
<td>+ ve</td>
</tr>
<tr>
<td>Fixed Oil</td>
<td>+ ve</td>
<td>+ ve</td>
</tr>
<tr>
<td>Tanin</td>
<td>+ ve</td>
<td>+ ve</td>
</tr>
<tr>
<td>Steroid</td>
<td>- ve</td>
<td>- ve</td>
</tr>
<tr>
<td>Alkaloid</td>
<td>+ ve</td>
<td>+ ve</td>
</tr>
<tr>
<td>Glycoside</td>
<td>+ ve</td>
<td>- ve</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>- ve</td>
<td>- ve</td>
</tr>
</tbody>
</table>

Table 5. Results of estimation of Strychnine and Brucine in raw and purified samples of Kupeelu by HPTLC.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Amount of Strychnine found (% w/w)</th>
<th>Amount of Brucine found (% w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw Kupeelu (KR)</td>
<td>1.442</td>
<td>0.659</td>
</tr>
<tr>
<td>Kupeelu purified by A.F.I. approved method (KGMDG)</td>
<td>0.411</td>
<td>0.303</td>
</tr>
</tbody>
</table>
The intensely bitter raw Kupeelu became sweetish bitter after processing and grayish powder of raw seeds turned into grayish white in color after processing. (Table 1). It was found that 63.90% of purified Kupeelu seed powder was obtained as final product (KGMDG) after Shodhana. (Table 2).

Differences in all the physico-chemical parameters were observed among the raw and Shodhita Kupeelu samples. (Table 3.) Qualitative tests revealed only the absence of glycoside in Shodhita Kupeelu sample however, other functional groups remained same. (Table 4).

The Rf values of Strychnine and Brucine standard were found as 0.54 & 0.34 respectively in HPTLC chromatogram under UV spectrum at 254 nm (Figure 1 & Figure 2) and the peak areas of Strychnine (Figure 3) and Brucine (Figure 4) in both the samples were exposed. Calibration curves of Strychnine and Brucine were prepared by plotting concentrations of Strychnine and Brucine in the range of 2-6 μg/spot versus average area of the peak. The responses for concentrations of standard Strychnine and Brucine were found to be linear (Figure 5 & Figure 6). The amounts of Strychnine and Brucine in raw & purified samples were computed from the calibration curves which suggest the reduction of Strychnine and Brucine content by 71.49% and 54.02% respectively in the Shodhita sample (Table 5) It might be due to the collective impact of the three media in the process of Shodhana. During Shodhana with Gomutra and Godugdha, some amount of strychnine & brucine were removed from the seed by the process of extraction. It is also reported that boiling in milk converted the strychnine into less toxic isostrychnine (Cai et al., 1990). Finally frying with Goghrita might have been converted strychnine & brucine into less toxic derivatives like isostrychnine, isobrucine, strychnine N- oxide, brucine N-oxide etc.

**Conclusion**

Shodhana of Kupeelu seeds with successive three media i.e., Gomutra, Godugdha
and Goghrita (as approved by A.F.I.) successfully reduce the toxic alkaloids from the seeds. These findings strongly confirm the claims of ancient classics of Ayurveda that Shodhana process reduces the toxic effects of the drugs.

Acknowledgement
The authors are very much thankful to Director of I.P.G.T & R.A, G.A.U, Jamnagar, Gujarat, India for providing all necessary facilities for carrying out this research work.

References

25. Mitra S, Shukla VJ and Acharya RN: Effect of Shodhana (processing) on Kupeelu (Strychnos nuxvomica Linn.) with special reference to strychnine and brucine content. Article accepted and ahead of print for publication in AYU 2012

Address for correspondence: Dr. Rabiranaryan Acharya, Associate Professor, Dept. of Dravyaguna, Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University, Jamnagar, Gujarat (India). E-mail: drrnacharya@gmail.com

532_2012
EVALUATION OF BIOCHEMICAL EFFECTS OF HOMEOPATHIC PREPARATION OF CHELIDONIUM MAJUS IN POTENCIES OF 30 C AND 200 C ON LIVER FUNCTIONAL MARKERS

ROOPALI GAJRAJ REDKAR* AND SADHANA SATHAYE1

Department of Pharmaceutical Sciences and Technology (Now UGC-CAS), 1 Institute of Chemical Technology (ICT), Nathalal Parekh Marg, Matunga, Mumbai – 400019 Maharashtra (India)

Abstract: Homeopathy is a complete medical system based on principles of “similaris” and “dilutions” that have an effect on the potentization of the drug activity. However, for a number of people Homeopathy is a controversial area of Complementary and Alternative Medicine (CAM) that provided a scientific rationale for testing to reevaluate its principle and theory by proving its effects on healthy experimental animals. The hepatoprotective preparation of Chelidonium majus in 30 C and 200 C potencies were subjected to antioxidant assay. Their effects on behavioral pattern and serum biochemical variables including protein content, glutamic oxaloacitic transaminase (SGOT), glutamic pyruvic transaminase (SGPT), total and direct bilirubin in healthy Wistar rats were evaluated in a 28 days single-dose study period. The results revealed moderate antioxidant activity and hepatotoxic effects indicated by significant increase in SGOT and SGPT levels in the first 14 days were exhibited predominantly by 200 C than 30 C is in view of the similie principle.

Keywords: Law of similars, Law of dilutions, Chelidonium majus, Potencies, Biochemical variables, Homeopathy.

Introduction

Homeopathy, by a certain treatment of the crude medicinal substances, advances them into the state of progressive and high development of their indwelling forces, in order that it may then use them in curing in the perfect manner (Hahnemann, 2006). The principle of similars (or “like cures like”) is a central homeopathic principle, which states that a disease can be cured by a substance that produces similar symptoms in healthy people (Stehlin, 1996). In homeopathy as per the principle of dilutions (or “law of minimum dose”), a solution that is more dilute is described as having a higher potency, and more dilute substances are considered by homeopaths to be stronger and deeper-acting “remedies” (Smith, 1989). It is based on the belief that a substance in large doses will produce symptoms of a specific disease and in extremely small doses, cures it; the concept, often referred to as the Law of Infinitesimals. Homeopathic products are formulated by taking a sample of a substance and repeatedly diluting it with water, water/alcohol, or milk sugar with succession, the dilutions of which are designated as X and C. At first, Hahnemann used material doses for proving, but he later advocated proving with remedies at a 30 C dilution, and most modern proving are carried out using ultradilute remedies in which it is highly unlikely that any of the original molecules remain (Vallance, 1998; Dantas et al., 2007). The classical experimental approach to laboratory animal experimentations, typical of modern medicine and the current international scientific literature, can help us to understand only some of the aspects of homeopathy.

Alcoholic hepatitis is a form of acute injury to liver tissue that is also a precursor of cirrhosis, and carries significant morbidity and mortality (Cassedy, 1999). The homeopathic preparation of Chelidonium majus, is recommended in various hepatic disorders including alcohol...
Redkar and Sathaye

induced hepatitis. Experimental studies have been carried out in animals for its hepatoprotective effects in alcohol-induced hepatitis (Mehta et al., 2006a; Mehta et al., 2006b; Sathaye et al., 2007). Despite its clinical uses, there is a lack of data on the effects of this miniature homeopathic dosage form on normal animals as proven for its hepatoactive potential. In particular, the potential role of oxidative stress has become clearer, although immune mechanisms and apoptosis remain uncertain. Despite its medicinal uses, there is a lack of sufficient data on toxicity and preliminary biochemical data (Madhotra and Gilmore, 2003). In the last 2 years, almost 10 cases of acute hepatitis induced by herbal preparations of greater celandine (Chelidonium majus) which are frequently prescribed to treat gastric and biliary disorders were observed (Benninger et al., 1999; Moro et al., 2009; Crijns et al., 2002).

The present study was carried out to investigate the effects of oral administration of the homeopathic miniature dosage forms of C. majus in potencies of 30 C and 200 C in normal healthy Wistar rats on blood serum biochemical variables including the total protein, the liver functional enzymes including serum glutamate oxalotransaminase SGOT (AST), serum glutamate pyruvate transaminase SGPT (ALT) and bilirubin as hepatocellular and hepatobiliary markers respectively. Effects of drug dilutions on average body weights, observational behavioral, motor functional tests and sensorimotor responses also served as an additive determinant that were conducted during the first 14 and also for the next 14 days of the experimental period. Estimation of body weights served as a key parameter to follow any progress in liver damage. Since generation of free radicals is an important mediator in the etiology and pathogenesis of liver disorders, antioxidant assays were intended to be performed by the most widely used 1,1-diphenyl-2-picryl hydrazyl radical (DPPH) method. This approach may help to test under controlled conditions the principle of homeopathy “Similia similibus Curentar” of drug action and the effects of dilution and dynamization (potentiation) on the drug activity. The objective of this study was therefore to evaluate whether the marketed homeopathic preparation could exhibit any damage on hepatocytes and/or on the biliary system in the normal experimental animals in terms of their effects on liver functional markers and any changes in observational behavioral pattern under the suggestion of a consulting homeopath.

Material and Methods

Chelidonium majus is one of the herbs in a polyherbal homeopathic formulation available in different potencies including 30 C and 200 C. It is used in liver disorders and affects the liver function tests biochemically.

Evaluation of antioxidant activity using 1,1-diphenyl-2-picryl hydrazyl (DPPH) assay

The antioxidant activity in terms of % inhibition of 200 μM DPPH by Chelidonium at potencies of 30 C and 200 C was calculated at 517 nm and evaluated using DPPH assay (Govindarajan et al., 2003).

\[
\text{Percentage DPPH scavenging activity} = \frac{A_{(\text{DPPH})} - A_{(\text{DPPH} + \text{sample})}}{A_{(\text{DPPH})}} \times 100
\]

where A= absorbance

Chemicals and Drugs

C. majus in 30 C and 200 C potencies (liquid dilutions) obtained from SBL Pvt. Ltd., New Delhi were further diluted as per the suggestions of the Homeopath.

5 ml of liquid dilution was diluted to 100 ml with distilled water and 1 ml administered orally to the Group II and Group III animals. All the chemicals used in biochemical estimation were of analytical grade (Ecoline Merck).

Study design

The preparation was suitably diluted and administered to the animals for the total period of 28 days (4 weeks), i.e. each study period comprised of 14 days.
Adult albino Wistar female rats weighing 150-200 g were procured from Bharat Serum, Thane (India). They were maintained under standard laboratory conditions at the ambient temperature of 24 +/- 2°C with the 12 h light/12 h dark cycles. The study was conducted as per the guidelines provided by the Committee for the purpose of Control and Supervision of Experimentation on Animals (CPCSEA). The animals were then acclimatized to Institute of Chemical Technology (ICT) animal house for about a week and fed with the standard diet pellets. All animal experiments were conducted after the approval of Institutional Animal Ethics Committee (IAEC) at ICT (IAEC Approval No: ICT /PH/IAEC/1204/20).

**Evaluation of hepatoactive potential of 30 C and 200 C**

The animals were divided into 3 major groups comprising of 6 animals (Wistar rats) each:-

- **Group I**: Vehicle control. Distilled water *ad libitum*
- **Group II**: 30 C liquid dilution for 28 days
- **Group III**: 200 C liquid dilution for 28 days

The observational behavioral pattern and motor functional battery of tests were evaluated in the experimental animals during the entire 28 days study period.

**Locomotor activity**

This was evaluated in terms of the speed and vigor of movement. i.e. exaggerated scratching or self-biting (S), restlessness (R) and writhing (W). Any bizarre or stereotyped behavior i.e. head flicking, circling or retropulsion where the animal walks backward, appearance of arousal (excited) or stupor (state of unconsciousness); alertness or excitement were recorded. Any vocalizations or urination-defecation exhibited during handling were also observed.

**Sensorno-motor responses**

Visual placing where the animal was lifted vertically by mid-tail above the wire-mesh grid and lowered to elicit visual placing response characterized by extension of both forelimbs and hind limbs.

**Neurologic**

A. **Posture** - Evaluated during forward movement of the animal. It reflects both behavioral and neurological state of animal, since tail elevation are usually increased by excitation or rigidity and decreased by stupor or flaccidity.

B. **Muscle tone- grip strength** – Here, the animal was allowed to stand on a wire-mesh grid and a horizontal pull is applied to the tail to slowly draw the animal backwards.

C. **Equilibrium and gait righting reflex** – Any neurological impairment in terms of inability of the animal to land squarely on all 4s when somersaulted into the air or placed on its back for initial testing was observed.

**Autonomic**

Secretions and excretions- salivation and lacrimation, diarrhea (any liquid stool) and miscellaneous – piloerection, any occurrence of cyanosis and hypothermia as determined by palpation, only its occurrence was noted.

**Ophthalmological examination**

The ocular examination in terms of exophthalmos when the animal remains undisturbed was performed on all animals before the study began, on control and high-dose animals at the end of the study.

The biochemical variables that serve as liver functional markers were evaluated for the hepatoactive activity on 0, 14th and 28th day are as follows:-

i. SGOT and SGPT done on the same day of bleeding of the animals and collection of serum.

ii. Total protein by Biuret method colorimetrically.

iii. Total bilirubin.

iv. Direct Bilirubin.
**Statistical analysis**

Results are expressed as mean ± SEM. The levels of significance * and ** (p<0.05 and 0.01 respectively) are measured by ANOVA followed by Dunnet’s Multiple method (of comparison) with Day 0 as control for comparison.

**Results**

The *in vitro* antioxidant activity of *C. majus* in potencies of 30 C and 200 C in the volume of 0.2 ml exhibited 14.7 % and 3.5 % DPPH inhibitory activity respectively whereas 0.8 ml of 30 C and 200 C exhibited 24.7 % and 7.9 % DPPH radical scavenging activity (Table 1).

On treatment of animals with the diluted preparation of *Chelidonium*, not much significant changes in observational behavioral pattern and locomotor activity were observed in the treated groups except watery discharge of stools.

The findings on average body weights, the biochemical variables including total protein content, SGOT, SGPT and bilirubin levels in vehicle control and diluted *C. majus* treated animals (30 C and 200 C potencies) on [0 and 14 days] and [0 and 28 days] are given separately (Tables 2 and 3).

**Table 1.** The % DPPH radical scavenging activity of 30 C and 200 C homeopathic *Chelidonium* preparations.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Volume In ml</th>
<th>% DPPH radical scavenging activity of 30 C</th>
<th>% DPPH radical scavenging activity of 200 C</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.2</td>
<td>14.7</td>
<td>03.5</td>
</tr>
<tr>
<td>2</td>
<td>0.4</td>
<td>21.0</td>
<td>05.6</td>
</tr>
<tr>
<td>3</td>
<td>0.8</td>
<td>24.7</td>
<td>07.9</td>
</tr>
<tr>
<td>4</td>
<td>1.6</td>
<td>-</td>
<td>19.66</td>
</tr>
<tr>
<td>5</td>
<td>2.0</td>
<td>-</td>
<td>20.18</td>
</tr>
</tbody>
</table>

†Results are expressed as mean ± SEM. All data values are in triplicate (n=3)

In control as well as in treated group, increase in body weight in first 14 days and insignificant decrease in subsequent 14 days were noted indicating growth phase. Both 30 C as well as 200 C did not produce any significant rise in body weight.

In the case of protein content, slight increase in first 14 days followed by slightly more increase in the next 14 days were observed in the control group. Whereas, in both 30 C group and in 200 C, decrease in protein content in first 14 days and compensatory increase in the next 14 days were observed.

**Table 2.** A 14-day study period on effects of 30 C and 200 C on biochemical parameters against the vehicle control group.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Biochemical variables</th>
<th>Days</th>
<th>Control</th>
<th>30 C</th>
<th>200 C</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Body weights g</td>
<td>00</td>
<td>171 ± 2.449</td>
<td>184 ± 4</td>
<td>180 ± 4.47</td>
</tr>
<tr>
<td></td>
<td></td>
<td>14</td>
<td>197.4 ± 4.556**</td>
<td>193.6 ± 4.354</td>
<td>199.8 ± 5.24*</td>
</tr>
<tr>
<td>2</td>
<td>Total protein content g/dl</td>
<td>00</td>
<td>7.5391 ± 0.0191</td>
<td>7.62 ± 0.09625</td>
<td>7.62 ± 0.09625</td>
</tr>
<tr>
<td></td>
<td></td>
<td>14</td>
<td>7.57 ± 0.02757</td>
<td>4.825 ± 0.857*</td>
<td>5.199 ± 0.7294</td>
</tr>
<tr>
<td>3</td>
<td>Total bilirubin mg/dl</td>
<td>00</td>
<td>0.206 ± 0.02694</td>
<td>0.1982 ± 0.0327</td>
<td>0.198 ± 0.0314</td>
</tr>
<tr>
<td></td>
<td></td>
<td>14</td>
<td>0.2232 ± 0.0321</td>
<td>0.2034 ± 0.041</td>
<td>0.302 ± 0.084</td>
</tr>
<tr>
<td>4</td>
<td>Direct bilirubin mg/dl</td>
<td>00</td>
<td>0.155 ± 0.033</td>
<td>0.0592 ± 0.0066</td>
<td>0.1548 ± 0.034</td>
</tr>
<tr>
<td></td>
<td></td>
<td>14</td>
<td>0.1616 ± 0.037</td>
<td>0.080 ± 0.011</td>
<td>0.282 ± 0.092</td>
</tr>
<tr>
<td>5</td>
<td>SGOT (AST) U/l</td>
<td>00</td>
<td>47.6 ± 4.64</td>
<td>48.9 ± 4.085</td>
<td>48.9 ± 2.759</td>
</tr>
<tr>
<td></td>
<td></td>
<td>14</td>
<td>50.206 ± 4.623</td>
<td>56.56 ± 0.447</td>
<td>57.36 ± 0.22*</td>
</tr>
<tr>
<td>6</td>
<td>SGPT (ALT) U/l</td>
<td>00</td>
<td>63.5 ± 7.173</td>
<td>64.03 ± 7.17</td>
<td>66.42 ± 6.402</td>
</tr>
<tr>
<td></td>
<td></td>
<td>14</td>
<td>64.73 ± 7.45</td>
<td>107.5 ± 6.133**</td>
<td>137.09 ± 8.79**</td>
</tr>
</tbody>
</table>

†Results are expressed as mean ± SEM. The level of significances are indicated by * at P< 0.05; ** at P< 0.01 and *** at P< 0.001 with vehicle control as the group of comparison.
Both 30 C and 200 C did not produce any significant rise in bilirubin although there was significant increase in control but within the normal range. The liver enzymes namely SGOT exhibited significant rise (57.36 ± 0.22 U/l, p<0.05) after 200 C administration on 14th day but non-significant increase in 30 C group. The SGPT levels in the control group showed marginal increase in first 14 days and marginal decrease in the next 14 days. However, significant increases of SGPT in both 30 C (107.5± 6.133 U/l, P<0.01) and in 200 C groups (137.09 ± 8.79 U/l, P< 0.001) were observed in first 14 days with 200 C showing comparatively higher SGPT values. It was found that SGPT level reverted back to normal after 28 days of drug administration.

In summary, in a total 28-days study period, the rats in 200 C showed significantly high SGOT and SGPT levels in the first 14 days indicating hepatotoxic effects in comparison to 30 C.

**Discussion**

The lack of convincing substantiation supporting efficacy (Maddox et al., 1988) of homeopathy and its use of “remedies” without active ingredients have led to characterizations as pseudoscience and quackery (Ndububa, 2007 and Wahlberg, 2007) or in the words of a 1998 medical review, “placebo therapy at best and quackery at worst” (Atwood, 2003; Ernst, 2007).

The proposed rationale for these extreme dilutions – that the water contains the “memory” or “vibration” from the diluted ingredient – is counter to the laws of chemistry and physics, such as the law of mass action (Teixeira, 2007). Homeopathic “remedies” are safe at high dilutions recommended by Hahnemann, since they likely contain no molecules of the original substance, at the same time they may not be safe at lower dilutions (Chakraborti et al., 2003).

Practitioners of homeopathy contend that higher dilutions produce stronger medicinal effects. This idea is inconsistent with the observed dose-response relationships of conventional drugs (Shang et al., 2005), where the effects are dependent on the concentration of the active ingredient in the body (Levy, 1986) as reflected by the decrease in the antioxidant activity of Chelidonium preparations with increasing dilutions.
In the hepatoactive study of the effects of the homeopathic formulations in the healthy experimental animals, the most commonly used blood serum biochemical variables that were evaluated included SGOT (AST), SGPT (ALT) and bilirubin levels for hepatocellular and hepatobiliary damage respectively (Tennant, 1999; Ramaiah, 2007). SGOT is an enzyme found primarily in the cells of the liver, heart, skeletal muscles, kidneys, pancreas and to a lesser extent, in red blood cells. Its serum concentration is in proportion to the amount of cellular leakage or damage (Kesari et al., 2007). The significance of SGPT, an enzyme found primarily in the liver, is far greater. Its enhanced release into the bloodstream is the result of liver abnormality. It therefore serves as a fairly specific indicator of liver status and its elevated levels in serum indicate liver damage. The rise in serum levels of ALT and AST has been attributed to the damage in the structural integrity of the liver, since these enzymes are located cytoplasmically and released into the blood after cell damage (Janbas and Gilani, 2000; Venkatesh et al., 2011).

A decrease in body weight in treated groups indicates a decrease in food consumption, many molecular mechanisms are involved in the liver toxicity including changes in lipid and carbohydrate synthesis, metabolism and transportation out of the liver (Monteiro et al., 2009). The hepatotoxic effects by single-dose administration of Chelidonium preparations in healthy animals are well indicated by significant increase in SGOT levels and still significantly high SGPT levels as in 14-days by 200 C, which retained its normalcy at the end of 28-days study period in accordance to the homeopathic law of similars.

Regarding the interpretation of data in view of the simile principle, we observe that there are different levels of similarity and that the experimental data give support to this principle, but have not yet yielded the ultimate answer to the action mechanism of homeopathy.

Conclusion

The collective weight of scientific evidence has found homeopathy (Ernst, 2002), to be no more effective than a placebo (Altunc et al., 2007; Levy, 1986; Barrett, 2007). Depending on the dilution, homeopathic “remedies” may not contain any pharmacologically active molecules (Ernst, 1998) and for such “remedies” to have pharmacological effect would violate fundamental principles of science (Ernst, 2005).

Evidence of the biological activity of highly diluted-dynamized solutions is slowly accumulating, with some conflicting reports. Others point to observational and anecdotal evidence that homeopathy does work and argue that it should not be rejected just because science has not been able to explain it.

In conclusion, the importance of these laboratory studies lies in the fact that they have made it possible to obtain some preliminary evidence of the effects of high dilutions and dynamisations as in Chelidonium preparations under conditions that exclude any possible effect of suggestion.

Acknowledgements

We are grateful to Dr.S.S.Apte, Director, Association for Research in Homeopathy (ARH), Mumbai (India) for his valuable contributions in the practice of homeopathy.

The study is self-supported and does not involve any sponsor’s assistance ship.

There are no conflicts of interest amongst any of the researchers conducting these animal studies. No financial assistance was funded by any sponsoring agency.

References

2. Atwood, KC: ‘Neurocranial Restructuring’ and Homeopathy, Neither Complementary nor
Evaluation of Homeopathic Chelidonium majus on Liver Biomarkers in Wistar Rats

Alternative (Letters to the Editor). Arch Otalaryngol Head Neck 2003; 9(12):1356-1357


__Address for correspondance:__ Dr. Sadhana Sathaye, Associate Professor in Pharmacy, Pharmacology Research Laboratory-II, Department of Pharmaceutical Sciences and Technology (Now UGC-CAS), ICT, Nathalal Parekh Marg, Matunga, Mumbai – 400019 Maharashtra (India). E-mail: sadhanasathaye@gmail.com
STATUS OF IMPLEMENTATION OF INDIAN SYSTEMS OF MEDICINE AND HOMOEOPATHY (ISM & H) POLICY, 2002- 
A SECONDARY DATA ANALYSIS

BALPREET SINGH,1* AMARJEET SINGH2 AND MANOJ KUMAR3

Centre for Public health, 1,3
Institute of Emerging Areas in Science and Technology, Panjab University, Chandigarh, UT (India)
Department of Community Medicine, 2
Post Graduate Institute of Medical Education and Research (PGIMER), Chandigarh, UT (India)

Abstract: Context: India has designed ISM&H Policy in 2002 to emphasise the development of AYUSH due to public patronage of these systems. It is important to evaluate the implementation status of the policy in the period of eight years of its course. Aims: To ascertain the degree to which ISM&H Policy has been implemented. Design: A record based secondary data analysis was conducted during November 2010 to April 2011. Methods and Material: An implementation assessment tool containing 85 items was designed which had various domains described in policy. Various documents issued by MOHFW, Department of AYUSH, CCIM, CCH, CCRAS, CCRH, CCRUM, CCRYN, NMPB, CDSCO, Ministry of Commerce and Industry, Planning Commission etc. were analyzed. Websites of concerned agencies were also scrutinized. Score of ‘one’ was assigned to strategy/provision which had been implemented. The total score per component of policy and overall scores was calculated into the percentage and graded as Excellent, Good, Fair and Poor. The data was analyzed with the help of Microsoft Excel 2007. Results: Overall implementation score of ISM&H Policy was found to be 76.5%. Implementation status was found to be excellent, good, fair and poor for ten, two, four and one components of ISM&H Policy respectively. Conclusions: Implantation status of ISM&H policy was excellent. At same time, implementation status of some provisions of policy was found to be rudimentary. Government should support ISM&H by a significant increase in budget.

Keywords: ISM & H Policy, AYUSH, Implementation, Evaluation, CCIM, CCH, CCRAS, CCRH, CCRUM, NMPB, CCRYN.

Introduction

India has fairly well-organized system of medicine, known as Ayurveda, which is millennia old. Other traditional systems which developed a little later (but which are equally ancient) include Siddha, Amichi and Unani. The Indian tradition of health care has also promoted several non-drug therapies like Yoga (as old as Ayurveda) and Naturopathy (fairly recent). The system of homeopathy is also quite widespread within the country. Though Homoeopathy came to India in 18th Century, it completely assimilated in to the Indian culture and got enriched like any other traditional system hence it is considered as part of Indian Systems of Medicine. Thus India has the unique distinction of having seven recognized systems of medicine which are Ayurveda, Siddha, Unani, Yoga, Naturopathy, Amchi and Homoeopathy. These systems are better known as ISM & H (Indian systems of Medicine and homoeopathy) or AYUSH. The distinguished features of Indian systems of Medicine are their holistic nature. These systems have sustained themselves from long past. ISM systems are firmly embedded in the belief systems of people and are ‘culturally compatible’. These systems enjoy significant public patronage.

Despite of public faith and strengths of systems, Indian systems of Medicine were dominated by Western Medicine during British
rule. Only western medicine was recognized as legitimate and Eastern systems were actively discouraged. At the dawn of twentieth century, with the assertion of Indian nationalism, interest in Indian art and science reawakened and Indian systems of medicine began a gradual renaissance. Bhore Committee and Mudaliar Committee, which brought revolution in Health System of India, identified the importance of ISM.\cite{1,2} After that, the Indian government set up several high-level committees to advise it on the course of action it should adopt in relation to ISM. Later on the strengths of traditional system were internationally recognized and concern was voiced in the World Health Organization (WHO) Assembly in early 1970s. International apprehensions for Traditional Medicine, poor health indicators and inaccessible modern medical services forced Government of India to work towards the development of Indian systems of medicine. As a result, National Health Policy formulated in 1983 assigned an important role to ISM in the delivery of primary health care. Moving step forward separate National Policy on Indian systems of Medicine and Homeopathy was developed in 2002. This policy addressed all issues directly or indirectly related with ISM and provides directions to Government to move forward.

Mere formation of a policy is not sufficient. Policies denote just expectations. It is only at implementation stage that shortcomings appear in practice. Implementation of a policy depends upon various factors like governmental support, resources, organization, efficiency of institutions etc. To assess the achievement of stated objectives of a policy, its adequacy and efficiency evaluation should be done. Thus, it is important to find out how effectively the Policy on ISM &H has been implemented in the period of eight years.

Keeping the above mentioned points in view, the present investigation was undertaken with the objective to ascertain the degree to which ISM&H Policy has been implemented.

**Material and methods**

A record based secondary data analysis was conducted during November 2010 to April 2011. To assess the degree of implementation of ISM&H Policy, an implementation assessment tool was designed. It had various domains according to strategies described in policy. The tool was given to some of the experts for content validity. Based on their suggestions and recommendations, the tool was revised. There were 85 items in all. Various documents issued by Ministry of Health and Family Welfare (MOHFW), Department of Ayurveda Yoga & Naturopathy, Unani, Siddha and Homoeopathy (AYUSH), Central Council for Indian Medicine (CCIM), Central Council for Homoeopathy (CCH), Central Council for research in Ayurveda and Siddha (CCRAS), Central Council for Research in Homoeopathy (CCRH), Central Council for Research in Unani Medicine (CCRUM), Central Council for Research in Yoga and Naturopathy (CCRYN), National Medicinal Plant Board (NMPB), Central Drugs Standard Control Organization (CDSCO), Ministry of Commerce and Industry, Planning Commission etc. were analyzed. Websites of concerned agencies were also scrutinized.\cite{25-39}

**Analysis**

Each item number carried equal score. Score of ‘one’ was assigned to strategy/provision which had been implemented. Score of ‘zero’ was assigned to strategy/provision which had not been implemented or regarding which no information is available. The total score per component of policy was calculated into the percentage and graded: Excellent 76-100%, Good 51-75%, Fair 26-50% and Poor 0-25%. Similarly overall implementation status was also graded. The data was analyzed with the help of Microsoft Excel 2007.

**Results**

Overall implementation score of ISM&H Policy was found to be ‘Excellent’ (76.5%). Implementation of strategies addressing issues
of research, medicinal plants, drug standardization & quality control, ISM industry, revitalization of LHT, operational use of ISM&H in RCH, administration of ISM&H, development of special areas, sensitization of modern medical graduates to ISM&H and building awareness was found to be excellent. Implementation of strategies addressing issues of education and integration of ISM&H in healthcare system was found to be good. Implementation of strategies addressing issues of financing ISM&H, veterinary medicine, medical tourism and intersectoral cooperation was found to be fair. Implementation of strategies addressing issue of home remedy kit was found to be poor. Status of implementation of various provisions of ISM&H Policy is shown in table 1 and Implementation scores are shown in table 2.

**Discussion**

Patwardhan *et al.* (2005) identified urgent need to improve quality education. He questioned the hybrid curricula which produces inadequately trained graduates in modern and traditional systems. Documents revealed many initiatives which supports the betterment of education standards like ‘Education Policy Cell’ of department of AYUSH to deal with the matters related to quality education; Centrally Sponsored Scheme for Development of AYUSH Institutions, of research, medicinal plants, drug standardization & quality control, ISM industry, revitalization of LHT, operational use of ISM&H in RCH, administration of ISM&H, development of special areas, sensitization of modern medical graduates to ISM&H and building awareness was found to be excellent. Implementation of strategies addressing issues of education and integration of ISM&H in healthcare system was found to be good. Implementation of strategies addressing issues of financing ISM&H, veterinary medicine, medical tourism and intersectoral cooperation was found to be fair. Implementation of strategies addressing issue of home remedy kit was found to be poor. Status of implementation of various provisions of ISM&H Policy is shown in table 1 and Implementation scores are shown in table 2.

**Table 1. Implementation status of various provisions of ISM&H Policy (ISM&H Policy Implementation Assessment Tool)**

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Strategies for various components of ISM&amp;H Policy</th>
<th>Stage of Implementation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Not done</td>
</tr>
<tr>
<td><strong>Component: Education</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.</td>
<td>Legislative measures taken to regulate starting of a new college, increase in intake and introduction of new course of study</td>
<td></td>
</tr>
<tr>
<td>2.</td>
<td>Accreditation system whereby silver and platinum standards be given to ISM&amp;H colleges</td>
<td>√</td>
</tr>
<tr>
<td>3.</td>
<td>Establishment of model colleges and Centers of Excellence of ISM&amp;H</td>
<td>√</td>
</tr>
<tr>
<td>4.</td>
<td>National Institutes made fully functional as Centers of Excellence</td>
<td>√</td>
</tr>
<tr>
<td>5.</td>
<td>Study of Sanskrit in Ayurveda discipline and Urdu/Persian in Unani incorporated in the curricula</td>
<td>√</td>
</tr>
<tr>
<td>6.</td>
<td>The course curricula reviewed</td>
<td>√</td>
</tr>
<tr>
<td>7.</td>
<td>The course curricula revised to weed out unnecessary teaching materials, incorporate what is relevant keeping in view present requirements, include research achievements, reduce the component of modern medicine</td>
<td>√</td>
</tr>
<tr>
<td>8.</td>
<td>Nursing (AYUSH) education introduced</td>
<td>√</td>
</tr>
<tr>
<td>9.</td>
<td>Nursing (AYUSH) education regulated through existing or new regulatory councils</td>
<td>√</td>
</tr>
<tr>
<td>10.</td>
<td>Pharmacy education introduced for AYUSH</td>
<td>√</td>
</tr>
<tr>
<td>11.</td>
<td>Pharmacy education regulated through existing or new regulatory councils</td>
<td>√</td>
</tr>
<tr>
<td>12.</td>
<td>Schemes for providing vocational training for housewives, dais, nurses, etc</td>
<td>√</td>
</tr>
<tr>
<td>13.</td>
<td>Course for dietitians based on Ayurvedic and naturopathy approach to food and nutrition</td>
<td>√</td>
</tr>
<tr>
<td>14.</td>
<td>A separate regulatory council for Yoga and Naturopathy</td>
<td>√</td>
</tr>
<tr>
<td><strong>Component: Research</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15.</td>
<td>Priority accorded to research covering clinical trials, pharmacology, standardization, toxicology, study of pharmacology kinetics in respect of already identified areas of strength</td>
<td>√</td>
</tr>
<tr>
<td>16.</td>
<td>The research areas prioritized keeping in view the strengths of the systems and contemporary relevance giving due emphasis on preventive and promotive aspects</td>
<td>√</td>
</tr>
<tr>
<td>17.</td>
<td>Research on fundamental principles of ISM&amp;H</td>
<td>√</td>
</tr>
<tr>
<td>18.</td>
<td>Drug research to establish efficacy and safety of ISM &amp;H medicine to be accelerated by adopting rapid screening of herbs <em>invivo</em> or <em>invitro</em> in experimental settings</td>
<td>√</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>---</td>
<td>-------------------------------------------------------------------------------------------</td>
<td>---</td>
</tr>
<tr>
<td>19.</td>
<td>Disease oriented clinical drug research following “reverse pharmacology approach”</td>
<td>✓</td>
</tr>
<tr>
<td>20.</td>
<td>Identification and evaluation of promising and widely accepted practices and skills of</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>traditional healers in rural and tribal areas</td>
<td></td>
</tr>
<tr>
<td>21.</td>
<td>Revival of ancient literature – survey, collection, transcription/translation, editing and</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>publication of classical literature and text books on ISM</td>
<td></td>
</tr>
<tr>
<td>22.</td>
<td>Research studies introduced for reproductive systems of plants, shelf-life, storage</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>conditions</td>
<td></td>
</tr>
<tr>
<td></td>
<td><strong>Component: Medicinal Plants</strong></td>
<td></td>
</tr>
<tr>
<td>23.</td>
<td>Medicinal Plants Board acquire Statutory status (to be able to regulate registration of</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>farmers and cooperative societies, transportation, marketing of medicinal plants and</td>
<td></td>
</tr>
<tr>
<td></td>
<td>proper procurement and supply to pharmaceutical industry.)</td>
<td></td>
</tr>
<tr>
<td>24.</td>
<td>Priority given to encourage cultivation of medicinal plants recommended for cultivation</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>by Expert Committees in the first instance</td>
<td></td>
</tr>
<tr>
<td>25.</td>
<td>Schemes to provide proper buy-back arrangements</td>
<td>✓</td>
</tr>
<tr>
<td>26.</td>
<td>Central Government assists following States on priority to avail of the benefits of</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>Medicinal Plant Sector Schemes to Chhattisgarh, Jharkhand and Uttrakhand</td>
<td></td>
</tr>
<tr>
<td>27.</td>
<td>Encouragement given for R&amp;D on rare and endangered plants</td>
<td>✓</td>
</tr>
<tr>
<td>28.</td>
<td>Schemes for growing medicinal plants for production and sale of plant based products</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>including herbal tea encouraged through Women’s groups and Tribal agencies</td>
<td></td>
</tr>
<tr>
<td>29.</td>
<td>Issues related to export import of Medicinal Plants addressed through Identification of</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>market, Segmentation of market, simplifying import and export procedures</td>
<td></td>
</tr>
<tr>
<td></td>
<td><strong>Component: Intellectual Property Rights &amp; Patents</strong></td>
<td></td>
</tr>
<tr>
<td>30.</td>
<td>Progressive creation of digital library</td>
<td>✓</td>
</tr>
<tr>
<td>31.</td>
<td>A <em>sui generis</em> system set up to provide grassroots innovators of plant based knowledge</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>an incentive to disclose knowledge</td>
<td></td>
</tr>
<tr>
<td></td>
<td>**Component: Integration of ISM &amp; H and National Health Care Programmes and Delivery System</td>
<td></td>
</tr>
<tr>
<td>32.</td>
<td>Efforts made to integrate and mainstream ISM&amp;H in health care delivery systems</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>including National Programmes</td>
<td></td>
</tr>
<tr>
<td>33.</td>
<td>A range of options for utilization of ISM &amp; H manpower in the health care delivery system</td>
<td>✓</td>
</tr>
<tr>
<td>34.</td>
<td>Goal oriented role and responsibility to the ISM workforce</td>
<td>✓</td>
</tr>
<tr>
<td>35.</td>
<td>An ISM&amp;H wing encouraged and supported at the primary health care level</td>
<td>✓</td>
</tr>
<tr>
<td>36.</td>
<td>Setting up of specialty centers and ISM clinics At the PHC and district hospital level</td>
<td>✓</td>
</tr>
<tr>
<td>37.</td>
<td>Referral ISM hospitals in the country be renovated, modernized and upgraded to provide</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>the full range of ISM treatment</td>
<td></td>
</tr>
<tr>
<td>38.</td>
<td>Identification of the referral hospitals made according to current availability of</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>motivated staff, OPD &amp; IPD attendance and locational advantages</td>
<td></td>
</tr>
<tr>
<td>39.</td>
<td>Central government assist specialty hospitals of allopathy who wish to establish</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>Panchkarma and Kaharshruta facilities</td>
<td></td>
</tr>
<tr>
<td>40.</td>
<td>Private allopathic hospitals encouraged to set up specialist treatment centers of ISM&amp;H</td>
<td>✓</td>
</tr>
<tr>
<td>41.</td>
<td>Hiring charges of Vaidyas/Hakims/Homoeopaths reimbursed to Private allopathic hospitals</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>entering into research collaboration protocols</td>
<td></td>
</tr>
<tr>
<td>42.</td>
<td>States encouraged to reenact or modify laws governing the practice of modern medicine</td>
<td>✓</td>
</tr>
<tr>
<td></td>
<td>by ISM &amp; H practitioners</td>
<td></td>
</tr>
<tr>
<td>43.</td>
<td>States encouraged to raise the salary of ISM &amp; H practitioners</td>
<td>✓</td>
</tr>
<tr>
<td>44.</td>
<td>States encouraged to raise social/professional status of ISM&amp;H practitioners</td>
<td>✓</td>
</tr>
</tbody>
</table>
### Table 1. Continued...

<table>
<thead>
<tr>
<th>Component: Drug Standardization and Quality Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>45. Drugs &amp; Cosmetics Act amended (to cover grant of manufacturing licenses for intermediate or partially processed herbal mixes and pharmacopoeial standards evolved for these intermediaries)</td>
</tr>
<tr>
<td>46. New Legislation covering a vast range of nutraceuticals and food supplements which are neither covered by the drug licensing nor food licensing</td>
</tr>
<tr>
<td>47. Quality Control Centers (ISM &amp; H) set up or recognized on a Regional basis</td>
</tr>
<tr>
<td>48. Pharmacopoeial work related to drugs of Ayurveda Unani Siddha Homoeopathy</td>
</tr>
<tr>
<td>49. Financial support given for acquisition of ISO 9000 certification by ISM industry</td>
</tr>
<tr>
<td>50. States would be advised to augment facilities for drugs manufacture and testing of the drugs.</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: ISM industry</th>
</tr>
</thead>
<tbody>
<tr>
<td>51. ISM industry given priority industry status</td>
</tr>
<tr>
<td>52. Efforts made to secure fiscal incentives and Tax concessions to ISM industry within overall taxation policy to make them viable and competitive</td>
</tr>
<tr>
<td>53. ISM Industry encouraged to adopt modern dosage form and follow reasonable shelf-life</td>
</tr>
<tr>
<td>54. Use of classical preparations encouraged</td>
</tr>
<tr>
<td>55. Guidelines framed for patent and proprietary medicines</td>
</tr>
<tr>
<td>56. Efficacy and safety studies made mandatory for manufacturers to grant license for new Patent Proprietary medicines</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Revitalization of Local Health Traditions</th>
</tr>
</thead>
<tbody>
<tr>
<td>57. Revitalization of folk health traditions related to birth attendants, herbal healers, bone settlers, Visha healers etc., figured in the agenda of the ISM sector to be selectively Identified, Reinforced Validated, Propagated for use in a wider community</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Home Remedy Kits</th>
</tr>
</thead>
<tbody>
<tr>
<td>58. Scheme for supply of identified medicine in Home Remedy Kit</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Veterinary Medicine</th>
</tr>
</thead>
<tbody>
<tr>
<td>59. Any Ayurvedic institute for veterinary medicine/ care</td>
</tr>
<tr>
<td>60. Any Homoeopathy institute for veterinary medicine/ care</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Operational Use of ISM in RCH</th>
</tr>
</thead>
<tbody>
<tr>
<td>61. ISM &amp;H medicines introduced in Reproductive &amp; Child Health (RCH)</td>
</tr>
<tr>
<td>62. Operational research studies of ISM&amp;H in RCH</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Financing ISM&amp;H</th>
</tr>
</thead>
<tbody>
<tr>
<td>63. ISM &amp;H share of National Health Budget raised to 10% of the total health plan at the Central level</td>
</tr>
<tr>
<td>64. Budget growth of ISM&amp;H sector designed to climb at the rate of 5% in eleventh Five Year Plan</td>
</tr>
<tr>
<td>65. Centre Government directly provided or earmarked budgets for Consolidation of infrastructure</td>
</tr>
<tr>
<td>Purchase of drugs</td>
</tr>
<tr>
<td>Support for opening specialty clinics and ISM services</td>
</tr>
<tr>
<td>Drugs listed in the Essential Drug Lists for Ayurveda, Unani and Homeopathy Medicine for specialty centres and ISM clinics established at PHC and District hospitals</td>
</tr>
</tbody>
</table>
**Table 1. Continued...**

<table>
<thead>
<tr>
<th>Component: Administration of the ISM Sector</th>
</tr>
</thead>
<tbody>
<tr>
<td>66. States encouraged to post state level Secretaries and Directors of ISM &amp; H</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Developments of Special Areas – North East &amp; New States</th>
</tr>
</thead>
<tbody>
<tr>
<td>67. Encouragement in North Eastern States for Utilization of medicinal plants, Identification of tribal medical practices</td>
</tr>
<tr>
<td>68. Setting up of dispensaries and Need based teaching institutions for ISM</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Medical Tourism</th>
</tr>
</thead>
<tbody>
<tr>
<td>69. Facilities for Panchakarma and Yoga encouraged to be offered in hotels</td>
</tr>
<tr>
<td>70. Road Shows organized abroad by providing services of Vaidas and Hakims and Yoga demonstration</td>
</tr>
<tr>
<td>71. Scheme for accreditation of Panchakarma &amp; Yoga facilities introduced</td>
</tr>
<tr>
<td>72. ISM parks developed in collaboration with State Tourism authorities</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Inter-Sectoral Co-operation</th>
</tr>
</thead>
<tbody>
<tr>
<td>73. Linkages established with other departments (like Culture, Tourism, Tribal, Rural development, Railways, ESI, etc.) to promote and propagate the use of ISM&amp;H through the establishment of clinics of ISM&amp;H or by allowing reimbursement of treatment charges of ISM&amp;H</td>
</tr>
<tr>
<td>74. Introduction of knowledge relating to the properties of medicinal plants and preparation of simple home remedies from ISM in the school curriculum</td>
</tr>
<tr>
<td>75. Naturopathy diets encouraged in schools, colleges and offices</td>
</tr>
<tr>
<td>76. Yogic exercise encouraged in schools, colleges and offices</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Exposing the Foreign and Indian Modern Graduates to ISM</th>
</tr>
</thead>
<tbody>
<tr>
<td>77. Modules formulated for introducing Ayurveda and Yoga to medical schools and institutions abroad</td>
</tr>
<tr>
<td>78. Courses of long duration say one year to two years started for allopathic doctors from India and abroad</td>
</tr>
<tr>
<td>79. A package of introductory lectures given during the regular medical course for foreign students.</td>
</tr>
<tr>
<td>80. At PG and Doctorate level, scholarships given to undertake medical research on ISM</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Component: Building Awareness</th>
</tr>
</thead>
<tbody>
<tr>
<td>81. Programmes on the utility and effectiveness of ISM&amp;H launched through the electronic and print media</td>
</tr>
<tr>
<td>82. Special incentives given to colleges and groups of students who come up with innovative ideas for popularizing ISM&amp;H</td>
</tr>
<tr>
<td>83. Students of Management and Social Science courses offered internships to work on popularizing and marketing ISM&amp;H</td>
</tr>
<tr>
<td>84. A significant portion of the budget would be assigned for IEC on healthy life styles and preventive health through ISM&amp;H approaches</td>
</tr>
<tr>
<td>85. NGOs used for popularizing Yoga in primary schools, residential colonies and in industrial units</td>
</tr>
</tbody>
</table>

Scheme for Grant-in-aid to non-profit/ nongovernmental AYUSH organizations/institutions for up gradation to centers for Excellence, Scheme for supporting re-orientation training continuing medical education and exposure programs of AYUSH and 'Assistance for
### Table 2. Implementation scores of various components of ISM&H Policy

<table>
<thead>
<tr>
<th>ISM&amp;H Policy Strategy No.</th>
<th>Component</th>
<th>Maximum Score</th>
<th>Score obtained</th>
<th>Percentage</th>
<th>Implementation status</th>
</tr>
</thead>
<tbody>
<tr>
<td>16.1</td>
<td>Education</td>
<td>14</td>
<td>9</td>
<td>64.3%</td>
<td>Good</td>
</tr>
<tr>
<td>16.2</td>
<td>Research</td>
<td>8</td>
<td>8</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.3</td>
<td>Medicinal plant</td>
<td>7</td>
<td>7</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.4</td>
<td>Intellectual property rights and patents</td>
<td>2</td>
<td>2</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.5</td>
<td>Integration of ISM&amp;H</td>
<td>13</td>
<td>7</td>
<td>53.8%</td>
<td>Good</td>
</tr>
<tr>
<td>16.6</td>
<td>Drug standardization and quality control</td>
<td>6</td>
<td>6</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.7</td>
<td>ISM industry</td>
<td>6</td>
<td>6</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.8</td>
<td>Revitalization of Local Health Traditions</td>
<td>1</td>
<td>1</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.9</td>
<td>Home Remedy Kits</td>
<td>1</td>
<td>0</td>
<td>0%</td>
<td>Poor</td>
</tr>
<tr>
<td>16.10</td>
<td>Veterinary medicine</td>
<td>2</td>
<td>1</td>
<td>50%</td>
<td>Fair</td>
</tr>
<tr>
<td>16.11</td>
<td>Operational Use of ISM in RCH</td>
<td>2</td>
<td>2</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.12</td>
<td>Financing ISM&amp;H</td>
<td>3</td>
<td>1</td>
<td>33.3%</td>
<td>Fair</td>
</tr>
<tr>
<td>16.13</td>
<td>Administration of the ISM Sector</td>
<td>1</td>
<td>1</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.14</td>
<td>Developments of Special Areas</td>
<td>2</td>
<td>2</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.15</td>
<td>Medical Tourism</td>
<td>4</td>
<td>2</td>
<td>50%</td>
<td>Fair</td>
</tr>
<tr>
<td>16.16</td>
<td>Inter-Sectoral Co-operation</td>
<td>4</td>
<td>2</td>
<td>50%</td>
<td>Fair</td>
</tr>
<tr>
<td>16.17</td>
<td>Exposing the Foreign and Indian Modern Graduates to ISM</td>
<td>4</td>
<td>4</td>
<td>100%</td>
<td>Excellent</td>
</tr>
<tr>
<td>16.18</td>
<td>Building Awareness</td>
<td>5</td>
<td>4</td>
<td>80%</td>
<td>Excellent</td>
</tr>
<tr>
<td>All strategies/ provisions of ISM&amp;H Policy</td>
<td>85</td>
<td>65</td>
<td>76.5%</td>
<td>Excellent</td>
<td></td>
</tr>
</tbody>
</table>

exchange programme/seminar/conference/workshop on AYUSH; The IMCC (Amendment) Bill, 2005 and the HCC (Amendment) Bill 2005 to bringing about transparency and accountability in the functioning of the regulatory Councils; The Indian Medicine and Homoeopathy Pharmacy Council Bill 2005 to establish a Central Pharmacy Council for Indian Medicine and Homoeopathy to regulate & standardize Pharmacy and Establishment of an All India Institute of Ayurveda at Sarita Vihar, New Delhi as “Center of Excellence for development and scientific validation of Ayurveda. Implantation of these strategies to improve AYUSH education was found to be ‘good’. Still lacunae are left. There is no separate regulatory council for yoga & naturopathy, nursing (AYUSH) and pharmacy (AYUSH). However it is learnt that CCRYN is discharging the function of regulatory body for Yoga and Naturopathy. No accreditation system was introduced for academic institutions.

In this study, 100% implementation of all strategies related to research was found on analyzing the government sources. Research councils have started clinical drug research following reverse pharmacological approach which was also advocated by Patwardhan et al. (2008) in his article. Golden Triangle Partnership Project and Extra Mural Scheme are worthy efforts of government for promotion of research in ISM&H. Government is favoring research in AYUSH probably because these
systems are getting attention of vast population even in western countries. Research is very important to validate principles, products and therapies of these systems. Validating these will provide the government a firm position in global market. This will yield higher benefits in long run.

Implementation of strategies related to medicinal plants was found to be excellent. Most of health systems in ISM&H are primarily herbal based. Framing of National Medicinal Plants Board, implementation of projects for development of Good Agricultural and Good Collection Practices for medicinal plants in association with WHO and Centrally Sponsored Scheme of National Mission on Medicinal Plants is significant steps to ensure authentic raw material. Medicinal Plants are backbone in most of systems of AYUSH and responsible for survival of these systems. Herbal products have high potential of marketing and these are more acceptable than herbo mineral products in global market. Medicinal plants are also being extinct due to increasing agricultural land. So government is focusing on conservation of medicinal plants.

**Mukherji and Wahile (2006)** considered safety, efficacy and quality helpful to rationalize the use of natural products in healthcare.[5] **Shinde et al. (2009)** advocated incorporation of traditional knowledge of herbs and modern standardization techniques to result in ‘Total Quality Management’. [16] **Unikrishnan (2010)** emphasized the need to seriously consider safety, efficacy and quality of traditional medicine.[7] **Banerjee (2002)** found that standardization and validation of traditional medical products are key issues addressed in ISM&H Policy.[8] Government sources claimed 100% implementation of strategies focused on drug standardization and quality control. Centrally Sponsored Scheme of Quality Control of ISM&H drugs for enforcement of provisions of Drug & Cosmetic Act and Drug Control Cell (AYUSH) is working in the Department to deal with the matters pertaining to Drug Quality Control and regulation of Ayurveda, Siddha and Unani drugs were comes out as major initiatives. Recent controversies regarding the safety of herbo-mineral drugs and absence of standard protocol for various AYUSH products Government of India to adopt measure to ensure the safety, standardization and quality control of AYUSH products.[9,10,11] Global market especially in the West is very much concerned with safety and quality control of AYUSH products. It is very important to ensure that all traditional medicine manufacture is in accordance with good manufacturing practices.[12,13,14]

**Unikrishnan (2010)** found it essential to appropriately integrate traditional medicine with mainstream health system without compromising diversity and unique aspects.[7] **Vaidya (2005)** advocated synergy between ISM and modern medicine.[15] **Rao et al. (2011)** advocated to encourage task-shifting and mainstreaming doctors and practitioners who practice ISM&H to work in these areas while adopting other innovative ways of augmenting human resources for health.[16] **Kumar (2010) and Chanana (2010)** recommended the use of Ayurvedic and Homoeopathic workforce in Public Health. ISM&H Policy provides various strategies to integrate ISM&H in mainstream health system.[17,18] In present study, implementation status of these strategies was found to be 53.8%. Government has started working on the recommendation on integration and cafeteria approach which was pending from a long period. National Rural Health Mission (NRHM) has been the most important step towards development of AYUSH. An ambitious initiative is also under way to provide the services of AYUSH doctors in CHCs/PHCs under the NRHM. The Department of AYUSH has been providing substantial financial assistance to States for opening of AYUSH wings in district hospitals and specialty/OPD clinics in other hospitals with a view to provide AYUSH facilities along with modern medicine under one roof. Central sector scheme of grant-in-aid for promotion of AYUSH intervention in public health initiatives is a step ahead by Department of AYUSH.

ISM&H Policy directs operational use of ISM&H in RCH. Implementation of these
strategies was found to be excellent. Nine Ayurvedic and five Unani drugs are being supplied under the National Reproductive Child Health (RCH) Programme. These drugs have been identified for the treatment of common ailments of pregnant women, adolescent girls and children. Under National Rural Health Mission one Ayurvedic drug ‘Punarnavadi Mandura’ for anaemia for pregnant women and adolescent girls is included in the ASHA Kit. Role of traditional medicine in RCH has been recognized to a large extent. Infant mortality rate and maternal mortality rates are major indicators of health performance and overall development of a nation. India is lagging behind the targets set for such indicators. Biomedical approach to RCH has not resulted in significant decline in IMR and MMR. It lacks to address indigenous culture. Government has perceived the need to incorporate AYUSH systems to improve such indicators as these are more acceptable and practiced in Indian society.

Implementation status of strategies which address issues of intellectual property rights, revitalization of LHT and ISM industry was found to be 100%.

Intellectual property rights and patents of AYUSH products and therapies are major controversial issues as traditional wisdom is being validated by some agencies and getting patented by them. Moreover there can be various interpretations of ancient texts describing these medicines and therapies. Bodekar (1999) emphasized on development information resource and exchange on issues pertaining to Intellectual property rights over traditional medical knowledge.[19]

Traditional Knowledge Digital Library (TKDL) has been created in five international languages in order to enable access by International Patent Office under a non-disclosure agreement for facilitating patent searches so as to prevent the grant of wrongful patents based on Indian traditional medicinal knowledge. Other projects like Ayusoft, Triskandha Kosha and medicinal plant database are also attempting systemic documentation of AYUSH in India.[20]

Government has also recognized importance of Local Health Traditions as these has potential to address some major health issues. Moreover these are only available health services to a large rural and tribal population of India. For revitalization of local health traditions, scheme for grant-in-aid to nonprofit/non-governmental AYUSH organizations/institutions for revitalization of local health traditions, midwifery practices etc in order to enhance health security of rural community is being promoted. “Dadi ma ka batua” is an innovative scheme in the J&K, which plans to include traditional home remedies in the AYUSH drug kit. Madhya Pradesh has an innovation called Gyaan ki Potli which too plans to include prevalent and useful local health traditions / remedies which are accessible and affordable for various ailments as a step forward for LHT revitalization.[21]

Central sector scheme for development of AYUSH cluster addresses issues of ISM industry. Efforts have been made for tax concessions for ISM industry. Government is largely focusing on implementation of strategies to enhance ISM industry as these involve direct and immediate monitory benefits.

Implementation status of strategies which address issues of building awareness regarding ISM&H was found to be 80%. To build Public awareness Ksharasutra Campaign, National Campaign on Homoeopathy for Mother and Child Care, National Campaign on Geriatric Care through Ayurveda, Yoga for Health, Amla campaign and Arogya fairs are significant steps.

ISM&H Policy directs use of Ayurveda and Homoeopathy in Veterinary medicine. National Veterinary - Ayurveda Research Institute, Lucknow is appreciable step to promote use of Ayurveda in veterinary. But there are not any Homoeopathic, Unani or Siddha institutions for this purpose. Still use of Ayurveda and Homoeopathy in veterinary medicine is an unexplored area despite of the fact that it has been described in ancient Ayurveda. There is need for research to explore these systems along with Siddha, Naturopathy and local Health Traditions related with animals.
ISM&H Policy directs to raise share of ISM&H to 10% of central health budget. Even after nine years of enforcement of ISM&H Policy share of ISM&H still stands at approx. 2% in central health budget. Implementation status of strategies directed to financing ISM&H was found to be 33.3%. Despite the recommendations by various international and national agencies, government is not able to increase budget share of ISM&H. \textit{Patwardhan et al. (2005)} recognized paucity of funds to ISM&H as major concern which was in accordance with results of our study.\textsuperscript{[1]} The share of health budget in total central budget and further the share of AYUSH in central health budget has been major lacuna in health sector of India. \textit{Bodekar (2000)} considered that through marginalized budget share in National Health budget, traditional medicine research, quality and training suffers adversely.\textsuperscript{[22]}

Implementation of strategies directed to increase medical tourism was found to be ‘fair’. Kerala and Himachal has been torch bearer for whole nation which successfully attracted tourists from entire world for AYUSH treatments. \textsuperscript{[23]} Government of Kerala also initiated accreditation system for hotels in state on the basis of ISM services which these provide. It has significantly contributed to state’s economy. This success story can be replicated to all states and at central level. Government of India is moving ahead to make India hub of medical tourism. Few states like Gujarat and Tamil Nadu has come forward to promote medical tourism through ISM.\textsuperscript{[40,41]} Himachal Pradesh government is giving added priority to boost health tourism, develop health resorts and popularise Panchkarma.\textsuperscript{[42]} ISM can attract whole world due to their ancient wisdom. It can contribute to medical tourism effectively. Central and state governments need to identify the potential of traditional wisdom and showcase AYUSH services to world.

All ancient health systems give emphasis on healthy diet. Dietary approaches described in ISM can be promoted to combat emerging trend of unhealthy fast food. No major scheme was found to promote AYUSH home remedies. Government has largely ignored this sector. This can be because of the fact that it do not yield good market prospect. Promotion of proved home remedies can be a good measure to promote the health of people. These can be very cost effective and have potential to serve large rural and poor population of India. Concept of Ayurvedagram in Chhattisgarh is good step towards introducing Ayurveda in routine life of people.\textsuperscript{[21]}

\textbf{Conclusion}

Overall implementation status of ISM&H Policy was found to be ‘Excellent’. Government has largely focused on issues of research, medicinal plants, drug standardization and quality control as these yield profits in global market. Operational use of AYUSH in RCH, revitalization of LHT and development of special area are also found to be area of interest of government as these have capability to improve health indicators and quality of life index. Education and integration of AYUSH with mainstream health system were found to be on priority list of government for implementation but a still lot is to be done. Range of options for utilization of AYUSH manpower and referral system was found to be rudimentary. No protocol was found for goal oriented role and responsibility to the ISM workforce. Use of AYUSH in Veterinary medicine emerged as one of most unexplored area despite of recognition in policy. Emphasis on ‘Home remedies’ and ‘Promotion of diet based on Ayurvedic and Naturopathy approach’ was inadequate. Scarcity of funds emerged as major factor to hinder the implementation of ISM&H Policy. Efforts to enhance medical tourism through AYUSH were limited to only specific states. Government should support ISM&H by a significant increase in budget. SWOT (Strengths, Weaknesses, Opportunities, and Threats) approach is needed.\textsuperscript{[24]}

Each system must focus in their strong points.

\textbf{References}


24. Rastogi S: Counting the strengths and countering the weaknesses: Applying SWOT analysis into AYUSH for its better appreciation and application. Annals Ayurvedic Medicine 2012; 1(1): 10-17

For the convenience of readers, referred websites are also written:


26. Department of AYUSH, MOHFW, GOI: http://www.indianmedicine.nic.in/


33. National Medicinal Plants Board (NMPB): http://nmpb.nic.in/
34. Homoeopathic Pharmacopoeia Laboratory (HPL): http://www.hplism.nic.in/
35. Indian Medicines Pharmaceuticals Corporation Ltd. (IMPCL): http://www.impclmohan.nic.in/
36. Pharmacopoeia Laboratory of Indian Medicine (PLIM): http://www.plimism.nic.in/
38. Ministry of Tourism, GOI: http://tourism.gov.in/
41. Tamil Nadu Medical Tourism: http://www.medicaltourismintamilnadu.com/

Address for correspondence: Dr. Balpreet Singh, Centre for Public health, Institute of Emerging Areas in Science and Technology, Punjab University, Chandigarh (India). Email: drbalpreetsaini@gmail.com
EFFICACY AND SAFETY OF POLY HERBAL FORMULATION WH-1 FAS-3 KIT AND FLUCONAZOLE IN FEMALE PATIENTS WITH VAGINOSIS / VAGINITIS

KULKARNI CHANDA,¹ DEB JARITA,² MISHRA DEVESH,³ DIAS MARY,⁴ MHASKAR RITA⁵ AND A.S. MOHAMMED⁶

Division of Clinical Pharmacology, St. John’s Medical College, Bangalore - 560034 Karnataka (India)

Abstract: Context: Vaginosis/vaginitis is one of the commonest symptoms in gynaecological outpatient setting and involves multiple causative organisms – bacteria, fungi and protozoa. The available treatment options produce a variety of adverse effects and do not decrease recurrence.

Aims: The purpose of this study was to examine if poly herbal formulation WH-1 has better efficacy and safety profile compared to the modern medication available for treatment of vaginosis/vaginitis.

Methods and Material: The present open labelled, randomized trial in adult women, with diagnosis of vaginosis/vaginitis, involved study patients in two groups who received either – WH-1 or standard treatment [FAS-3 kit or fluconazole], based on microbiological evaluation. Efficacy outcome measures were assessed subjectively, clinically and microbiologically, while safety was assessed subjectively and using biochemical investigations. The data was analysed using unpaired ‘t’ test, χ² test, and ‘t’ test as appropriate.

Results: Between the groups comparison for demographic profile and chronicity of infection did not show significant difference. The clinical and microbiological improvement was comparable in two groups. The microbiological improvement in patients from both WH-1 and FAS-3 kit treated groups was similar while the adverse effects were significantly lower in patients treated with WH-1 [P < 0.05].

Conclusions: The efficacy outcome measures with poly herbal preparation WH-1 was found to be similar compared to standard treatment with lesser adverse effects. WH-1 may therefore be considered as a safer and alternative treatment option in subjects with vaginosis/vaginitis. However, studies involving long term treatment and follow up in larger number of patients are necessary to confirm the findings.

Keywords: WH-1, FAS-3 kit, fluconazole, vaginosis, vaginitis, clinical evaluation, medicinal plants, Poly herbal formulations, Woodfordia floribunda, Cyperus scariosus, Bombax malabaricum, Symlocos racemosa and Caesalpinia bonduc.

Introduction

Vaginosis/vaginitis, is one of the most common reasons for a female patient to consult a physician in a primary care setting. The infection often becomes chronic with difficulty in its eradication. The three most common types of acute vaginitis are Bacterial Vaginosis (BV) Vaginal Candidiasis (VC) and Trichomonal Vaginitis (TMV). It is reported that BV is the most common cause of vaginitis in reproductive age group accounting for 50% of cases with nearly 75% of all adult women having had at least one genital Candida infection in their lifetime.¹

The standard treatment includes systemic/topical antifungal, anti-protozoal and antibacterial agents, which decrease infection temporarily and often disrupt the normal vaginal flora which may lead to recurrent infection.

A preliminary single arm trial with WH-1 in the form of soft gelatine capsule containing - Woodfordia floribunda (Dhataki flower), Cyperus scariosus (Musta root), Bombax malabaricum (Mocharas gum), Symlocos racemosa (Lodhra

1. Professor & Head, Division of Clinical Pharmacology   2. Tutor, Pharmacology   3. Lecturer, Clinical Pharmacology   4. Assistant Professor, Microbiology   5. Professor, Obstetrics & Gynaecology   6. Assistant Professor, Community Health
root) and *Caesalpinia bonduc* (Lata Karanja seed) is reported to be useful in BV, VC as well as TMV\(^2\) due to its antifungal, antimicrobial, antiseptic, astringent and demulcent properties.\(^3\)

Therefore, the present study was designed to compare safety, efficacy and spectrum of activity of WH-1 with that of FAS-3 Kit and fluconazole, in patients with vaginosis/vaginitis.

**Materials and Methods**

Study procedure, patient selection and randomization:

The study was carried out by the Division of Clinical Pharmacology, in collaboration with the departments of Gynaecology and Microbiology after obtaining approval from Institutional Ethical Review Board and ICH-GCP-2008 Seoul amendment and ICMR-2006 Guidelines were followed during the study procedure. The study was registered in Clinical Trial Registry of India [CTRI-020911479-0607200997748].

This was a randomized, open-labelled, comparative safety and efficacy study in subjects with vaginosis / vaginitis. Married women between 18 to 60 yrs of age, diagnosed to have vaginitis on the basis of symptoms, clinical and microbiological examination, were enrolled after obtaining written and informed consent from them as well as from their spouses. Pregnant and lactating women, with clinical and biochemical evidence of major systemic disorder, women with multiple partners with history of alcohol, drug dependence, participation in any other study within six weeks preceding day one of the present study, were excluded.

A specially designed case record form (CRF) was used to enter characteristic demographic data, and clinical symptoms of both acute [< 2 months duration] and chronic [> 2 months duration] vaginosis/vaginitis. The microbiological assessment was carried out to determine type of infection and subjects with confirmed diagnosis of vaginitis/vaginosis were then randomized manually to receive test or standard treatment.

**Objectives**

The primary objective was to examine the efficacy and safety profile of poly herbal test formulation WH-1 and compare the same with FAS-3 kit or fluconazole. The spectrum of activity of study drugs against various organisms causing vaginitis/vaginosis was also evaluated.

**Manufacturing process**

The raw material from various plant sources - *Woodfordia fruticosa* flower, *Cyperus*...
rotundus Rhizome, Bombax ceiba gum, Symlocos recemosa stem bark and Asteracantha longifolia seeds were identified as per API parameters (Table 1). An extract of first three products was mixed with five times the quantity of water/solvent. The last two items were then pulverized into a fine mesh powder. The extract and powder were mixed with ghee and dispensed as soft gelatine capsules for study purpose.

Medication intervention

The study products WH-1, poly herbal preparation 650 mg capsule each and standard drug - FAS-3 kit or fluconazole, were administered to two separate groups. The female study subjects along with respective partners also received study medications as a standard protocol. Group – A, received test drug WH-1, one capsule three times a day with water after food for a period of 10 days [composition Table 1].

Subjects under standard treatment Group – B, received contents of FAS-3 Kit [1 tablet fluconazole, 150 mg, after breakfast; one tablet of azithromycin 1 gm, one hour before lunch; and 2 tablets of secnidazole 1 gm each after dinner] as a single dose. Subjects who were positive for Candida infection were randomized to receive fluconazole 150 mg, stat or WH-1 as mentioned above. There was one follow-up on day fourteen.

Instructions to subjects

The study subjects and their partners were instructed not to take any other medication during the trial period.

Compliance to study medications and adverse reactions were monitored by counting the number of unused medications at each clinic visit and on the day of final follow up visit and recording through a calendar provided to each patient.

Assessment of safety and efficacy outcomes

The following subjective and microbiological assessments were carried out and compared on two occasions - prior to and after the treatment period to evaluate treatment outcomes.

1. Assessment of efficacy

a. Subjective assessment for improvement in severity of symptoms viz - itching, discharge, dysuria and dyspareunia was scored –
   Mild = 1, Moderate = 2 or Severe = 3.
b. Clinical assessment was by gynaecological examination for presence or absence of infection and was graded as - cured, improved, not improved or aggravated.
c. Microbiological assessment was carried out by collecting two vaginal swabs from each subject at the first visit and on day 14 after the treatment period and processed for –
   (i) pH of vaginal smear using pH paper
   (ii) Whiff test using 10% KOH and amine odour production. A strong fishy odour was considered suggestive of Bacterial Vaginosis [BV]
   (iii) vaginal smears, were subjected to Gram staining and graded based on Nugent score for the diagnosis BV
   (iv) the wet mount preparation examined for presence of yeast cells, clue cells and actively mobile trophozoites as Trichomonas vaginalis [TMV]
   (v) smear for diagnosis of candidiasis when showed presence of numerous yeast cells and pus cells
   (vi) presence of - bacteria, candida, trichomonals and numerous pus cells in a smear was grouped under mixed infection
   (vii) lastly samples which did not fit into - Bacterial vaginosis, Candidiasis, Trichomoniiasis; but had numerous pus cells with normal vaginal flora; or have altered vaginal flora showing abnormal Gram positive bacilli not resembling lactobacilli, or Gram negative bacilli not resembling Gardnrella/Prevotella morphotype were categorised as nonspecific vaginitis. Smears with presence of Polymorphonuclears (PMNs) was graded in a semi quantitative manner as – none and +, ++, +++ representing - mild, moderate, and severe infection respectively.
The follow up swabs collected from each of the subjects at clinic visit were processed similarly and microbiological improvement was considered as positive for –

(i) Bacterial vaginosis: when Nugent score dropped from > 6 before the treatment to < 3 after treatment
(ii) Vaginal candidiasis: with reduction in yeast cells and PMNs on Gram staining
(iii) Trichomoniasis: with absence of motile trophozoites in wet mount and
(iv) nonspecific vaginitis: when reduction noticed in PMNs from +++ to ++ or +; from ++ to + or none, with return of lactobacilli as normal vaginal flora.

2. Assessment of Safety

Safety of treatment medications was by eliciting information subjectively through non-leading questions and biochemical investigations, carried out for female subjects entering the study and respective partners who received WH-1. The adverse effects were graded as – Mild = 1, Moderate = 2 or Severe = 3 and were recorded from the calendar provided and categorised based on the system affected.

The Biochemical parameters were estimated on day zero and day 14 for those patients with respective partners who received WH-1 and included - random blood sugar (RBS), Kidney Function Tests (KFT) and Liver Function Tests (LFT). The values were considered normal when they matched with standard laboratory values. Since the safety of standard medications is well established patients in this group were not subjected for biochemical investigations.

Statistical analysis

The values for age distribution and chronicity of vaginal infection was compared between test and standard treatment groups using unpaired ‘t’ test. The statistical analysis for parameters of microbiological and clinical improvement was carried out using $\chi^2$ test. The comparison of subjective improvement in symptoms between the groups, and safety parameters for biochemical as well as adverse reactions were carried out using ‘t’ test and $\chi^2$ test respectively.

Results

A total 57 female subjects with confirmed diagnosis of vaginitis/vaginosis were enrolled in the study. There were 48 subjects positive for mixed infection/nonspecific vaginitis and nine positive for candida. All subjects met the inclusion/exclusion criteria, were randomized to group A and B and 41 subjects completed the study; seven were lost for follow up and were considered as dropouts. Subjects positive for candida were not included in the statistical analysis.

Twenty one subjects received study drug WH-1 and twenty subjects received standard treatment (Table 2).

The test and standard treatment groups were homogenous with respect to age and chronicity of infection \( [P > 0.05] \) (Table 3). The improvement between the groups with respect to microbiological and clinical parameters calculated using $\chi^2$ test did not show a significant difference \( [P > 0.05] \).

The difference in mean post-treatment values for subjective improvement when compared between the test \( [1.65 \pm 0.988] \) and standard \( [2.50 \pm 1.539] \) groups using unpaired students ‘t’ test, showed marginally higher improvement in the group which received standard treatment \( [P = 0.044] \).

The comparison of biochemical parameters between the two groups, as a part of evaluation of safety profile using ‘t’ test, did not show
Efficacy and safety of poly herbal formulation for vaginosis/vaginitis

The adverse reaction was seen in 1/21 patients in the test drug group viz. drowsiness, as against 6/20 in the standard treatment group who showed mild – gastritis=2, diarrhoea=2, nausea and vomiting=1, body ache and fatigue = 1. The occurrence of adverse drug reactions was significantly higher among subjects who received standard treatment compared to group treated with test drug [$\chi^2$ test, $p = 0.0396$].

There were nine subjects who were microbiologically positive for Candida and among these, eight completed the study with one dropout. Five subjects from this group received WH-1 and three fluconazole. They were analyzed descriptively after the treatment period. According to Clinical / Microbiological assessment in WH-1 group, three patients improved while two did not, whereas in the fluconazole group all three patients showed improvement.

**Discussion**

Several studies have been carried out to examine the factors influencing, as well as treatment outcome of bacterial vaginosis in terms of efficacy and safety. National Health Examination Analytic and Reporting Guidelines, have been formulated to study the condition (2005). It is well established that vaginosis when left untreated increases the risk of pelvic inflammatory disease [PID] in turn leading to infertility, premature delivery, low birth weight, in addition to endometritis and cervical neoplasia as some of its long term complications. Data clearly suggests that bacterial vaginosis is an important predictor of adverse reproductive outcome and more complete dynamics connecting the sociodemographic characteristics is anticipated to create targeted interventions.

Although BV is a common condition with distressing symptoms a high proportion of women remain asymptomatic. Change in vaginal ecology resulting in overgrowth of Gardnerella vaginalis and anaerobes with replacement of normal vaginal flora [lactobacilli] seem to be primarily responsible for this condition with recurrence leading to adverse pregnancy outcomes. However, Clinical Guidelines from a review based on pooled data of several studies to examine value of screening and treatment of BV to reduce adverse pregnancy outcomes among asymptomatic women at various levels of risk for preterm delivery has turned out inconclusive.

In the present study since pregnancy was one of the exclusion criteria it is difficult to comment on safety as well as usefulness of WH-1 in pregnant women with vaginitis.

Table 3. Comparison of subjective improvement of symptoms of vaginitis / vaginosis in patients receiving WH-1 [Grp- A] vs FAS-3 kit [Grp B]

<table>
<thead>
<tr>
<th>Subjective improvement</th>
<th>Group A</th>
<th>Group B</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pre-treatment</td>
<td>$2.75 \pm 1.517$</td>
<td>$3.27 \pm 1.383$</td>
<td>$&gt; 0.315$</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Not Significant</td>
</tr>
<tr>
<td>Post-treatment</td>
<td>$1.10 \pm 1.210$</td>
<td>$0.7 \pm 0.0733$</td>
<td>$&gt; 0.214$</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Not Significant</td>
</tr>
<tr>
<td>Post treatment</td>
<td>$1.65 \pm 0.988$</td>
<td>$2.50 \pm 1.539$</td>
<td>$&lt; 0.044$</td>
</tr>
<tr>
<td>Between groups</td>
<td></td>
<td></td>
<td>Significant</td>
</tr>
</tbody>
</table>
alternative traditional remedies (96%) that are available and these were usually educated health conscious women.\(^{12}\) Few of the specific therapies tested included – lactobacilli recolonization by oral and vaginal administration, douching, boric acid, tea tree oil and garlic. While, review supports the benefits of these ingredients, it has also highlighted the adverse effects associated with each of them. Comparative studies with various douching formulations either have revealed increased prevalence of PID, risk of ectopic pregnancy, endometritis and salpingitis associated with this procedure or some are in-vitro to draw conclusive evidence on efficacy.\(^{13}\) Routine douching has been shown to double the risk of acquiring vaginitis. Further, boric acid treatment is reported to produce vaginal excoriation and use of topical tea tree oil or garlic is known to cause allergic reaction while oral garlic is said to produce heartburn and bloating.\(^{14}\) A trial with povidon-iodine as vaginal suppository for BV has shown some promising results.\(^{15}\) In addition the recommendations of previous review include - improvement in the study design, acceptable diagnostic criteria and examining validated outcome measures. The test preparation, WH-1 in the present study did not have any significant adverse effects. In this context the present study meets few of these recommendations such as study design, well defined assessment criteria, and outcome measures with reliable results, hence may be considered superior to earlier studies.

A preliminary study carried out elsewhere, with WH-1, revealed both symptomatic and clinical improvement in subjects with vaginitis of varied aetiology but this was not a comparative study. Also, objective assessment using microbiological parameters and statistical analysis was lacking in the previous study.\(^3\)

The present study was designed to evaluate systematically the efficacy and safety of test drug WH-1, in comparison with standard treatment regimen FAS-3 kit, in women suffering from mixed / non-specific vaginitis using subjective as well as objective criteria. The study also included assessment of lactobacilli count which was not a part of the earlier studies. The important efficacy parameters such as microbiological as well as clinical examination showed that WH-1 is equipotent to standard therapy. It is reported that the combination of antibacterial, antifungal and antiprotozoal agents in the modern therapy alter the normal vaginal flora leading to super infection and /or increasing chances of recurrence of infection. Interestingly, the present test preparation WH-1 showed absence of such associated complications. This finding appears to be unique to WH-1 and may be attributed to multiple mechanisms of actions of the various components of this formulation which are reported to reduce irritation, inflammation and secretion with added anti-septic activity.\(^3\)

In addition either maintenance or improvement in the lactobacilli count was seen with WH-1, in the present study. In contrast 2/20 patients on standard treatment had aggravation of symptoms and super infection with candida respectively while only 1/20, had aggravation with super infection. Study by Bluestein C (1991) suggests possibility of predicting the occurrence of antibiotic induced candidal vaginitis, this was not considered since the number of patients was small.\(^{16}\) Although, a long term follow up was not a part of the present study protocol, some patients were contacted telephonically at one to two months after completion of treatment with WH-1 and these showed subjective improvement compared to those who received the standard allopathic treatment, as far as relapse/recurrence was concerned. It is interesting to note that although the duration of treatment was ten days with WH-1 as against single dose treatment with standard FAS-3 kit, all patients except one demonstrated good compliance. This difference may be attributed to the fact that vaginitis/vaginosis produces disturbing symptoms hence the patients may not be averse to taking medication for longer duration.

Patients with candida infection also showed clinical and microbiological improvement with
Efficacy and safety of poly herbal formulation for vaginosis/vaginitis

WH-1, but the number of patients was small and so a statistical comparison to standard fluconazole therapy could not be performed.

To conclude, our study confirms the observations of the previous preliminary study reports.\(^3\) WH-1 was found to be equally efficacious as standard drugs of FAS-3 kit and fluconazole in mixed infection, nonspecific vaginitis as well as vaginal candidiasis. The numbers of adverse drug reactions [ADRs] were higher in standard modern treatment group.

A few of the positive findings of the present study with WH-1 were its global efficacy in the treatment of vaginitis/vaginosis caused by multiple pathogens, minor incidence of ADRs, a noticeable increment in lactobacilli count, and 100% medication compliance with WH-1 despite longer duration of treatment. In addition there was no biochemical evidence of ADRs in patients who received test drug. Therefore WH-1 may be considered to be an effective and safe option for treatment of vaginitis/vaginosis.

However, studies in larger number of patients with longer duration of treatment as well as follow up are necessary to confirm benefits of WH-1, in preventing recurrence of BV, and in the treatment of VC or TMV as a complication of underlying diabetes mellitus, STDs or AIDs.

**Key Messages**

1. The safety and efficacy outcome measures with poly herbal formulation WH-1 in treatment of vaginitis/vaginosis revealed comparable efficacy profile with lesser adverse effects compared to modern medication.
2. Results are suggestive of WH-1 as one of the safer and alternative options in the treatment of vaginosis/vaginitis.

**Acknowledgement**

The authors wish to gratefully acknowledge Dr. V. R. Bapat, for free supply of WH-1 capsules and for financial assistance to conduct this study and Dr. S.C. Savitri, District AYUSH Officer, Govt. of Karnataka for her scientific input.

**References**

7. Pirotta MV, Garland SM: Genital candida species detected in samples from women in Melbourne Australia before and after treatment with antibiotics. *J Clin Microbial* 2006; 44(9):3213-7
8. Centre for disease control and prevention (CDC), national centre for health statistics (NCHS), national health and nutrition examination analytic and reporting guidelines Yattsville (MD):U.S. department of health and human services, centres for disease control and prevention 2005


Address for correspondence: Dr.(Mrs.) Chanda Kulkarni, MD; Ph.D; FSASMS; Professor & Head, Division of Clinical Pharmacology, St. John’s Medical College, Bangalore - 560034 Karnataka (India).
Email: drchandakulkarni@gmail.com
EVALUATION OF BHAVANA SAMSKARA WITH REFERENCE TO PIPPALI CHURNA AND CHAUSASTA PIPPALI

P. B. PIMPALGAONKAR1 ASHLESHA RAUT2 AND R. S. SAWANT3

Department of Rasashastra,1 Govt. Ayurved College, Osmanabad, Maharashtra (India)
Nutritional Consultant and Health Educator, 2 B.C., Integrative Medicine, Director, Main Street Yoga, Bloomington, IL
Department of Rasashastra & Bhaishajya Kalpana,3 Govt. Ayurved College, Vazirabad road, Nanded, Maharashtra (India)

Abstract: Chausasta Pippali is commonly used medicine in respiratory disorders. It is an herbal preparation. There are different traditions of preparing Chausasta Pippali, one is triturate Pippali churna with Pippali decoction for 64 prahar (prahar = 3hrs) and another is triturate Pippali churna with Pippali decoction for 64 times one after another. In current study healthy Pippali fruits were sun dried and were finely powdered. This powder was processed in the decoction of Pippali for 64 times. For each Bhavana fresh decoction was added to the preparation. We adopted above method because primarily we intended to study Bhavana Samskara and there is tradition to follow this method. An attempt was made to find difference between Pippali churna and Chausasta Pippali. Samples were analyzed after 0, 32nd, 64th Bhavana on organoleptic, physico-chemical characters and photochemical behavior of these compounds. We observed interesting qualitative and quantitative differences between ‘Pippali Churna’ and ‘Chausasta Pippali’.

Keywords: Bhavana Samskara, Pippali, Chausasta Pippali, photochemical character, HPTLC.

Introduction

Bhavana Samskara is a type of trituration process. In this process a dry medicinal substance is poured with the wet medicinal substance (most often plant juice/decoction) and then macerated together till entire amalgam becomes dry again.1 The Bhavana Samskara may have the following main implications.

· To enhance potency of drug.
· To reduce the dosage for clinical efficiency.
· To suppress the toxicity.
· To improve the bioavailability and bioacceptability.

Bhavana Samskara is one of the most commonly carried out pharmaceutical processing which has multidimensional clinical implications and hence it was decided to study this process.

Here an attempt was made to study some aspect of chemical analysis to evaluate Bhavana Samskara by studying the difference between Pippali churna and Chausasta Pippali.

Objectives

· To understand the concept of Bhavana Sanskara by studying the difference between Pippali Churna and Chausasta Pippali.
· To prepare and Standardize Chausasta Pippali.
· To observe for qualitative and quantitative differences between Pippali churna and Chausasta Pippali.
· To compare our laboratory prepared Chausasta Pippali with available marketed samples by HPTLC analysis.

Chemical Composition of Pippali

The major alkaloid piperine (C17H19NO3) and sesamine (C20H18O6) have been isolated from fruits and stem. Other than piperine (4 to 5 %) and pipilartine two new liquid alkaloids one of which designated as alkaloid ‘A’ is closely related to pellitorine producing marked salivation, numbness and tingling sensation of mucus.
membrane of mouth. Sample of dried fruit of Piper longum on steam distillation gave 0.7 % of an essential oil with spicy odour.²

**Method of Preparation of Chausasta Pippali**

There are two types of Chausasta Pippali-
1. Chausasta Pippali
2. Chausasta prahari Pippali.

On screening the literature for Chausasta pippali it was revealed that there is no pertinent reference of this formulation in earlier Ayurvedic treaties. The description of this formulation is documented by recent Ayurvedic Scholars viz. Vd. S. P. Khare in Aushadhi Nirmana, Vd. G. A. Phadake in Dravyagunashastram and Vd. Ramaraksha Pathak in Ayurved Sarasamgraha.

It can be inferred from these description that Chausasta Pippali is prepared in different ways.

1. **With Bhavana Dravya**
   I. Fresh powder of Piper Longum fruits is to be impregnated by decoction of Piper longum and then triturated in pestle and mortar for 64 prahara (approx. 192 hours).³
   II. Fresh powder of Piper Longum fruits is to be impregnated by decoction of Piper longum and then triturated till it becomes dry again. This is considered as one bhavana. Such a bhavana repeated 64 times. Every time fresh decoction is added in earlier prepared bhavit Pippali.⁴

2. **With Substituted Bhavana Dravya**
   Here the procedure is same either of above only difference is the decoction or liquid used for process of trituration is substituted with other liquid or decoction of a substance having similar properties and potency that of Piper longum.

3. **Without Bhavya Dravya**
   In this procedure fresh fruits of Piper longum is powdered and sieved through cloth and is then ground in mortar and pestle for 8 days and nights. No liquid or decoction is used for trituration. Some vaidyas add gold foils in this preparation.

**Material and Methods**

Dry Pippali fruits were procured from market and authenticated by following pharmacognostical methods like:-

1. Macroscopy
2. Microscopy

**Preparation of Pippali Churna for Chausasta Pippali**

The completely dried fruits free from fungus and possible to break in pieces were selected and powdered in grinder until the powder passed through sieve no 120.

**Kwath (Decoction) Preparation for procedure of Bhavana**

The method described in Rasendra sarasangraha was followed for preparation of Kwath.

Dry Pippali fruits (1 part) + water (8 parts) = Decoction (1/8th of water used)

Then decoction was filtered from cloth and the filtrate was used to add to each bhavana. For each bhavana freshly prepared decoction of Piper longum fruits were used.

**Preparation of Chausasta Pippali**

As mentioned above there are various methods of preparation of Chausasta pippali, concern to our aim 1/1 method was adopted for planned study.

2 kg powder of pippali was taken initially for the preparation of Chausasta pippali. Then the pippali decoction was poured into it. When it became adequately wet the trituration was carried out in wet grinder.

**The Criteria for Addition of Amount of Decoction Required for Bhavana was as Follows**

1. The powder should get thoroughly soaked in the decoction.
2. The preparation should have muddy appearance but it should not be too batter.
Physico-chemical characters and photochemical behavior of compounds present in Pippali & Chausastā

Pippali Churna

3. While rotating in the grinder the mixture of Pippali churna and decoction should get shiny look.
4. The pesani should move smoothly in the grinder.

The process of trituration was carried out till mixture becomes dry. The time of trituration was not fixed because it was varying from bhavana to bhavana.

The atmospheric humidity, the changing consistency of prepared amalgam, etc are the factors which influenced varying time during Bhavana Samskara.

The Completion of Bhavana was Determined on Following Parameters
1. The shiny appearance of the above muddy material disappears and it becomes dry.
2. If the material is pressed between two fingers it does not stuck up to the finger surfaces.
3. The material become so dry a pill of it can made easily.
4. The final product becomes soft.
5. The Muller should not move freely in the grinder and while lifting it up much more pressure apply for it.

Following the above criteria each bhavana samskara was given to pippali churna. The samples for analysis taken out after 0, 32nd, 64th bhavana. At the end of 64th bhavana the final product yield was 3 kg.

Analysis of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

- **PL₀**: Pippali Churna at 0 Bhavana
- **PL₃₂**: Pippali Churna at 32nd Bhavana
- **PL₆₄**: Pippali Churna at 64th Bhavana

**Results**

| Table 1. Organoleptic Evaluation of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath |
|---|---|---|
| **Organo-leptic test** | **PL₀** | **PL₃₂** | **PL₆₄** |
| Shabda (Sound) | NIL | NIL | NIL |
| Sparsha (Texture) | Mrudu, Laghu | Mrudu, Khara, Laghu | Mrudu, Snigdha, Laghu |
| Rupa (Colour) | Greenish | Brownish black | Black |
| Rasa (taste) (Katu) | +++ | ++ | + |
| Gandha (Smell) (Ugra) | +++ | + | + |

**Table 1a. Determination of Pungency of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath**

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Mean of taster’s sensitivity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Std</td>
<td>5.0</td>
</tr>
<tr>
<td>PL₀</td>
<td>5.2</td>
</tr>
<tr>
<td>PL₃₂</td>
<td>6.6</td>
</tr>
<tr>
<td>PL₆₄</td>
<td>7.6</td>
</tr>
</tbody>
</table>

**Graph 1. Determination of Pungency of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath**

Data Interpretation of HPTLC Analysis

A) Piperine Quantification of Lab Prepared Samples

Piperine being a major alkaloid of Piper longum; its quantification was the first step to
Table 2. Determination of pH of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>PL0</td>
<td>5.0</td>
</tr>
<tr>
<td>PL32</td>
<td>4.5</td>
</tr>
<tr>
<td>PL64</td>
<td>4.6</td>
</tr>
<tr>
<td>Kwath</td>
<td>5.22</td>
</tr>
</tbody>
</table>

Graph 2. Determination of pH of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

Table 3. Determination Moisture Content of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Moisture content (%w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PL0</td>
<td>6.15</td>
</tr>
<tr>
<td>PL32</td>
<td>8.14</td>
</tr>
<tr>
<td>PL64</td>
<td>9.51</td>
</tr>
</tbody>
</table>

Graph 3. Determination Moisture Content of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

Table 4. Determination of Total Solids of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

<table>
<thead>
<tr>
<th>Sample No.</th>
<th>Total Solids</th>
</tr>
</thead>
<tbody>
<tr>
<td>PL0</td>
<td>23.90%</td>
</tr>
<tr>
<td>PL32</td>
<td>36.55%</td>
</tr>
<tr>
<td>PL64</td>
<td>47.50%</td>
</tr>
</tbody>
</table>

Graph 4. Determination of Total Solids of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

HPTLC analysis. After confirmation, the quantification of Piperine content in each sample was carried out. The peak correlating the same Rf of 0.55 in each sample was calculated by height and by area to quantify the Piperine content.

Graph 5. Determination Ash value of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath
Physico-chemical characters and photochemical behavior of compounds present in Pippali & Chausasta

Table 5. Determination of Ash Value of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Sample</th>
<th>Residue on Ignition (%w/w)</th>
<th>Total Ash (%w/w)</th>
<th>Water soluble Ash (%w/w)</th>
<th>Acid insoluble Ash (%w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>PL₀</td>
<td>5.32</td>
<td>7.2</td>
<td>3.25</td>
<td>14.22</td>
</tr>
<tr>
<td>2</td>
<td>PL₃₂</td>
<td>8.58</td>
<td>10.59</td>
<td>5.10</td>
<td>10.45</td>
</tr>
<tr>
<td>3</td>
<td>PL₆₄</td>
<td>9.81</td>
<td>12.56</td>
<td>5.20</td>
<td>9.47</td>
</tr>
</tbody>
</table>

Table 6. Determination of Extractive Values of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Water (%w/w)</th>
<th>Ethanol 90% (%w/w)</th>
<th>Benzene (%w/w)</th>
<th>Petroleum ether (%w/w)</th>
<th>Chloroform (%w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PL₀</td>
<td>23.99</td>
<td>13.51</td>
<td>6.64</td>
<td>4.95</td>
<td>6.57</td>
</tr>
<tr>
<td>PL₃₂</td>
<td>36.51</td>
<td>17.62</td>
<td>6.22</td>
<td>1.82</td>
<td>5.02</td>
</tr>
<tr>
<td>PL₆₄</td>
<td>47.57</td>
<td>24.81</td>
<td>6.08</td>
<td>0.82</td>
<td>2.2</td>
</tr>
</tbody>
</table>

Table 7. Phyto-chemical Constituents’ Estimation of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath

<table>
<thead>
<tr>
<th>Sample</th>
<th>Total Alkaloids (%w/w)</th>
<th>Total Glycosides (%w/w)</th>
<th>Total volatile oil (%w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PL₀</td>
<td>1.51</td>
<td>75</td>
<td>negligible</td>
</tr>
<tr>
<td>PL₃₂</td>
<td>1.44</td>
<td>82</td>
<td>negligible</td>
</tr>
<tr>
<td>PL₆₄</td>
<td>1.31</td>
<td>86.55</td>
<td>negligible</td>
</tr>
</tbody>
</table>

This shows a gradual reduction in Piperine content. The Bhavana process might be the causative factor of above results.

B) Piperine Quantification of Ethanolic Extract of Lab Prepared Samples

To evaluate Piperine content and recheck the previous observations the hot ethanolic soxhlet were analyzed on HPTLC. The calculated data is presented in following table.

<table>
<thead>
<tr>
<th>Sample (1mg extract)</th>
<th>Amount of Piperine (in ng)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PL₀</td>
<td>188.29</td>
</tr>
<tr>
<td>PL₃₂</td>
<td>49.971</td>
</tr>
<tr>
<td>PL₆₄</td>
<td>22.999</td>
</tr>
</tbody>
</table>

Graph 6. Phyto-chemical Constituents’ Estimation of Laboratory Prepared Pippali Churna, Chausasta Pippali and Pippali Fruit Kwath
Pimpalgaonkar, Raut and Sawant

were having more acidic ph than abavit sample. This acidic ph might be indicator of increased tikshnatwa after bhavana samskara. The moisture content is one of the physical constants to check adulteration. The observation found in present study may be helpful for quality control.

Ash value represent the inorganic material present in drug. The ash value (total ash, water soluble ash, residue on ignition) observation s of this project shows gradual increase. This indicates inorganic salts for eg. Ca, K, Na, etc. might have increased in bhavit pippali samples.

The results of total alkaloid percentage were gradually reduced in PL 0, PL 32, PL 64 samples which was also confirms on HPTLC showed gradual reduction in subsequent samples. Viz. namely PL 0, PL 32, PL 64. One need not infer that reduction in piperine in reduced potency of chausasta pippali. To evaluate the role of bhavana samskara in chaussasta pippali for conformation of drug potenciation experimental and clinical trial would be more productive.

Conclusion

Notwithstanding to the verse “Bhuyashcha Yesham Baladhanam” piperine content gradually reduced in subsequent samples on HPTLC analysis. It is traditionally belived; clinically experienced and widely excepted that repeatedly processed pippali by its own decoction is more potent than pippali churna. Hence the message that clinical efficacy of an ayurvedic formulation may not correlate with its single active principle.

References

1. K. Shatri: Rasatarangini, Motilal Banarasidas publication, New Delhi, India 2004; pp.21
4. S. P Khare: Aushadhi Nirman, Neelakantha publication, Pune, India 1975; pp.52

Address for correspondence: Dr. Ranjeet S. Sawant, Assistant Professor, Department of Rasashastra & Bhaishajya Kalpana, Government Ayurved College, Vazirabad Road, Nanded, Maharashtra – 431601 (India).
E-mail: drranjeet.sawant@gmail.com
ANTIMICROBIAL ACTIVITY OF AN INDIAN MISTLETOE, THE HEMIPARASITE DENDROPHTHOE FALCATA L. F. (LORANTHACEAE)

S. H. PATIL,1* V.S.PATIL,3 R. B.JADHAV,1 G. S. TALELE,2 S. J. SURANA1

R.C. Patel College of Pharmacy, 1,1* Near Karwand Naka, Shirpur, Dist. Dhule - 425405 (India)
N.G.S.P.M’s College of Pharmacy, 2 Trimbakeshwar, Nasik, M.S. - 422213 (India)
H.R. Patel College of Pharmacy, 3 Near Karwand Naka, Shirpur, Dist. Dhule - 425405 (India)

Abstract: Dendrophthoe falcata linn. is reported to contain biologically active substances such as flavonoids, β-sitosterol, β-amirin, oleonolic acid, chlorophyll, steroids and terpenoids. The present study was undertaken to evaluate the antimicrobial and antifungal potential of extracts of this plant. The extracts of Dendrophthoe falcata linn. leaves, showed significant antimicrobial activity against Gram positive and Gram-negative test organisms. Dendrophthoe falcata linn. is an hemiparasite plant belonging to Loranthaceae, Synonyms are Loranthus longiflorous also known as banda, vandal vrikshabhaksha and bandgul. This parasitic plant is one of the most widespread plants in the forests of India, usually occurring in deciduous forests of Western Ghats of India.

Keywords: Dendrophthoe falcata linn., leaves, Cup and Plate method, Antibacterial, Antifungal

Introduction

Even though pharmaceutical industries have produced a number of new antibiotics in the last three decades, resistance to these drugs by microorganisms has increased. In general, bacteria have the genetic ability to transmit and acquire resistance to drugs, which are utilized as therapeutic agents. Such a fact is cause for concern, because of the number of patients in hospitals who have suppressed immunity, and due to new bacterial strains, which are multi-resistant. Consequently, new infections can occur in hospitals resulting in high mortality. This fact has also been verified in other clinics around all over world.

The problem of microbial resistance is growing and the outlook for the use of antimicrobial drugs in the future is still uncertain. Therefore, actions must be taken to reduce this problem, for example, to control the use of antibiotic, develop research to better understand the genetic mechanisms of resistance, and to continue studies to develop new drugs, either synthetic or natural. The ultimate goal is to offer appropriate and efficient antimicrobial drugs to the patient.

For a long period of time, plants have been a valuable source of natural products for maintaining human health, especially in the last decade, with more intensive studies for natural therapies. The use of plant compounds for pharmaceutical purposes has gradually increased in Brazil. According to World Health Organization medicinal plants would be the best source to obtain a variety of drugs. About 80% of individuals from developed countries use traditional medicine, which has compounds derived from medicinal plants. Therefore, such plants should be investigated to better understand their properties, safety and efficiency.

Even though some literature on the medicinal properties of Loranthaceae plants is found, but information on genus Dendrophthoe is rare.1-7 Traditionally, the whole plant is used in indigenous system of medicine as aphrodisiac, astringent, diuretic, narcotic and in treatment of pulmonary asthma, menstrual disorders, and as antiviral herbal drug.8-13
Materials and methods

Plant material

The leaves of *Dendrophthoe falcata* parasitic on *Mangifera indica* (Anacardiaceae) were collected from Western Ghat region of Maharashtra (India) in February 2005. The plant specimen was authenticated from Botanical Survey of India, Pune (Voucher specimen no. PSH-1).

Reagents and Materials

The media used for antimicrobial testing was Muller Hinton agar and the media for antifungal testing was Potato dextrose agar. The media were purchased from Hi-media labs, India. Other solvents used in the test were of analytical reagent grade purchased from Rankem, India. The various strains of microorganisms were obtained from NCIM, Pune, India.

Preparation of extracts

The leaves were shade dried and coarsely powdered. The powdered leaves were extracted by soxhlet extraction by successive extraction method and the extractive yield for each of the extracts was calculated. Petroleum ether, Benzene, Chloroform, Acetone, Methanol, Ethanol, Ethylacetate, Butanol, and Aqueous extracts were prepared. The prepared extracts were then subjected to preliminary phytochemical investigations to estimate the presence of various phytoconstituents. The image of *Dendrophthoe falcata* L. f. is depicted in Figure 1.

Evaluation of Anti-microbial activity

The size of Inoculums for Anti-microbial tests were 1 X 10^8 bacteria per ml and the concentration of extracts used in the assays was 5mg/ml.

Antimicrobial assessment of the extracts using Agar Well Diffusion method

Preparation of inoculums

About 20ml of Muller Hinton agar medium for bacteria and Czapek Dox for fungi were allowed to set in empty sterile Petri plate. About 0.1ml of fungal inoculums was made in Petri plates preset for spore count, cell density and bacterial inoculums in respective medias. The cups of 6 mm diameters were bored on the agar media and were then filled with 0.5ml of plant extracts. The plates were then incubated at 30°C for 48 hours and 37°C for 24 hours respectively for fungi and bacteria. The zone of inhibition produced was read after incubation.

Culture medium

All the solutions used for testing were prepared in DMSO as a solvent.

Microorganisms

Antibacterial activity of extract was tested in vitro against *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Salmonella typhi* (Gram-negative) and *Bacillus subtilis* (Gram-positive), *Shigella sonnei*.

Antimicrobial Agent

The standard compound (Gentamycin) was dissolved in dimethyl sulfoxide to give a concentration of 5x10^-2 mm/ml.
The concentrations of various extracts were made to obtain a concentration of 1mg/ml. Cup plate agar diffusion method was used to determine the zone of inhibition of various extracts.
**Preparation of test solution**

The concentration of various extracts mentioned above were made in Dimethyl sulfoxide (DMSO) to give concentration of 200 mg/ml.

**Determination of zone of inhibition by cup plate method**

The antibacterial activity of the extracts was performed using Agar cup-plate method. About 20ml of sterile nutrient agar medium was poured into sterile petri-dishes and allowed to solidify. The petri dishes were incubated at 37°C for 24 hours to check for sterility. The medium was seeded with the organisms by pour plate method using sterile top agar (4 ml) contained 1 ml culture. Bores were made on the medium using sterile borer. Dried extracts were dissolved in Dimethyl sulfoxide (DMSO) to obtain different concentrations and sterilized by filtration through a Whatman filter paper no. 1, and 0.1 ml of the different concentrations of extracts were added to the respective bores. The plates were incubated overnight at 37°C with appropriate positive and negative controls.

The petri-dishes were kept in refrigerator at 4°C for 30 min for diffusion. After diffusion the petri-dishes were incubated at 37°C for 24 hours and zone of inhibition were observed and measured. Dimethyl sulfoxide was used as the control.

**Determination of anti-fungal activity**

Antifungal activity of extracts against was assessed with _Candida albicans_ (MTCC 227) and _Aspergillus niger_ (NCIM 545). The evaluation was performed similar to antibacterial activity by use of potato dextrose agar (PDA) as media for assay. The fungal sensitivity of extracts was evaluated for 7 days. Amphotericin B (20 mcg/ml) was used as a standard for this activity.

**Results and Discussion**

**Extraction of plant material**

Various extracts of plant material were prepared by soxhlet extraction and the extractive yield were calculated. The extractive yields of Petroleum ether, Benzene, Chloroform, Acetone, Methanol, Ethanol, Ethylacetate, Butanol and Aqueous are 3.99%, 1.26%, 2.10%, 2.23%, 4.16%, 2.21%, 3.2%, 1.25% and 9.4% respectively.

**Preliminary phytochemical investigation**

The preliminary phytochemical investigations were carried out to reveal the presence of various phytoconstituents. Petroleum ether and CHCl₃ extract gave positive tests for steroids, EtOAc and MeOH extracts revealed presence of glycosides and flavonoids.

**Antibacterial and antifungal activity**

The antibacterial studies confirmed that the extracts had zone of inhibition, but the MIC of extracts ensured no prominent action on the tested bacterial strains. The antifungal studies confirmed that the extracts had a effective zone of inhibition against the tested organisms. The results of antibacterial and antifungal extracts are shown in Table 1-4. In future one can formulate the active constituents into a topical dosage form with antimicrobial and antifungal effect.

**References**

Antimicrobial activity of *Dendrophthoe falcata* L.f. (Loranthaceae)

6. Z.P. Cerda, T. Fernandez and *et al.*: Ligaria cuneifolia flavonoid fractions modulate cell growth of normal lymphocytes and tumor cells as well as multidrug resistant cells. *Immunobiology* 2005; 209(10):737-749


10. N.A. Aleykuty, K.K. Srinivasan, and *et al.*: Diuretic and antilithiatic activity of *Dendrophthoe falcata*, *Fitoterapia* 1993; pp.64,4,325-331


---

**Address for correspondence:** Satish H. Patil, Research Scholar, R.C. Patel College of Pharmacy, Shirpur, Dist. Dhule, M.S.P.C - 425405 (India). E-mail: patilsatish181980@rediffmail.com
A CLINICAL STUDY ON THE ROLE OF GOKSHURADI YOGA IN THE MANAGEMENT OF KLAIBYA (ERECTILE DYSFUNCTION)

MANJUNATHA T SASANOOR,1 DHANARAJ NAGAR,2 HETAL DAVE3 AND BALDEV KUMAR4

Department. of P. G. Studies in Maulik Siddhant and Samhita,1,2,4
Department of P. G. Studies in Prasuti and Sri Roga,3
National Institute of Ayurveda, Amer Road, Jaipur, Rajasthan - 302002 (India)

Abstract: The rich heritage of Indian culture considered ‘kāma’ as one among the purushārthas, the objectives of life. The concept of ‘kāma’ reveals that the recreational aspects like enjoyment of pleasure is equally important to its procreational aspects. An apparent disparity between the subjective sense of pleasure and objective performance was always present. The present media culture additionally impacts unsatisfied inquisitiveness lead them to various misconceptions and sexual dysfunctions. The male sexual dysfunction includes all sort of disturbances of coital performance and sexual congress in male. Among the various phases of sexual response the most essential is the achieving of normal erection with sufficient rigidity for penetrative intercourse, the absence of which ends into failure and dissatisfaction. This condition has been elaborately described as ‘Klaibya’ in Ayurvedic classics and ‘Erectile dysfunction’ in modern texts. The disease klaibya is a multifactorial condition, mainly involving Bahu Dosavastha as a whole and Sukraksaya in specific, Mano Dosa, and Sukravaha Srot Dusti. Considering the grave nature of the disease though it does not reduces the life expectancy, it has been selected for the present study to find out a better cure.

Materials and Methods: 30 diagnosed patients of, aged between 20 – 50 years attending Opd of N.I.A Jaipur, 30 patients of, aged between 20 – 50 years, fulfilling the clinical criteria for diagnosis of Klaibya (Erectile dysfunction) were randomly selected irrespective of their caste, and religion from the OPD & IPD of Maulik Siddhant department of National Institute of Ayurveda Jaipur. were randomly selected and assigned into two groups. In Group A Gokshuradi yoga in the dose of 5gm/twice daily with water, for 45 days. and in Group B – Patients of this group received placebo capsule containing soybean powder (500 mg) in the dose of 2 capsule /twice daily with water, for 45 days. Follow up was done for 2 month. Assessment done on the basis of self scoring symptoms / subjective and objective parameters of Klaibya (Erectile Dysfunction). The study clearly showed that there is an improvement in both group with added effect in group A Patients of Klaibya (Erectile Dysfunction)

Keywords: Purushārthas, ‘Klaibya’, Erectile Dysfunction, Bahu Dosavastha.

Introduction
The disease klaibya is a multifactorial condition, mainly involving Bahu Dosavastha as a whole and Sukraksaya in specific, Mano Dosa, and Sukravaha Srot Dusti. It is commonly observed in the society, owing to the feeling of inadequacy less commonly reported, even than Master and Johnson reported a fear of impotence in all men above 40 years. Considering the grave nature of the disease though it does not reduces the life expectancy, it has been selected for the present study to find out a better cure.

Vajikarana has been described specially to improve the sexual health to enhance the status of Sukra and to please the mind. The amount of sexual dysfunction prevalent in the society is difficult to estimate if not impossible. About half of the married couples experience sexual difficulties in one or the other way at various stages of their married life (MC carthy et al. says that, The percentage of all men treated for sexual disorders who have impotence (erectile dysfunction) as the chief complaint ranges from 35 to 50 percent.

1. P. G final scholar 3. Lecturer 4. Associate Professor at NIA, Jaipur
2. Presently Lecturer at Shri Ganganagar College of Ayurveda, Science & Hospital, Ganganagar, Rajasthan
According to Acharya Charak Klaibya is defined as inability to perform sexual act\(^1\) and in sutra sthana 28th chapter, he says klaibya means lack of penile erection.\(^2\) According to Acharya Dalhana, klaibya means impotent.\(^3\) According to Acharya Vagbhata, it means inability to perform sexual intercourse with female partner,\(^4\) According to Astanga Sangraha, it means lack of penile rigidity,\(^5\) and According to Yoga Ratnakara, it means incapability of a person to perform sexual act and its form is Klaibya.\(^6,7\)

**Aim and objects**

1) To study the patients of male sexual dysfunction in general and the erectile dysfunction in particular from Ayurvedic point of view and its correlation with various types of Klaibya.

2) To study the efficacy of the gokshuradi yoga administered orally in the management of Klaibya.

3) To compare the effects of gokshuradi yoga With Placebo Drug.

**Materials and methods**

For the present study, 30 patients fulfilling the clinical criteria for diagnosis of Klaibya (Erectile dysfunction) were randomly selected irrespective of their caste, and religion from the OPD & IPD of Maulik Siddhant department of National Institute of Ayurveda Jaipur.

**Plan of the study:**

1. Patients were selected randomly. It is a comparative study with Pre-test – post-test design.
2. The selected patients were assigned into 2 groups, each consisting of 15 patients.
3. Group A: Patients received test drug in the dose of 5 gm/twice daily with water, for 45 days.
4. Group B: Patients of this group received placebo capsule containing soybean powder (500 mg) in the dose of 2 capsule /twice daily with water, for 45 days.

**Inclusion criteria**

The following was the main clinical criteria for the selection of the patients:

1) All Married men (age 20-50 years)
2) Incapability to perform sexual act.
3) Premature Ejaculation
4) Flaccidity of the penis even after psychic or physical stimulation

**Exclusion criteria**

Patients suffering from mental retardation, Unmarried persons (more than 50 years of age), congenital anomalies, infectious diseases of brain, vascular causes, toxic causes, metabolic causes of seizures were excluded from the study.

**Assessment criteria**

**Subjective criteria**

1. Sexual desire,
2. Breathlessness during coitus,
3. Excessive perspiration during coitus,
4. Premature ejaculation,
5. Erectile dysfunction,
6. Pain during ejaculation.

**Objective criteria**

**Investigations**

Routine haematological biochemical and urine investigations like Hb, Tc, De, ESR, Random Blood Sugar and Serum Cholesterol were carried to rule out other pathologies before starting the treatment, and after the completion of treatment to assess the overall effect of the therapy.

**Semen examinations**

To see the effect of the drug on the seminal parameters, the semen examination was done. The samples were collected by masturbation. The semen analysis was carried out by physical, chemical and microscopic examination to assess it qualitatively and quantitatively both, before and after the treatment in the all patients.

**Preparation of medicine**

The drug Kapikacchu was purified by boiling with milk and then dried and powdered, after that this was mixed with other drugs powder which are taken in equal quantity and prepared 150 gms pack.
### 1. Sexual desire

<table>
<thead>
<tr>
<th>Sexual desire</th>
<th>Grading</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal desire</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lack of desire</td>
<td></td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Desire only on demand of partner</td>
<td></td>
<td>2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No desire at all</td>
<td></td>
<td>3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### 2. Breathlessness

<table>
<thead>
<tr>
<th>Breathlessness</th>
<th>Grading</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Breathlessness</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mild Breathlessness which does not disturb the act</td>
<td></td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moderate Breathlessness sometimes disturb the act</td>
<td></td>
<td>2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Severe Breathlessness which hamper every act</td>
<td></td>
<td>3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### 3. Excessive perspiration

<table>
<thead>
<tr>
<th>Excessive perspiration</th>
<th>Grading</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>No perspiration</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mild perspiration</td>
<td></td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moderate perspiration</td>
<td></td>
<td>2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Severe perspiration which disturbs the act</td>
<td></td>
<td>3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### 4. Premature Ejaculation

<table>
<thead>
<tr>
<th>Premature Ejaculation</th>
<th>Grading</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>Every time can control ejaculation till both get satisfied</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Can control ejaculation till He get satisfaction</td>
<td></td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ejaculation before penetration</td>
<td></td>
<td>2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ejaculation during foreplay</td>
<td></td>
<td>3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### 5. Erectile Dysfunction

<table>
<thead>
<tr>
<th>Erectile Dysfunction</th>
<th>Grading</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proper stiffness to maintain erection and to continue the sexual intercourse till last.</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Some loss of stiffness but can maintain the erection and continue the act till last</td>
<td></td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Some loss of stiffness, able to maintain erection, but unable to continue act till last</td>
<td></td>
<td>2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total loss of stiffness and unable to initiate the sexual intercourse</td>
<td></td>
<td>3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### 6. Pain during ejaculation

<table>
<thead>
<tr>
<th>Pain during ejaculation</th>
<th>Grading</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>No Pain during ejaculation</td>
<td></td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Occasionally Mild Pain during ejaculation</td>
<td></td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Usually Moderate Pain during ejaculation</td>
<td></td>
<td>2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Severe Pain during ejaculation</td>
<td></td>
<td>3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

#### A) Volume of semen (in ml)

<table>
<thead>
<tr>
<th>Volume of semen</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt; 3-0 ml</td>
<td></td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>upto 2&amp;3 ml</td>
<td></td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>upto 1&amp;2 ml</td>
<td></td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>&lt;1 ml</td>
<td></td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>0 ml</td>
<td></td>
<td>4</td>
<td></td>
</tr>
</tbody>
</table>

#### B) Sperm Count Million/ml

<table>
<thead>
<tr>
<th>Sperm Count</th>
<th>Score</th>
<th>B.T</th>
<th>A.T</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt;40 million/ml</td>
<td></td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>30&amp;39 million/ml</td>
<td></td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>20&amp;29 million/ml</td>
<td></td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>10&amp;19 million/ml</td>
<td></td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>0&amp;9 million/ml</td>
<td></td>
<td>4</td>
<td></td>
</tr>
</tbody>
</table>
In the present clinical study Group A was administered with gokshuradi yoga powder with a dosage of 5 gms bd with water, Group B was administered with 2 placebo capsules bd with water.

**Review of the different components of the formulations.**

**Gokshura**

Oral administration of 100 mg/kg of test drug has proven anabolic effect as evidenced by body weight gain in the body and reproductive organs. Improvement in sexual behavior of male rats was characterized by increased amount and intromission frequency. Penile erection index (PEI) was also considerably enhanced without any noticeable toxicity, and the testosterone level and sperm count also significantly increased, and the results are comparable to that of standard drug, sildenafil citrate. Findings of the present study validate the traditional use of *Tribulus terrestris* for its role in enhancing sexual behavior and potential to be used in the treatment of Erectile Dysfunction (ED).

**Kokilaksha**: A long history of traditional use in Brazil and India as an aphrodisiac. Clinical studies in India have validated that the plant does indeed have aphrodisiac activity. It also has reported with anabolic and growth hormone stimulant properties. The anabolic effect of the seed is due to its ability to increase testosterone. In 2002, a U.S. patent was filed on the use of velvet bean to stimulate the release of growth hormone in humans. Research cited in the patent indicated that the high levels of L-dopa in mucuna seed were converted to dopamine which stimulated the release of growth hormone by the pituitary gland. L-dopa and dopamine are also effective inhibitors of prolactin. Prolactin is a hormone released by the pituitary gland; increased levels are considered to cause erection failure in males. In one study, oral intake of the seeds in 56 human males was able to improve erection, duration of coitus, and post-coital satisfaction after only four weeks of treatment. The seed also has documented fertility promoting and sperm producing effects in human males (being able to improve sperm count and motility).

**Black gram**

Black gram is black colored bean of a plant. It is nutritious bean commonly cooked for healthy diet.

**Nutrition Facts and Information about Black Gram**: Black gram is rich in potassium, phosphorus and calcium with good amount of sodium. It also has small amount of iron in it.

**Vitamin Content of Black Gram**: Black gram is rich in Vitamin A, B1 and B3 and has small amount of thiamine, riboflavin, niacin and Vitamin C in it.

**Calorie Content of Black Gram**: 100g of Black Gram has 347 calories. Calories from fat are 5.

**Health Benefits of Black Gram**: Black Gram cures diabetes, sexual dysfunction, nervous disorders, hair disorders, digestive system disorders and rheumatic afflications.

**Kapikacchu**: A long history of traditional use in Brazil and India as an aphrodisiac. Clinical studies in India have validated that the plant does indeed have aphrodisiac activity. It also has reported with anabolic and growth hormone stimulant properties. The anabolic effect of the seed is due to its ability to increase testosterone. In 2002, a U.S. patent was filed on the use of velvet bean to stimulate the release of growth hormone in humans. Research cited in the patent indicated that the high levels of L-dopa in mucuna seed were converted to dopamine which stimulated the release of growth hormone by the pituitary gland. L-dopa and dopamine are also effective inhibitors of prolactin. Prolactin is a hormone released by the pituitary gland; increased levels are considered to cause erection failure in males. In one study, oral intake of the seeds in 56 human males was able to improve erection, duration of coitus, and post-coital satisfaction after only four weeks of treatment. The seed also has documented fertility promoting and sperm producing effects in human males (being able to improve sperm count and motility).

**Drugs of the Gokshuradi Yoga**

<table>
<thead>
<tr>
<th>Sl. No.</th>
<th>Drugs</th>
<th>Latin names</th>
<th>Part used</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Gokshura</td>
<td>Tribulus terrestris</td>
<td>Fruit</td>
</tr>
<tr>
<td>2.</td>
<td>kokilaksha</td>
<td>Asteracanth longifolia</td>
<td>Root</td>
</tr>
<tr>
<td>3.</td>
<td>Black gram</td>
<td>Phaseolus mungo</td>
<td>Seed</td>
</tr>
<tr>
<td>4.</td>
<td>Kapikacchu</td>
<td>Mucuna prurita</td>
<td>Seed</td>
</tr>
<tr>
<td>5.</td>
<td>Shatavari</td>
<td>Asparagus racemosus</td>
<td>Rizome</td>
</tr>
</tbody>
</table>

---

8. Gokshura

9. Kapikacchu

10. Black gram

11. Black gram

12. Kokilaksha
Role of Gokshuradi Yoga in the Management of Klaibya (Erectile Dysfunction) 117

Discussion
In the present clinical study, after analyzing etiological factors of klaibya, it was found that aharaja, viharaja, and manasika factors play a role in the manifestation of this. Quality and quantity of shukra has direct relation with klaibya. Acharya caraka considered klaibya as lakshana under shukra kshya but klaibya will manifest both as a disease and a symptom, current parallel science has also supported the same.

Assessment was done on the bases of, sexual desire, breathlessness during coitus,

Table 1. Assessment of sexual desire in both groups

<table>
<thead>
<tr>
<th>Mean</th>
<th>N</th>
<th>B.T.</th>
<th>A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A</td>
<td>11</td>
<td>1.90</td>
<td>0.72</td>
<td>1.18</td>
<td>62.10</td>
<td>0.60</td>
<td>0.18</td>
<td>6.55</td>
<td>&lt;.001</td>
</tr>
<tr>
<td></td>
<td>Group B</td>
<td>10</td>
<td>1.80</td>
<td>1.30</td>
<td>0.50</td>
<td>27.77</td>
<td>0.52</td>
<td>0.16</td>
<td>3.125</td>
<td>&lt;.025</td>
</tr>
</tbody>
</table>

1 Sexual desire (Table 1)
In group A, out of 15 patients, 11 patients that is 62.10% found relief, stastically it is highly significant (P<.001).
In group B 10 patients out of 15 patients that is 27.77% found relief, stastically it is significant (p< .025).

Table 2. Assessment of Breathlessness during coitus in both groups

<table>
<thead>
<tr>
<th>Mean</th>
<th>N</th>
<th>B.T.</th>
<th>A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A</td>
<td>09</td>
<td>1.88</td>
<td>0.66</td>
<td>1.22</td>
<td>64.89</td>
<td>0.65</td>
<td>0.21</td>
<td>5.80</td>
<td>&lt;.001</td>
</tr>
<tr>
<td></td>
<td>Group B</td>
<td>11</td>
<td>1.81</td>
<td>1.36</td>
<td>0.45</td>
<td>24.86</td>
<td>0.52</td>
<td>0.15</td>
<td>3.00</td>
<td>&lt;.025</td>
</tr>
</tbody>
</table>

2 Breathlessness during coitus (Table 2) &
In group A, out of 15 patients, 9 patients that is 64.89% found relief, stastically it is highly significant (P<.001).
In group B 11 patients out of 15 patients that is 24.86% found relief, stastically it is significant (p<.025).

Table 3. Assessment of Excessive perspiration during coitus in both groups

<table>
<thead>
<tr>
<th>Mean</th>
<th>N</th>
<th>B.T.</th>
<th>A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A</td>
<td>13</td>
<td>1.92</td>
<td>0.69</td>
<td>1.23</td>
<td>64.06</td>
<td>0.70</td>
<td>0.19</td>
<td>6.47</td>
<td>&lt;.001</td>
</tr>
<tr>
<td></td>
<td>Group B</td>
<td>14</td>
<td>1.92</td>
<td>1.57</td>
<td>0.35</td>
<td>18.22</td>
<td>0.49</td>
<td>0.13</td>
<td>2.69</td>
<td>&lt;.02</td>
</tr>
</tbody>
</table>

3 Breathlessness during coitus (Table 3)
In group A, out of 15 patients, 13 patients that is 64.06% found relief, stastically it is highly significant (P<.001).
In group B 14 patients out of 15 patients that is 18.62% found relief, stastically it is significant (p<.025).

Table 4. Assessment of Premature Ejaculation in both groups

<table>
<thead>
<tr>
<th>Mean</th>
<th>N</th>
<th>B.T.</th>
<th>A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Group A</td>
<td>13</td>
<td>2.30</td>
<td>1.23</td>
<td>1.07</td>
<td>46.52</td>
<td>0.90</td>
<td>0.25</td>
<td>4.28</td>
<td>&lt;.01</td>
</tr>
<tr>
<td></td>
<td>Group B</td>
<td>15</td>
<td>1.86</td>
<td>1.46</td>
<td>0.40</td>
<td>21.50</td>
<td>0.63</td>
<td>0.16</td>
<td>2.44</td>
<td>&lt;.05</td>
</tr>
</tbody>
</table>

4 Premature ejaculation (Table 4)
In group A, out of 15 patients, 13 patients that is 46.52% found relief, stastically it is significant (P<.010).
In group B 14 patients out of 15 patients that is 46.52% found relief, stastically it is significant (p<.050).
Table 5. Assessment of Erectile Dysfunction in both groups

<table>
<thead>
<tr>
<th>N</th>
<th>Group A</th>
<th>Mean B.T.</th>
<th>Mean A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td>11</td>
<td></td>
<td>2.09</td>
<td>0.72</td>
<td>1.37</td>
<td>65.55</td>
<td>0.16</td>
<td>0.20</td>
<td>6.8</td>
<td>&lt;.001</td>
<td>HS</td>
</tr>
<tr>
<td>12</td>
<td>Group B</td>
<td>2.0</td>
<td>1.41</td>
<td>0.59</td>
<td>29.50</td>
<td>0.66</td>
<td>0.19</td>
<td>3.02</td>
<td>&lt;.02</td>
<td>S</td>
</tr>
</tbody>
</table>

5 Erectile dysfunction (Table 5)
In group A, out of 15 patients, 11 patients that is 65.55% found relief, statistically it is highly significant (P<.001). In group B 12 patients out of 15 patients that is 18.62% found relief, statistically it is significant (p<.025).

Table 6. Assessment of Pain during ejaculation in both groups

<table>
<thead>
<tr>
<th>N</th>
<th>Group A</th>
<th>Mean B.T.</th>
<th>Mean A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td></td>
<td>1.50</td>
<td>1.0</td>
<td>0.50</td>
<td>33.33</td>
<td>0.52</td>
<td>0.16</td>
<td>3.00</td>
<td>&lt;.02</td>
<td>S</td>
</tr>
<tr>
<td>11</td>
<td>Group B</td>
<td>1.54</td>
<td>1.18</td>
<td>0.36</td>
<td>23.37</td>
<td>0.50</td>
<td>0.15</td>
<td>2.39</td>
<td>&lt;.05</td>
<td>S</td>
</tr>
</tbody>
</table>

6 Pain during ejaculation (Table 6)
In group A, out of 15 patients, 10 patients that is 33.33% found relief, statistically it is highly significant (P<.001). In group B 11 patients out of 15 patients that is 23.37% found relief, statistically it is significant (p<.050).

Table 7. Assessment of Volume of Sperm in both groups

<table>
<thead>
<tr>
<th>N</th>
<th>Group A</th>
<th>Mean B.T.</th>
<th>Mean A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td></td>
<td>2.57</td>
<td>3.10</td>
<td>-0.53</td>
<td>-20.62</td>
<td>0.55</td>
<td>0.14</td>
<td>3.70</td>
<td>&lt;.01</td>
<td>S</td>
</tr>
<tr>
<td>15</td>
<td>Group B</td>
<td>2.90</td>
<td>2.86</td>
<td>0.04</td>
<td>1.37</td>
<td>0.14</td>
<td>0.03</td>
<td>1.103</td>
<td>&gt;.10</td>
<td>IS</td>
</tr>
</tbody>
</table>

7 Volume of the semen (Table 7)
After the treatment all the patients of group A found 20.62% benefit, statistically it is significant (P<.01). Whereas in group B 1.37% of benefit was found, statistically it is non-significant (p<.10).

Table 8. Assessment of Sperm count in both groups

<table>
<thead>
<tr>
<th>N</th>
<th>Group A</th>
<th>Mean B.T.</th>
<th>Mean A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td></td>
<td>46.33</td>
<td>55.20</td>
<td>-8.86</td>
<td>-19.12</td>
<td>8.44</td>
<td>2.18</td>
<td>4.068</td>
<td>&lt;.01</td>
<td>S</td>
</tr>
<tr>
<td>15</td>
<td>Group B</td>
<td>51.33</td>
<td>50.53</td>
<td>0.80</td>
<td>1.55</td>
<td>2.04</td>
<td>0.52</td>
<td>1.517</td>
<td>&gt;.10</td>
<td>IS</td>
</tr>
</tbody>
</table>

8 Sperm count (Table 8)
After the treatment all the patients of group A found 19.12% benefit, statistically it is significant (P<.01). Whereas in group B 1.55% of benefit was found, statistically it is non-significant (p<.10).

Table 9. Assessment of Motility of sperm in both groups

<table>
<thead>
<tr>
<th>N</th>
<th>Group A</th>
<th>Mean B.T.</th>
<th>Mean A.T.</th>
<th>Dif</th>
<th>% of Change</th>
<th>S.D.</th>
<th>S.E.</th>
<th>t</th>
<th>P</th>
<th>R</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td></td>
<td>51.33</td>
<td>61.66</td>
<td>-10.33</td>
<td>-20.12</td>
<td>10.93</td>
<td>2.82</td>
<td>3.66</td>
<td>&lt;.01</td>
<td>S</td>
</tr>
<tr>
<td>15</td>
<td>Group B</td>
<td>52.33</td>
<td>51.66</td>
<td>0.67</td>
<td>1.28</td>
<td>2.85</td>
<td>0.66</td>
<td>1.00</td>
<td>&gt;.10</td>
<td>IS</td>
</tr>
</tbody>
</table>

9 Motility of Sperm (Table 9)
After the treatment all the patients of group A found 20.12% benefit, statistically it is significant (P<.01). Whereas in group B 1.28% of benefit was found, statistically it is non-significant (p<.10).
excessive perspiration during coitus, premature ejaculation, erectile dysfunction, pain during ejaculation.

**Probable Mode of action of Gokshuradi yoga**

1. **Sexual desire**
   In the present clinical study *Gokshuradi yoga* containing gokshura, black gram, kapikacchu seed, shatavari, etc does nourishment, nourine tonic, increases sexual vigor and increases sexual stamina, along with this satvavajaya treatment will boost the effect.

2. **Breathlessness during coitus**
   As *Gokshuradi yoga* does nourishment of dhatu, vata shaman and increases the strength of the body mind, so intern helps in the relieving this symptom.

3. **Excessive perspiration during coitus**
   To combat this symptom along with this yoga satvavajaya treatment is beneficial.

4. **Premature ejaculation**
   Due to premature ejaculation dhatu kshaya will be the effect to enrich shukra dhatu gokshuradi yoga is beneficial because combination of drugs having properties like shnigdha guna, seeta veerya etc.

5. **Erectile dysfunction**
   The drugs of the gokshuradi yoga having properties nourishing, neuron tonic, which intern helps in the Erectile dysfunction which is leading cause for klaibya.

6. **Pain during ejaculation**
   Vitiation of vata dosha due to nidana sevan intern manifest in the form of pain during ejaculation the gokshuradi yoga containing drugs, having properties like guru, snigdha, ushanveerya, madhura rasa helps in pain during ejaculation.

**Discussion on tables**

Discussion on the symptoms of klaibya (which shows statistically highly significant Value)

1) **Breathlessness during coitus (Table 2)** — In group A, out of 15 patients, 9 patients that is 64.89% found relief, statistically it is highly significant (P<.001).
   In group B 11 patients out of 15 patients that is 24.86% found relief, statistically it is significant (p<.025).

2) **Breathlessness during coitus (Table 3)** — In group A, out of 15 patients, 13 patients that is 64.06% found relief, statistically it is highly significant (P<.001).
   In group B 14 patients out of 15 patients that is 18.62% found relief, statistically it is significant (p<.025).

3) **Erectile dysfunction (Table 5)** — In group A, out of 15 patients, 11 patients that is 65.55% found relief, statistically it is highly significant (P<.001).
   In group B 12 patients out of 15 patients that is 18.62% found relief, statistically it is significant (p<.025).

**Discussion on laboratory investigations**

1. **Volume of the semen (Table 7)** — after the treatment all the patients of group A found 20.62% benefit, statistically it is significant (P<.010). Whereas in group B 1.37% of benefit was found, statistically it is non-significant (p<.10).

**Summary**

In the present clinical study entitled “A Clinical Study on the Role of Gokshuradi Yoga in the Management of Klaibya Roga”

Description regarding causative factors, symptoms, pathogenesis of klaibya, and mode of action of gokshuradi yoga seen.

1. Regarding the principle (siddhant) of the study
   The combination of all the drugs in this gokshuradi yoga, nourishs all the 7 dhatu, because of quality of drugs having snigdha, guru, seeta, which are similar to shukra dhatu. Based on the concept samanaguna bhuhista and samanyam ektvakara, this supports the principle of this study.
2. Regarding klaibya

By taking different kind of vata doshakara ahara, vihara, manasika causes, by vyadhi karshana etc causes vitiats vatadi doshas, which intern vitiats agni ,because of this their will be uttarottar dhatu kshaya which leads to klaibya roga.

Conclusion

In the present study patients were divided into two groups, namely group A and groupB

Group A :- subjects were administerd with gokshuradi yoga. The relief found in the symptoms are as follows, sexual desire (62.10%), breathlessness during coitus(64.89%), excessive perspiration during coitus (64.06%), premature ejaculation (65.55%), erectile dysfunction (46.52%), and pain during ejaculation (33.33%). stastically it is significant (p< .05).

Group B :- subjects were administerd with placebo cap containg soybean powder. The reliefs found in the symptoms are follows, sexual desire (27.77%), breathlessness during coitus (24.86%), excessive perspiration during coitus (18.62%), premature ejaculation (46.52%), erectile dysfunction (29.50%) and pain during ejaculation (23.37%).

Comparing both Groups the over all 56.99 % improment in group A and were as in group B is 24.17% found

References


9. Database on medicinal plants used in Ayurveda & Siddha CCRAS Vol.1, 3 & 4


Address for correspondence: Dr Manjunatha.T.Sasanoor, M.D (Samhita), Dept. of P. G. Studies in Maulik Siddhant and Samhita, National Institute of Ayurveda, Amer Road, Jaipur, Rajasthan - 302002 (India). E-mail: drmanjuss@gmail.com
AGE RELATED MACULAR DEGENERATION: THERAPEUTIC TARGETS AND POSSIBLE ROLE OF AYURVEDA

MANOJ KUMAR,1* S.C. VARSHNEY2 AND O.P.S. MAURYA3

Department of Shalakya Tantra, 1*  Department of Shalya Tantra, 2 Faculty of Ayurveda, Department of Ophthalmology, 3 Faculty of Medicine, Institute of Medical Sciences, Banaras Hindu University, Varanasi – 221005  U.P. (India)

Abstract: Age related macular degeneration (ARMD) is a leading cause of blindness in developed countries. The prevalence of ARMD is rising due to rise in aged population. Despite of better understanding of its pathogenesis, promising therapeutic agents are yet not available. Therefore, preventive strategies are likely to be more helpful and should be emphasized. New approaches are needed to slow the progression and limit the damage caused by this disease. Ayurveda is well recognized for its role in aging related problems. An integrative approach, taking into account the knowledge of both Ayurveda and Biomedicine, to manage this problem may be more rewarding. The integrative approach need to be based upon pathophysiological correlations between both systems. This paper reviews the pathophysiology of ARMD with a view to understand therapeutic targets and discusses the possible role of Ayurveda in its management.

Keywords: Age related macular degeneration (ARMD), Degeneration, Timira, Ayurveda.

Introduction

Age related macular degeneration (ARMD) was first described in 1874 as senile macular degeneration, a symmetrical central choroidoretinal disease.1 It has also been termed as age related maculopathy (AMD). It is a leading cause of blindness2 and the most common cause of untreatable blindness in the developed countries with a prevalence of 0.05% before the age of 50 years that rises to 11.8% after the age of 80 years.3 The prevalence of age related macular degeneration is expected to rise in coming decades because of the projected increase in aging population.3,1

The magnitude of ARMD in developing countries like India is also disturbing. A study conducted on South Indian population concludes that the age-gender-area–adjusted prevalence of AMD in population was 1.82% which is found similar to that reported in white populations. Exudative AMD is responsible for 80% of the AMD related blindness.2 Age related macular degeneration is categorized as dry or non-exudative form and wet or exudative form. There is no preventive or curative treatment available for dry form of ARMD. Wet form of ARMD is treated with anti-angiogenic drugs, thermal laser and photodynamic therapy. ARMD involves macular region of ocular fundus with characteristic lesions, the drusen and the areas of hyper or hypo-pigmentation. In early stages of age-related macular degeneration, the visual symptoms are unremarkable,4 but in late stages severe loss of vision is usual.1

As no satisfactory treatment is available for ARMD, new approaches are needed to slow the progression and limit the damage caused by this disease. Ayurveda is well recognized for its role in aging related problems. An integrative approach, taking into account the knowledge of both Ayurveda and Biomedicine, to manage this problem may be more rewarding. The integrative approach needs to be based upon pathophysiological correlations between both systems.

This paper reviews the pathophysiology of ARMD with a view to understand therapeutic
targets and discusses the possible role of Ayurveda in the management of ARMD.

**Clinical presentation**

Age-related macular degeneration is diagnosed on the basis of clinical signs. Visual acuity usually remains normal in early stages.\(^5\) It is characterized by the presence of drusen, \textit{- the dot like lesions of whitish or yellow in colour}, \textit{- in the macular region.}

Age-related macular degeneration is divided into the early stage, in which visual symptoms are not present or may be minimal\(^4\) and the late stage, in which severe loss of vision is usual.\(^1\) Early age-related macular degeneration is characterized by drusen or by hyperpigmentations or small hypopigmentations. Drusen become visible on ophthalmoscopy when their diameter exceeds 25 \(\mu m\).\(^6\) Larger drusen indicate higher risk of visual loss due to ARMD.\(^7\)

**Dry age-related macular degeneration**

Dry ARMD is also referred to as non-exudative, atrophic or geographic ARMD. Ophthalmoscopically, it is visible as atrophy of the neuroretina with presence of drusen which gradually grow in number and size. It starts with a round hypopigmented spot in macular region through which large choroidal vessels are visible.

The initial symptoms of dry age-related macular degeneration are scotomas presented as ‘gaps in an image, as if letters had dropped out of a line of text’.\(^1\) In early course of the disease visual acuity remains normal but as disease advances there is gradual distortion and loss of central vision.

**Wet age-related macular degeneration**

This is also referred to as exudative or neovascular ARMD. In addition to the features of dry ARMD it is characterized by choroidal neovascular membrane (CNV) formation. The neovascularization tend to grow towards the fovea.\(^1\)

Choroidal neovascular membrane (CNV) may result in bleeding and scar formation under the retina. The scar formation results in severe and irreversible visual loss. Wet AMD accounts for 50% of registrations of blindness due to AMD.\(^8\)

Wet age-related macular degeneration may also lead to retinal detachment as serous fluid or blood collects under neuroretina. The detachment disturbs the fine arrangement of the photoreceptors and causes image distortion called metamorphopsia. It is usually a bilateral condition, but can also occur unilaterally.

**Risk factors**

The most important risk factor for ARMD is smoking. The genetic and environmental factors also play a significant role.

**Smoking**

Smoking has shown positive correlation with age-related macular degeneration\(^9,10\) and reduces the concentration of macular pigment by as much as 50% in a dose-response relationship.\(^11\)

**Light-induced toxic effects**

Excessive exposure to light is associated with age-related macular degeneration.\(^12,13\) Light can induce formation of reactive oxygen species, which in turn lead to the formation of toxic lipid and protein peroxidation products.\(^14\)

**Anatomy and Pathophysiology**

The inner retina is present towards the vitreous side and the outer retina towards the choroidal side. The outer retina includes the photoreceptors (rods and cones) and the retinal pigmentary epithelium (RPE). RPE lies on the Bruch’s membrane. The choriocapillaries, the capillary layer of the choroid, are related to the outer side of RPE. The inner retina is supplied by retinal vessels and outer retina by choriocapillaries. The nutrients and oxygen pass through Bruch’s membrane to reach outer retina. The cones are concentrated in macular region and rod density is highest in parafoveal region. This structural organization provides optimal environment for retinal function i.e. high resolution and color vision (cones) and peripheral vision and vision at dusk (rods).\(^1\)

**The Retinal Pigment Epithelium**

The RPE plays a key role in pathogenesis of age-related macular degeneration. It is a
cuboidal monolayer of cells with a very high metabolic rate. The most important functions of RPE are
1. Regeneration of bleached visual pigments.
2. Formation and maintenance of the interphotoreceptor matrix and Bruch’s membrane.
3. Phagocytosis

**RPE as Phagocytic System**

The RPE acts as a phagocytic system that is essential for the renewal of photoreceptors. Each photoreceptor has an inner and outer segments. A study on rhesus monkeys found that the outer segment of each rod consists of about 1000 disks, and the outer segment of each cone has about 700 disks. The disks are involved in conversion of light into electrical signals with the help of a transmembrane protein called rhodopsin. A large number of photoreceptor outer segments are shed daily which are then taken up by RPE and degraded. In the rhesus monkey, about 3000 disks are taken up daily by each RPE cell. The ingested material from photoreceptors combines with lysosomes to form phagolysosomes where it is digested. Proteins, nucleic acids and lipids are recovered in process and are recycled back for reuse. The contents of the phagolysosomes are, however, not completely degraded by lysosomal enzymes and form the residual bodies that are the substrates for lipofuscin formation.

The lipofuscin keeps on accumulating within RPE throughout life. The highest concentration of lipofuscin is found in the parafoveal region. This is the area where rod density is highest and dry age related macular degeneration begins. In late age related macular degeneration, the lipofuscin may disappear due to RPE cell death and their removal by phagocytes.

**Lipofuscin and A2E**

Lipofuscin contains a retinoid called A2E. High concentration of A2E inhibits the proton pump of lysosomes which results in leakage of lysosomal contents into the cytoplasm of RPE cells and cause cell injury. It can also damage the DNA and mitochondrial membranes in RPE which may induce apoptosis.

**Chromophores**

The number of RPE cells is reduced with age, resulting in compromised phagocytic system. In old age, pheomelanin and all-trans retinal dimers are also found in higher concentrations. They impair lysosomal function and further injure the RPE cells. The injured RPE cells attract dendrites from choroidal dendritic cells. Chromoidal dendritic cells are antigen presenting cells and constitute the core of approximately 40% of all drusen. The accumulation of debris in the RPE cells also increases the risk of phototoxic damage. In people over 80 years of age, the debris can occupy more than one fifth of the total volume of an RPE cell.

**Bruch’s Membrane**

Bruch’s membrane covers the outer aspect of RPE. It has three layers, an inner collagenous layer, a central elastic layer and an outer collagenous layers. Inner collagenous layer is lined by the basement membranes of the RPE and the outer collagenous layer is covered by choriocapillaris. Proteoglycans are also the constituents of Bruch’s membrane which are negatively charged molecules. Their negative charge helps to maintain homeostasis in the RPE. ARMD leads to altered homeostasis resulting in the deposit formation between the basement membrane of the RPE and the inner collagenous layer of Bruch’s membrane.

**Drusen**

Drusen are deposits that accumulate in the area between RPE and the inner collagenous zone of Bruch’s membrane and considered as the hallmark of ARMD. Their size may vary from just ophthalmoscopically visible dots to large deposits of approximately up to 250 μm in size. Few small drusen are found in 95% of the aged population but larger and numerous drusen are related to ARMD. The presence of large and numerous drusen, especially accompanied by hypopigmentation, is a major risk factor for developing choroidal neovascularization and the loss of central vision.
Drusen formation is a complex process influenced by the activation of the immune system and local inflammation. The drusen is composed of RPE remnants, dendritic cell processes, immunoglobulins, components of the complement cascade and the membrane attack complex (MAC). This composition of drusen shows that immunological and low grade inflammatory components are involved in drusen formation. The presence of drusen leads to degeneration of adjacent structures viz. photoreceptors, RPE cells and Bruch’s membrane, and release of angiogenic factors resulting ultimately in choroidal neovascularization.

Thickness of Bruch’s membrane increases with age due to deposition of lipids resulting in decreased fluid and nutrient transport across the membrane. There is reduction in concentration of metalloproteinases (MMPs) within Bruch’s membrane which leads to impaired proteinase activity and formation of deposits resulting in increased thickness of Bruch’s membrane. Bruch’s membrane thickening affects the survival of RPE cells as well as choriocapillaries. Altered properties of Bruch’s membrane result in diminished cell adhesion, and apoptosis.

The deposits between RPE and Bruch’s membrane evoke low grade chronic inflammation, and invasion by dendritic cells, macrophages and immune cells. The injured RPE cells, macrophages, and dendritic cells release inflammatory cytokines. The macrophage activity may be significantly increased and clears the deposited material (drusen), a finding that indicates advanced tissue damage and may mark the onset of neovascularization.

Photoreceptors consume more than 90% of retinal oxygen supply. In dark adapted states, oxygen requirements are further increased. The choroidal circulation is substantially decreased with increasing age. Further, the diffusion of oxygen through thickened Bruch’s membrane from choroid to RPE is reduced. The oxygen supply to inner retina is relatively much less and cannot compensate for compromised state of oxygenation in outer retina of ARMD patients. A near-hypoxic environment is created in outer retina. Hypoxia is further increased in dark adapted states when oxygen demand is increased. Hypoxia increases the secretion of growth factors such as vascular endothelial growth factor (VEGF), which cause choroidal angiogenesis.

Angiogenesis occurs due to imbalance between pro-angiogenic (e.g. vascular endothelial growth factor, VEGF) and anti-angiogenic (e.g. pigment epithelium derived factor) factors. Hypoxia may shift the balance in favour of neovascularization. The new vessels are not normal vessels as they are fragile and easily bleed. Degenerative changes, neovascularization, haemorrhages and scar formation result in vision loss in ARMD.

Correlations with Ayurvedic Pathophysiology

ARMD as Timira

Ancient Ayurvedic literature describes twelve categories of diseases that affect vision. The diseases that usually present as painless loss of vision in absence of any other ocular sign (if observation with unaided eyes) are called as timira in Ayurvedic literature. Timira are further classified according to the involvement of doshas. Timira may present either as diminution of vision as sole feature, or may be associated with visual symptoms like metamorphopsia, floaters, scotomas etc.

ARMD presents with following features
1. In early stages ARMD is asymptomatic but as disease progresses visual acuity diminishes.
2. Central scotoma is caused by advanced macular degeneration or macular hemorrhages.
3. Visual loss may be severe and deteriorates up to light perception (PL+).
4. Scar formation at macula may cause metamorphopsia.
5. No other sign, which can be observed with unaided eye, is present.

All these features are characteristic of timira, therefore, ARMD should be considered as a type of timira.
Involvement of Doshas

The earliest clues of involvement of doshas come from the study of risk factors. ARMD is exclusively found among elderly people. Its association with old age points to the involvement of vata dosha (vata is a term that describes regulatory mechanisms and neurological phenomenon that occur in the body) in the pathogenesis of ARMD, as old age is associated with predominance of vata. There is degeneration and loss of neural tissue (photoreceptor degeneration) in the posterior pole of eye. The degeneration of neural tissue again indicates a role of vata, as neural tissue is considered as a component of vata in the body.

In later stages of disease, however, the involvement of other doshas, viz. rakta (cellular component of blood) and pitta (pitta correlates with inflammatory and proinflammatory molecules; digestive and lysosomal enzyme system) is also visible as neovascularization and bleeding are caused by abnormality of rakta; and inflammation is a feature of vitiated pitta.

Mala Samchaya

The immediate cause of degeneration in ARMD is mala samchaya (deposition of waste material) and reduced tissue capability to deal with waste material. According to Ayurveda, the agni (agni is a term that describes the factors that transform raw nutrients into simpler products which can be used by the body) transforms the nutrient material into sarabahga (the products which can be used by body) and malabhaga (malas usually are the waste materials). Vata separates sarabahga from malabhaga. The diminished action of agni leads to formation of semidigested material and abnormal vata is incapable of separating sarabahga from malabhaga resulting in accumulation of unwanted material in the tissue. The accumulation of unwanted material in the body tissues is known as mala samchaya.

In ARMD outer segment of photoreceptors cannot be fully digested by lysozomal enzymes and remain entrapped within the RPE cells, ultimately resulting in degeneration. This aspect of ARMD pathogenesis correlates with and explained by the abnormal agni and vata.

We are concerned here with the aspect of agni which is involved in digestion of shed outer segments of photoreceptors. The agni in the form of lysozomal enzymes works on photoreceptor segments forming two products viz. the malas as described above, and sara bhaga. The sara bhaga is circulated back to photoreceptors so that it can be reused for formation of new photoreceptor segments.

ARMD occurs as a result of inadequate action of agni (agnimandya) on shed outer segments leaving them semiprocessed (apakva). This inadequately processed product is called as ama (literally ama means uncooked). Therefore, minute amounts of ama keeps on accumulating within RPE cells (ama samchaya) for years, and ultimately manifest as visible pathology in fundus.

Ama, according to Ayurveda, forms amavisha in dhatus i.e. in the intracellular and extracellular milieu. In modern parlance, it correlates with enhanced toxic state. This involves the mechanisms by which low grade chronic inflammation is evoked. These mechanisms involve signaling, genetic expression of certain molecules, recruitment of leucocytes, formation of immunoglobins and increase in concentration of cytokines.

There is an increase in Bruch’s membrane thickness which results from deposition of lipids within the membrane. Deposition in Bruch’s membrane is associated with decreased MMP activity suggesting the reduced action of proteases in continuous remodeling of Bruch’s membrane. This also correlates with agnimandya and consequent deposition of waste materials (malas samchaya).

Srotodushti

The deposition in Bruch’s membrane and drusen formation (mala samchaya) in RPE cells produces hindrance to transportation of molecules. The thickening of Bruch’s membrane results in decreased transport of oxygen and creates hypoxic environment in retinal tissue. This aspect
of pathogenesis correlates with srotodushti, particularly pranavaha and rasavaha (the concept of srotas correlates with transport systems working in the body).

The presence of hypoxia indicates abnormality of prana vayu or pranavaha srotas. Oxygen, oxygen transport system and the mechanisms that regulate oxygen supply to tissues can be correlated with prana vayu and pranavaha srotas.

Hypoxia caused by abnormality in pranavaha srotas along with already vitiated pitta and rakta, results in release of angiogenic factors. The latter are responsible for choroidal neovascularization.

Conclusion

It can be concluded on the basis of above correlations that ARMD is primarily a vataja verity of timira. But in case of wet ARMD when neovascularization and hemorrhages occur, the vitiated pitta and rakta along with vitiated vata play a significant role in progression of ARMD.

With this understanding of the pathophysiological role played by various factors, a hypothesis is proposed here. ARMD can be considered due to abnormality of vata which leads to dhatwagnimandya, mala samchaya and ama formation. Ama may lead to chronic inflammation, formation of Amavisha and consequent tissue damage.

Correlations between pathophysiology of ARMD, and the general process of disease development as well as pathogenesis of timira provide important clues to an integrated approach to treat this condition. Ayurvedic drugs or modalities which exhibit vatahara, deepanapachana, srotoshodhaka and amapachana properties, and described in context of timira may provide ground for further research. They can be studied for their role in drusen formation, and for modifying the degenerative process occurring in macula. Rasayana therapy may be particularly useful in prevention of age related macular degeneration.

References

8. Mr Richard Newson: Age-related macular degeneration InnovAiT 2008; 1(10):710–713
Lipofuscin fluorophore inhibits DNA is a target of the photodynamic effects elicited in A2E-laden RPE by blue-light illumination. Invest Ophthalmol Vis Sci 2003; 44:2245-51


Bressler NM, Silva JC, Bressler SB, Fine SL, Green WR: Clinicopathological correlation of drusen and retinal pigment epithelial abnormalities in age-related maculardegeneration. Retina 1994; 14:130-142


Jerzy Z. Nowak Age-related macular degeneration (AMD): pathogenesis and therapy Pharmacological Reports 2006, 58, 353 363 ISSN 1734-1140


**Address for correspondence:** Dr. Manoj Kumar, MS (Ay.), Assistance Professor, Department of Shalakya Tantra, Faculty of Ayurveda, Institute of Medical Sciences, Banaras Hindu University, Varanasi - 221005. E-mail ID: mkumarbhui@gmail.com
CONFERENCES AND FORTHCOMING EVENTS

SEMINAR ON ‘HERBAL PRODUCTS: REGULATORY ASPECTS’

29th & 30th June 2012
TNMC & BYL NAIR CH. HOSPITAL,
DR. AL NAIR ROAD, MUMBAI CENTRAL, MUMBAI - 400008

The Department of Clinical Pharmacology, TNMC & BYL Nair Ch. Hospital is organizing a 2-day Seminar on “Herbal Products: Regulatory Aspects” on 29th & 30th June 2012. The objective of the Seminar is to review the current regulations on herbal products, and to update health care professional/stakeholders involved in herbal research/industry with respect to the regulatory requirements for evaluating the safety, efficacy and quality of herbal products. This interactive seminar will aid in facilitating better understanding of the current regulations pertinent to herbal medicines/products.

The organizers expect participation from both academia and industry who are actively involved in research, development and marketing of Medicinal Plants and herbal products.

Program highlights:
- Regulations to launch a Herbal Product (Traditional Medicine/Food/Health supplement/raw material)
- Regulatory requirements regarding Quality of Herbal Products
- Regulatory requirements for safety data documentation/generation of herbal products
- Recent notifications related to herbo-mineral formulations in India
- Claim substantiation; regulatory requirements
- AYUSH GCP
- IPR issues related to herbal products

Venue: TN Medical College Auditorium, BYL Nair Ch. Hospital, Dr. AL Nair Road, Mumbai Central, Mumbai - 400008

<table>
<thead>
<tr>
<th>Registration Fees*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Category</td>
</tr>
<tr>
<td>31st May 2012</td>
</tr>
<tr>
<td>Students#</td>
</tr>
<tr>
<td>Faculty/Professionals</td>
</tr>
<tr>
<td>Industry Delegates</td>
</tr>
</tbody>
</table>

* Includes Registration kit with breakfast/lunch/tea only. Accommodation not included.
# Proof from Head of Department/Principal/Dean required for students.

Registration Fees will be accepted in Cash or by DD or local cheque (payable at par) drawn in favour of “BYL Nair Hospital DDF”.

Last date for registration: **31st May 2012**

Registration Form available at [www.tnmcnair.com](http://www.tnmcnair.com)

For further inquiries kindly contact:

**Dr. Samidha Kalekar**
Organizing Secretary, Department of Clinical Pharmacology 5th Floor, G Building, TNMC & BYL Nair Ch. Hospital, Dr. AL Nair Road, Mumbai Central, Mumbai - 400008
Telephone: 022 23014713
E-mail: admin@dcpnairhospital.org