

Study of macroscopic and microscopic plant characters: Assure the presence of plant constituents in traditional ayurvedic formulation

KARUNAKAR SHUKLA, SWARNLATA SARAF and SHAILENDRA SARAF*

Institute of Pharmacy, Pt. Ravishankar Shukla University, Raipur - 492 010 (India)

(Received: April 15, 2007; Accepted: May 19, 2007)

ABSTRACT

The aim of the present research work is to determine macroscopic, microscopic characteristics of the plants constituents in raw material and in final product for assuring the presence of plant material in the Ayurvedic formulation. Three batches of *Bhaskar Lavan Churna* were prepared in laboratory and three samples from different manufacturers were procured and subjected to various analysis. The macroscopic characteristics of different plant parts used in the formulation are sufficient to establishing the identity. The microscopic characters of powdered plant material give idea about presence of the different herbs in Ayurvedic formulation. The determined microscopic characters when analyzed in laboratory and marketed formulation, absence of different characters have been observed in marketed formulations. The determined macroscopic and microscopic characters can be used as a standard in routine quality control of the formulation.

Key words: Microscopic, macroscopic, plant characters, Ayurvedic formulation.

INTRODUCTION

Macroscopic and microscopic characters of the different plant parts were used for identification and authentication purpose since long time¹. Keeping this in mind, affords have been made in determining macroscopic and microscopic plant characteristics for assuring the quality of the single plant as well as for compound formulations in traditional system of medicine^{2,5}. The most important problem with the compound formulation is that most of them consist of several plant constituents, the presence of which should confirm in the final product by developing certain botanical (microscopic and macroscopic characters) or by analytical mean. The present study was undertaken to determine macroscopic and microscopic characters of different plant parts with medicinal value available in Chhattisgarh neighbor region and application of these determined characters in compound formulation used in traditional system.

Bhaskar Lavan churna (BLC) is one of the most popular Ayurvedic formulation described in 'Sarangadharasamhita', Ayurvedic formulary of India has given the specification for the composition of *Bhaskar Lavan churna*^{3, 4}.

MATERIAL AND METHODS

Microscope used for determination of characters were MICRON OPTIC (BINO_{C XI}), all the microscopic characters were obtained by taking the samples from six different places in the container.

Procurement of Plant material

Crude drugs were procured from local market and identified by macroscopic and microscopic measurements^{6, 7}.

Preparation of laboratory formulation:

Three batches were prepared in laboratory according to strict methods of 'Ayurvedic formulary of India' and *Sarangadharasamhita*². Commercially

available brand BLC-A, BLC-B, and BLC-C, of *Bhaskar Lavan churna* was procured from local market.

Macroscopic evaluation of plant material^{8,9}

Sensory characteristic

Plant material obtained from different places was evaluated on the basis of sensory

Table - 1: Contents of *Bhaskar Lavan churna*

S. No.	Local name/ Botanical source
1.	Dhanyaka / (<i>Coriendrum sativum</i>)
2.	Pippali / (<i>Piper longum</i> ,Fr.)
3.	Pippali mula / (<i>Piper longum</i> ,Rt.)
4.	Krsna jirka / (<i>Caram carvi</i>)
5.	Patraka / (<i>Cinnamomum tamala</i>)
6.	Nagaksara / (<i>Mesua ferra</i>)
7.	Talisa / (<i>Abies webbiana</i>)
8.	Amlavetasa / (<i>Rheum emodi</i>)
9.	Marica / (<i>Piper nirgum</i>)
10.	Jiraka / (<i>Cuminum cyminum</i>)
11.	Visva / (<i>Zingiber officinale</i>)
12.	Dadima / (<i>Punica Granatum</i>)
13.	Tvak / (<i>Cinnamomum zeylanicum</i>)
14.	Ela / (<i>Elettaria Cardmomum</i>)
15.	Samudra Lavan / (Sea salt)
16.	Sauvarcala Lavan / (black salt)
17.	Vida Lavan / (Vida salt)
18.	Saindhav Lavan / (Rock salt)

organs and characters recorded in the terms of colour, odour, test, and shape. Size was measured with the help of scale and recorded in the term of length, width and diameter (Table -1).

pH

pH of 1 % solution of powdered drug were determined and mentioned (Tables-2).

Loss on drying

The moisture content was determined by heating the plant material at 105 °C in a oven to a constant weight. Weight about 5 gms of plant material heat it in a oven until the constant weight achieved and calculate the percentage loss on drying (Table -2).

Ash Values

The ash values of the plant material in term of total ash were determined using standard procedure. Weight about 3 gms of plant material in a tared silica crucible, incinerate the powdered drug

by gradually increasing the heat until the free from carbon. Keep it in desiccators, weight the ash and calculate the percentage of ash (Table -2).

Extractive values

Extractive values were determined as per the method given in Indian Pharmacopoeia and WHO publication. These are described below:

Water-soluble Extractives

3 gms of Powered plant material was dispersed in 100 ml of water and allowed to stand for 24 hours with occasional shaking and filtered and water was evaporated. The above procedure was performed for each plant powder and the dried water extractive was weighted (Table -2).

Methanol Soluble Extractives

3 gms Powered plant material was dispersed in 100 ml of methanol and allowed to stand for 24 hours with occasional shaking and filtered and methanol was evaporated (Table -2).

Microscopic evaluation of Powdered plant material⁹

For determining microscopic plant characteristics small quantity of all the samples of the powdered plant materials from different sides of container taken out and observed. For the purpose of microscopic study boil the powdered plant materials separately with chloral hydrate solution, washed with water stained with different laboratory dies and mounted in glycerin. (Table-3).

Determination of microscopic features in traditional formulation¹⁰

For determining microscopic plant characteristics small quantity of all the samples of the churna (laboratory and marketed formulations) from different sides of container along taken out and compared with the genuine samples of powdered plant material. For the purpose of microscopic study boil the churna separately with chloral hydrate solution, washed with water stained with different laboratory dies and mounted in glycerin. Another small quantity of respective samples washed to remove the sugar, cleared by heating with chloral hydrate, washed again, cooled and mounted in glycerin to identify the microscopic characters of the all ingredients (Table-4).

Table - 2: Macroscopic and Physicochemical characters of plants used in Bhaskar Lavan churna

S. No.	Name of the plant	Colour/ Odour/Taste	Shape/ Size Analysis	pH/Moisture Content/ Ash Content			Extractive values		Phytochemical Analysis	
				Water soluble	Alcohol soluble	Test performed	Result			
1	<i>Coriandrum sativum</i> (Dried Fruits)	Yellowish brown / Aromatic / Spicy and characteristic	Subglobular cremocarpous fruit/2-4 cm diameter, 3-4 cm in length	5 / 3.5% / 3 %	14 %	08 %	Biurate test	Positive (Protiens)		
2	<i>Zingiber officinale</i> (Dried Rhizome)	Yellowish brown/ Characteristic / Aromatic by warm sensations	laterally compressed, bearing short flat ovate branches/ 1.5-6.5 cm diameter	6 / 6 % / 4 %	11 %	04 %	Molish test	Positive (Carbohydrate)		
3	<i>Piper longum</i> (Dried fruits)	Dull dark brown to black / characteristic odour / bitter	small, ovoid, sunken structure embedded in flashy spike/ 2.5-4cm length, 1-2cm in dia	4.5 / 6 % / 6 %	16 %	14 %	Dragondroff test	Positive (alkaloids)		
4	<i>Piper longum</i> (Dried Roots)	dark brown to black / characteristic odour / bitter	small, cylindrical much branched/ 4-9 cm length, 0.5-1cm in diameter	4.5 / 8 % / 9 %	13 %	11 %	Dragondroff test	Positive (alkaloids)		
5	<i>Piper nigrum</i> (Dried Fruits)	black / Characteristic aromatic/ Pungent	globular or oblong / 4-6mm in diameter	4 / 8 % / 9 %	16 %	09 %	Dragondroff test	Positive (alkaloids)		
6	<i>Cuminum cyminum</i> (Dried Fruits)	dark brown / ridges are light brown /characteristic aromatic	Mericaip is having five longitudinal ridges / 4-6 mm in length, 2 mm thick characteristic aromatic	7 / 8 % / 7 %	18 %	12 %	Biurate test	Positive (protiens)		
7	<i>Abis webbiana</i> (Dried leaves)	dark brown / Odorless/bitter	opposite or sub-opposite, glabrous and are scaly, / 2-3 by 4-6 cm	5 / 8 % / 4 %	22 %	18 %	FecI3 test	Positive (terpenoids)		
8	<i>Punica Granatum</i> (Dried seeds)	dark brown /Odorless /bitter	opposite, glabrous and are scaly, shining above / 1-2 by 0.3-0.5 cm	4 / 2 % / 6 %	16 %	14 %	FecI3 test	Positive (terpenoids)		
9	<i>Carum carvi</i> (Dried Fruits)	yellowish brown /characteristic aromatic /Aromatic and sweet	Mericaip is elongated, narrow, tapering at the end shows presence of 5 yellow primary ridges /2- 7mm	7 / 3 % / 5 %	11 %	07 %				
10	<i>Cinnamomum zeylanicum</i> (Stem bark)	Dark brown / Fragrant / Aromatic and sweet	Found in compound quills / 1 m length, 1 cm diameter	5 / 2.5% / 3 %	12 %	10 %	FecI3 test	Positive (terpenoids)		
11	<i>Rheum emodi</i> (Dried rhizome)	Brownish red /odorless / bitter	barrel shaped cylindrical / 8-10 cm length, 3-4 cm in thickness	5 / 2 % / 6 %	09 %	08 %	Brontager test	Positive (glycoside)		
12	<i>Eleteria cardamomum</i> (Dried fruits)	green to pale buff /characteristic aromatic /aromatic, sweetish	ovoid or oblong 3 sided, sharply backed at top/2 cm in length, 1-3 cm	7 / 8 % / 5 %	12 %	10 %				
13	<i>Mesua terra</i> (Dried Stamens)	Dark brown / characteristic /bitter	Fragrant/2-3cm in length, 0.2-0.5 cm in diameter	6 / 7 % / 6 %	14 %	12 %	FecI3 test	Positive (terpenoids)		
14	<i>Cinamomum tamala</i> (Dried leaves)	Greenish brown /characteristic penetrating /bitter	opposite or sub-opposite, glabrous and are scaly, shining above / 5-7.5 by 12.5-20 cm	6 / 4% / 7 %	13 %	11 %				

Table - 3: Analysis of microscopic characters of different plant constituent's present in laboratory formulation















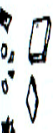





S.No	Characteristics	Shows presence	Inference
1.	 Microsette crystals of calcium of Calcium oxalate in the cells of coriander.	<i>Coriandrum sativum</i>	Presence
2.	 U-Shaped, lignified cells form the cinnamon bark.	<i>Cinnamomum zeylanicum</i>	Presence
3.	 Parenchyma cells contain starch grains from ginger.	<i>Zingiber officinale</i>	Presence
4.	 Broken glass like yellow fragment(Vittae) from carum.	<i>Carum carvi</i>	Presence
5.	 Calcium oxalate crystals from rhubarb.	<i>Rheum emodi</i>	Presence
6.	 Dark brown stone cells from piper longum	<i>Piper longum</i> (Fr.)	Presence
7.	 Polygonal cell with oil globules from Cumimum cyminum	<i>Cumimum cyminum</i>	Presence
8.	 Stone cells from Piper longum roots	<i>Piper longum</i> roots	Presence
9.	 Perisperm cell filled with oleo resin from Piper nigrum fruits	<i>Piper nigrum</i> fruits	Presence
10.	 Parenchymatous cells of perisperm with prisms from cardamomum	<i>Elleteria cardamomum</i>	Presence

Table - 4: Comparative analysis of microscopic characters of different plant constituents

S.No.	Characteristic	Formulation-L	A	B	C
1.	 <p>Microsette crystals of calcium of Calcium oxalate in the cells of coriander.</p>	+	+	+	-
2.	 <p>U-Shaped, lignified cells form the cinnamon bark.</p>	+	+	+	+
3.	 <p>Parenchyma cells contain starch grains from ginger.</p>	+	+	+	+
4.	 <p>Broken glass like yellow fragment(Vittae) from carum.</p>	+	-	-	-
5.	 <p>Calcium oxalate crystals from rhubarb.</p>	+	+	+	-
6.	 <p>Dark brown stone cells from piper longum</p>	+	+	+	+
7.	 <p>Polygonal cell with oil globules from Cumimum cyminum</p>	+	+	+	+
8.	 <p>Stone cells from Piper longum roots</p>	+	+	-	+
9.	 <p>Perisperm cell filled with oleo resin from Piper nigrum fruits</p>	+	+	-	+
10.	 <p>Parenchymatous cells of perisperm with prisms from cardamomum</p>	+	-	+	-

RESULTS AND DISCUSSION

In the present study plants material were studied for macroscopic and microscopic characters. Macroscopic characters were studied on the basis of sensory evaluations and compared with characters given in literature (Table-2). Microscopic characters of the different plant material were determined and recorded. Laboratory batches were evaluated for their microscopic parameters and compared with three marketed formulations. Microscopic examination of laboratory formulation gives idea about presence of plant constituents on the basis of their characters like microsette crystals of calcium of calcium oxalate in the cells of coriander, U-Shaped, lignified cells and starch grains form the *Cinnamomum zeylanicum*, parenchyma cells contain starch grains and fibers from the *Zingiber officinale*, Broken glass like yellow fragment (Vittae) from *Carum carvi*, calcium oxalate crystals from *Rheum Emodi*, polygonal cell with oil globules from *Cuminum cyminum*, stone cells from *Piper longum* roots, perisperm cell filled with oleo resin from *Piper nigrum* fruits, parenchymatous cells of perisperm with prisms from *Elleteria cardamomum* and Dark brown stone cells from piper longum. All these characteristics give idea about presence of plant constituents in the laboratory formulation (Table-3).

When marketed formulations evaluated for the presence of determined characteristics formulation A showing the nonappearance of broken glass like yellow fragment (Vittae) from *Carum carvi* and parenchymatous cells of perisperm with prisms from *Elleteria cardamomum*. Formulation B showing the nonappearance of broken glass like yellow

fragment (Vittae) from *Carum carvi*, stone cells from *Piper longum* roots and perisperm cell filled with oleo resin from *Piper nigrum* fruits. Formulation C showing the nonappearance of broken glass like yellow fragment (Vittae) from *Carum carvi*, calcium oxalate crystals from *Rheum emodi*, parenchymatous cells of perisperm with prisms from *Elleteria cardamomum* and microsette crystals of calcium of calcium oxalate in the cells of *Coriander sativum* (Table-4). These results show appearance or nonappearance of the plant material in the marketed formulations. Therefore the determined macroscopic and microscopic characters for the plant material gives idea about quality of Ayurvedic formulations *Bhaskar Lavan churna* and developed method for determination could be used as a valuable analytical tool in the routine analysis, to check the batch to batch variation

Conclusion

These studied macroscopic and microscopic plant characters can be utilized to assure the presence of plant material in powdered traditional (Ayurveda, Siddha, and Unani), herbal formulations and in collection of Medicinal plants. It can also be used as a primary quality control parameter for routine analysis of commercial formulation containing same plant materials.

ACKNOWLEDGEMENTS

The authors are highly thankful to AICTE and CGOST for financial assistance & Director, Institute of Pharmacy, Pt. Ravishankar Shukla University, Raipur (C.G.) for providing infrastructural facilities to carry out the work.

REFERENCES

1. P.K. Mukherjee, *Quality Control of Herbal Drug*, first ed., published by Eastern Publishers (Business Horizontal Ltd.), New Delhi, pp: 03-30,184-219. (2002).
2. S. Agarwal, R.H.Singh, "Proceedings of International Congress" Ayurveda, Abstract no.: 209, pp: 221. 28-30th January (2002).
3. C.K.Kokate, *Pharmacognosy*, twenty ninth ed. published by Nirali Prakashan, pp: 550-559. (2002).
4. *The Ayurvedic Formulary of India*, Part-I, second ed. Published by Govt. of India, Ministry of Health and Family Planning, Department of Health, pp: 103-119. (2003).
5. *Pharmacoepical standards for Ayurvedic Formulations*, Central Council for Research in Ayurvedic and Siddha, Ministry of Health and Family Welfare, pp: 112-123. (1987).
6. *Quality Standards Of Indian Medicinal Plants*, Volume I & II, Indian Council of Medicinal Research, New Delhi, pp: 95,168 (2003).
7. *Indian Herbal Pharmacopoeia*, Volume II, Regional Research Laboratory Jammu, Indian drug Manufacturing Association Mumbai, pp:90-93.162. (1999).
8. "Quality control methods for medicinal plant material", published by WHO regional office for western pacific manila pp: 08, 22-31. (1993).
9. K.R.Khandelwal, *Practical Pharmacognosy Techniques and Experiments*, ninth ed. published by Nirali Prakashan, pp: 152. (2002).
10. C.K. Kokate, *Practical Pharmacognosy*, fourth ed. published by Nirali Prakashan, pp: 143. (1996).