COMPARATIVE ‘NAMBURI PHASED SPOT TEST’ ANALYSIS OF SAMUDRAPHENA (CUTTLEFISH BONE)

Dr. PRAMODKUMAR¹, Dr. P.G.JADAR²

Abstract

Samudraphena (Cuttlefish bone) is one of the Pranija (Animal origin) sudha vargadravya (calcium compounds) used for eye and ear ailments mainly in Ayurveda. Shodhana of Samudraphena was done by triturating with citrus juice then subjected to organo-leptic & NPST analysis. NPST method is newly introduced in the field of Ayurvedic mineral pharmaceutical standardization accepted by CCRAS. NPST helps differentiate between, and thus identify, various bhasmas. It depends upon the pattern of the spot, which develops after a specific chemical reaction. Hence, present work was undertaken to test the purity of Samudraphena using above tests collected from Mumbai, Kolkata and Thiruvanathapuram. The organo-leptic properties of all the samples after shodhana shows no much difference but NPST spot test analysis of samples shows mild variations in their color in respective phases. The sample 1 which was collected from Mumbai in fresh form shows desired result compared to standard NPST result.

Keywords
Samudraphena, Sudhavarga, NPST, Cuttlefish, Cuttlebone, Ayurveda

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INTRODUCTION

The oceans are full of living organisms. In the course of evolution, marine organisms have adapted excellently to the marine environment, such as high salt concentration, low temperature, high pressure, and low nutrient availability. These extreme conditions require unique adaptation strategies leading to the development of new natural products, which differ from known structures of terrestrial organism\(^2\). One such natural product is *samudraphena* (cuttlebone) get separated when cuttlefish dies. In the Rasa literatures it is included in the *Pranija sudha vargardravya*. In the classical literatures it has great importance in the therapeutic point of view. Chemically it is having calcium carbonate, including various organic and inorganic elements. Due to its immense medicinal value Ayurveda has utilized it for various ailments.

In India generally *Samudraphena* (Cuttlefish bone) are obtained from four species of Cephalopods (cuttlefishes) along with their respective code numbers which are as follows \(^3\).

- *S. aculeate* - 5011
- *S. elliptica* - 5012
- *S. pharaonis* - 5013
- *S. prashadi* - 5014

As cuttlefishes are having very less life span (1-3 years) \(^4\) thus availability is never a concern. When the fish dies naturally the Samudraphena is collected near the coast so there is no question for ethical objection & are easily available in the market for less cost.

Namburi Phased Spot Test (NPST) is the study of spot and colors at three successive phases spreading over three different time intervals developed and standardized by Dr. Namburi Hanumantha Rao in 1970, it has been accepted by CCRAS, New Delhi. When a drop of clear solution of a substance (Bhasma or Sindura) under examination is put on Whatman paper impregnated with suitable reagent, a spot with series of changes in color and pattern will appear. In chemistry, techniques involving spot tests or chromatography are widely used. It has the advantage of measuring the sensitivity of reactions at different time intervals. This method is used to detect or study continual chemical reactions that take place gradually.
between two chemical substances on static media at every second. It is used to assess the bhasma qualitatively.

In this NPST technique a methodology evolved to identify bhasma (calx) of *Sudha varga dravya* (calcium compounds) with minute differences in over all chemical reactions, it has become necessary to study the organoleptic properties of our sample in comparison with organoleptic properties of standards\[^5\].

**MATERIALS AND METHODS**

1. Procurement of samudraphena: Three samples (sample 1, 2 & 3) were procured from three different places within India namely Mumbai (Maharashtra), Kolkata (West Bengal), and Thiruvananthapuram (Kerala) & were authentified in National Institute of oceanography, Dona poula, Goa.

2. Shodhana of Samudraphena of three samples was carried out in P.G Departament of Rasashastra, K.L.E U.’s Shri B.M.K.Ayurveda Mahavidyalaya, Belgaum.

3. Organoleptic & NPST analysis of all the three samples was carried in P.G Departament of Rasashastra, K.L.E U.’s Shri B.M.K.Ayurveda Mahavidyalaya, Belgaum.

**METHODOLOGY:**

**Shodhana of Samudraphena (cuttlebone):**

First its outer surface is to be scrapped and remaining is powdered in *khalwa yantra* (*Mortar & pestle*) then *bhavana (lavage)* of *Nimbuswarasa* (lemon juice) given for one day and it is dried \[^6\].

**Organoleptic analysis:**

The purified samples were subjected to various Organoleptic analysis after shodhana table no.1 & during heat as well as wet treatment is tabulated in table no.2

**Namburi Phased Spot Test:**

**Materials:**

a) Distilled water

b) Haridra paper: Whatman Paper No.1 impregnated and dried in an alcoholic Extract of Haridra swarasa (*Curcuma longa* Linn)

c) Test tubes: Three

d) Samudraphena churna (cuttlebone powder) (Sample 1 to 3)
Procedure:

All the three samples were subjected to NPST. Initially 0.25g of Samudraphena churna (cuttlefish bone powder) was put into test tube and heated on spirit lamp till the lower end of the test tube becomes red hot & allow the test tube to cool. Then 0.5 ml of distilled water was added to all the test tubes, shaked well and allowed to settle. Then one drop of clear solution of each sample was put on Haridra paper (prepared using Whatman's filter paper no.1) and observed for spot pattern in the following three phases:

1st phase: 0 to 5 min
2nd phase: 5 min to 20 min
3rd phase: 20 min to 1 day

(Figure -1a(sample 1 phase 1),1b(sample1 phase 2),1c(sample 1 phase 3) , 2a(sample 2 phase 1),2b(sample2 phase 2),2c(sample 2 phase 3), 3a(sample 3 phase 1),3b(sample3 phase 2),3c(sample3 phase 3))

Observations of NPST

NPST phased spot observations tabulated in table no.3 & standard phased spot observations given in tabulated in table no.4.

DISCUSSION AND CONCLUSION

All the three samples collected from three sources respectively from Mumbai, Kolkata, Thiruvananthapuram shows no much Organoleptic differences after shodhana that is due to all the calcium compounds after reacting with the any acids converts into salts of the respective compounds and shows same features but still the sample collected from the Mumbai shows fishy odour & turns into light ash colour that is because sample was collected in the fresh form & still some organic things like polysaccharides remains of the cuttlefish present in it. NPST analysis of all samples shows slight differences with respect to colour in all phases but the sample 1 which was collected from the Mumbai shows desired color change as compared to the standard. The classical text don’t have any parameters for checking the quality of the samudraphena, but NPST analysis is the only test available to check the quality of samudraphena, as the test is chemical reaction-based, with specific
results for specific bhasmas, we can differentiate between bhasmas clearly. If it is adulterated with any of the sudha varga dravya than it shows some other colour changes in NPST analysis. This technique is very helpful for quality assessment of Bhasma as per the standards of Rasashastra. In other words, bhasmas can be identified by their name given in Rasashastra by virtue of their quality differences, but not chemically. It is such a simple test that it can be carried out with minimum set up and requirements. CCRAS has also accepted the monograph of NPST, and so the quality of product can be checked before being used therapeutically. In the present study, though the samudraphena was purified by same method, there was difference in colour, and according to NPST analysis, only sample 1 (Mumbai) which was collected in the fresh form & prepared in our department, gave results as per standard.

Images of Sample 1 of three phases

1a (sample 1 phase 1)  1b (sample1 phase 2)  1c (sample 1 phase 3)
Images of Sample 2 of three phases

2a (sample 2 phase 1)  
2b (sample 2 phase 2)  
2c (sample 2 phase 3)
Images of Sample 3 of three phases

3a (sample 3 phase 1)  
3b (sample 3 phase 2)  
3c (sample 3 phase 3)
Table 1

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Sample 1</th>
<th>Sample 2</th>
<th>Sample 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>i. Colour</td>
<td>Ash</td>
<td>Creamish white</td>
<td>Creamish white</td>
</tr>
<tr>
<td>ii. Odour</td>
<td>Fishy</td>
<td>Unpleasant</td>
<td>Unpleasant</td>
</tr>
<tr>
<td>iii. Taste</td>
<td>Alkaline</td>
<td>Alkaline</td>
<td>Alkaline</td>
</tr>
<tr>
<td>iv. Touch</td>
<td>Smooth</td>
<td>Smooth</td>
<td>Smooth</td>
</tr>
</tbody>
</table>

Table 2

<table>
<thead>
<tr>
<th>Stages</th>
<th>Sample 1</th>
<th>Sample 2</th>
<th>Sample 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heat treatment:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Liberation of fumes</td>
<td>Black</td>
<td>Brownish Black</td>
<td>Brownish black</td>
</tr>
<tr>
<td>Odour</td>
<td>Unpleasant</td>
<td>Unpleasant</td>
<td>Unpleasant</td>
</tr>
<tr>
<td>Change of color</td>
<td>Nil</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>Wet treatment:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Exothermic/</td>
<td>Nil</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>Endothermic</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Color of solution</td>
<td>Straw</td>
<td>Light straw</td>
<td>Straw</td>
</tr>
<tr>
<td>Absorption</td>
<td>Normal</td>
<td>Normal</td>
<td>Normal</td>
</tr>
<tr>
<td>Settling time</td>
<td>Rapid</td>
<td>Rapid</td>
<td>Rapid</td>
</tr>
</tbody>
</table>

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Table 3
NPST Spot pattern observations of Samudraphena samples

<table>
<thead>
<tr>
<th>Phases</th>
<th>Sample 1</th>
<th>Sample 2</th>
<th>Sample 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>I phase (0-5 min)</td>
<td>Soon after dropping solution a bright wet darkish pink circle forms at the centre and gradually wet circle fades away leaving light pink circle at centre</td>
<td>Wet dark pinkish circle appears followed by widening of wet circle around the pink circle</td>
<td>Wet dark brownish pink circle appears followed by widening of the wet circle gradually seen</td>
</tr>
<tr>
<td>II\textsuperscript{nd} Phase (5-20 min)</td>
<td>Wet circle faded with reduction in the brightness of the pink coloured circle at the centre</td>
<td>Periphery of centre circle became light brownish pink at the end of II phase with widening of wet circle.</td>
<td>Wet circle faded away in this phase.</td>
</tr>
<tr>
<td>III\textsuperscript{rd} Phase (20 min-1 day)</td>
<td>Wet circle faded away with light pink circle at its centre</td>
<td>Widening of Wet circle with light brownish pink circle at the centre at the end of 2\textsuperscript{nd} phase.</td>
<td>Wet circle fades way.</td>
</tr>
</tbody>
</table>
Table 4

<table>
<thead>
<tr>
<th>Sample name</th>
<th>Phase I</th>
<th>Phase II</th>
<th>Phase III</th>
</tr>
</thead>
<tbody>
<tr>
<td>Samudraphena</td>
<td>A thin pink circle forms on a wide wet spot</td>
<td>Starts fading away</td>
<td>The entire circle fades away by 24 hours</td>
</tr>
</tbody>
</table>

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