FURTHER STUDIES ON CHEMICAL EVALUATION OF LAUHA BHASMA III

A. KESHRI, P.R.P VERMA and C.M PRASAD*
Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra,
Ranchi -835215, Bihar
Department of Pharmacy, Muzaffarpur Institute of Technology*,
Muzaffarpur – 842 002, Bihar

ABSTRACT: Samples of marketed Lauha Bhasma from different manufactures were evaluated chemically. Apart from the 81 -85% iron content, the 15-19% other constituents were determined therein. Ferrous ferric and total iron in a single aliquot were determined spectrophotometrically, Qualitative and chromatographic analysis indicate the presence of sodium, potassium, calcium copper and cobalt in the samples, silicious matter and traces of ascorbic acid were present while tannin was absent in lauha Bhasma. Quantitatively sodium and potassium were determined by flame spectrometry. Upon fractionation, water soluble and acid soluble contents were determined.

INTRODUCTION

On qualitative and quantitative analysis the samples of lauha bhasma, an ayurvedic drug, used in the iron deficiency anaemia, were found to contain both the ferric and the ferric oxide than ferrous oxizide1. Upon in vivo evaluation on healthy adult male rabbits, samples of lauha bhasma were considered better bhasma were considered better on the basis of increase I the haemoglobin content than the allopalhic iron preparation of ferrous sulphate2. This establishes its superiority over the allopathic iron preparation.

In the present study further chemical evaluation was done of the samples of Lauha bhasma, from not only the earlier manufacturer but also from other manufacturers. This study enables us to complete the chemical evaluation by determining the chemical constituents of the 15-19% apart from the total iron content.

MATERIAL AND METHODS

Lauha bhasma samples were from different manufacturers viz Baidyanath (all commonly available variants), sadhna, Dabur Gurukul Kangari Cooperative drug factory. All chemicals and reagents were of analar grade.

1. Determination of Iron content

Ferrous, Ferric and total iron content in a single aliquot were determined spectrohotometrically at 515 mm. 1,3-4 The results of the analysis are tabulated I Table - 1.

2. Evaluation of inorganic content

Qualitative test: Non – luminous Bunsen flame5,6 was used to detect the presence of calcium, sodium and potassium.

Chromatographic analysis: Descending paper chromatography was employed to detect the presence of cobalt and copper. In this Acetone: Ethylacetate: 6M HCl (9:9:2)
as solvent and 1% Rubeanic acid in ethanol as spraying reagent were used\(^7\).

**Quantitative test:** Amount of sodium and potassium were determined by flame spectrometry\(^8\) on systronic digital FPM 125 flame photometer. With the aid of appropriate filters in flame photometer and standard curve of potassium ions (made with solutions containing 20,10,5,2 ppm of potassium ions) and sodium ions (made with solutions containing 10,5,2,5,1 ppm of sodium ions). Different ions were determined in the solution of *Lauha Bhasma* this was repeated with different samples of Lauha Bhasma and the results obtained are presented in Table -1.

3. Evaluation of organic matter:

Tannin and ascorbic acid may be present in the samples of lauha bhasma triphla [a mixture of terminalia chebula, terminalia belerica and phyllanthus emblica which is added during of preparation may be a source of tannin and ascorbic acid.

Tannin was determined qualitatively in the test solutions of *Lauha Bhasma* by comparing with the presence of tannin in catechu solution 9-11 (Table -2).

Ascorbic acid (Vitamin C) was also determined qualitatively in the test samples12 and the test solution was compared with standard solution of ascorbic acid the results are depicted in Table -3.

4. Fractional analysis of lauha Bhasma:

100 mg of *Lauha Bhasma* was taken and shaken with 100ml of distilled water for half an hour, it was then filtered and the filter paper was dried to a constant weight the residue on filter paper was carefully washed off with dilute HCL (17ml of conc. HCL+140ml of distilled water). It was then heated at a temperature of 45oC to accelerate the solubility of acid soluble substances. Samples were again filtered and dried to a constant weight to get values of acid insoluble substances. The results are tabulated in table -4.

**RESULTS AND DISCUSSION**

On assaying the marketed *Lauha bahsma* samples from different manufacturers, % of ferric, % of ferrous and % of total iron content are found (Table 1). These results are more or less same as the results reported earlier1, except in the case of samples obtained form manufacturers D and H The % of ferric iron was found more than the % of ferrous iron in all the samples as it is evident in the form or ratios of ferric to ferrous (Table 1). This is because during the excessive heating process, the ferrous has been oxidized to ferriciron. Literature shows that although these bhasma are prepared from the same ayurvedic text the differences seen may be due to the variations in the processing of *Lauha Bhasma*.

The qualitative analysis and chromatographic analysis indicate the presence of sodium, potassium calcium, copper and cobalt. On quantitative analysis of Lauha Bhasma sodium (1.3 – 4.2 % W/W), potassium (0.6-2% W/W) ions are found in the samples. In the test different methods of preparation and purification of *lauha bhasma* are available. Also each method may be modified according to the physicians’experience. These factors may lead to the variation in their percentage in the different samples obtained from different manufacturers.

Tannin and ascorbic acid was determined qualitatively in the samples of *lauha bhasma*. From the tests, as depicted in tables 2&3, it is clear that Lauha Bhasma sample
do not contain tannin but traces of ascorbic acid has been found. The absence of other organic matter may be accounted by their loss as carbon dioxide, water etc during the numerous steps of heating the mixtures of Lauha churna and triphala. However,

Table 1
IRON, SODIUM AND POTASSIUM IONS IN THE SAMPLES OF LAUHA BHASMA

| S.No | Manufacturer | % of ferric* | % of ferrous* | % of total Iron* | Ratio of ferric to ferrous | % of sodium in the sample* | % of potassium in the sample* |
|------|--------------|-------------|---------------|-----------------|---------------------------|---------------------------|----------------------------|
| 1    | A            | 68.315      | 17.338        | 85.656          | 1:0.254                   | 4.253                     | 0.976                       |
|      |              | (0.125)     | (0.185)       | (0.124)         |                           | (0.109)                   | (0.019)                     |
| 2    | B            | 79.291      | 2.527         | 81.785          | 1:0.032                   | 4.185                     | 1.156                       |
|      |              | (0.241)     | (0.039)       | (0.03)          |                           | (0.076)                   | (0.012)                     |
| 3    | C            | 83.230      | 2.359         | 85.542          | 1:0.028                   | 4.257                     | 1.085                       |
|      |              | (0.178)     | (0.238)       | (0.047)         |                           | (0.020)                   | (0.089)                     |
| 4    | D            | 61.168      | 13.858        | 75.0.26         | 1:0.226                   | 3.596                     | 0.996                       |
|      |              | (0.131)     | (0.719)       | (0.681)         |                           | (0.053)                   | (0.023)                     |
| 5    | E            | 66.979      | 15.564        | 82.55           | 1:0.232                   | 3.174                     | 0.669                       |
|      |              | (1.378)     | (0.238)       | (1.581)         |                           | (0.014)                   | (0.018)                     |
| 6    | F            | 82.550      | 5.510         | 88.103          | 1:0.067                   | 1.645                     | 2.073                       |
|      |              | (0.294)     | (0.528)       | (0.776)         |                           | (0.113)                   | (0.039)                     |
| 7    | G            | 78.015      | 4.039         | 82.055          | 1:0.052                   | 1.742                     | 1.673                       |
|      |              | (0.115)     | (0.107)       | (0.092)         |                           | (0.040)                   | (0.054)                     |
| 8    | H            | 63.962      | 3.605         | 67.561          | 1:0.056                   | 1.318                     | 1.182                       |
|      |              | (0.201)     | (0.102)       | (0.302)         |                           | (0.054)                   | (0.076)                     |

*Mean of three runs. Values in parentheses indicate standard deviation.
## Table 2

**QUALITATIVE EVALUATION OF TANNIN IN LAUHA BHASMA**

| S.No | Test                        | 1% of catechu          | 1% of soln. of catechu       | Sample solution               | Inference          |
|------|-----------------------------|------------------------|-----------------------------|-------------------------------|--------------------|
|      |                             | In water               | In dil. HCl and then         | In water                      | In dil. HCl and then|                      |
|      |                             |                        | neutralized                 |                                | neutralized        |                      |
| 1.   | With FeCl₃ solution         | Green colour           | Greenish colour              | Green colour                  | No Colour          | Tannin absent       |
| 2.   | With lead subacetate solution | Coloured Ppt           | Coloured Ppt                 | Coloured Ppt                  | No Ppt             | Tannin absent       |
| 3.   | With gelatin solution       | Whitish Ppt            | Whitish Ppt                  | Whitish Ppt                   | No Ppt             | Tannin absent       |
| 4.   | With quinine sulphate solution | Pinkish white Ppt      | Pinkish white Ppt            | Pinkish white Ppt             | No Ppt             | Tannin absent       |

Ppt = Precipitate
| S.No | Test | Ascorbic acid standard soln. | Lauha bhasma water extract | Inference |
|------|------|------------------------------|----------------------------|-----------|
| 1.   | Test soln. + 5ml 2,6 di chlorophenol indophenol (0.05%) | Reagent soln. is decolourised | Reagent soln. is decolourised | Ascorbic acid absent |
| 2.   | 5ml test soln + 0.5ml 0.1N HCl + 3 drops of sod. Nitroprusside (5%) = 1ml of 0.1N NaOH soln. | Transient blue colour | No blue colour | Ascorbic acid absent |
| 3.   | 10ml test soln + 0.1 gm sod bicarbonate + 20mg ferrous sulphate stand for 20 minutes then 5ml of dilute H₂SO₄ was added | Purple colour appears | Purple colour on the top of layer | Ascorbic acid is present in traces |
The presence of traces of ascorbic acid may be due to the formation of sodium ascorbate which is supposed to be fairly stable at high temperatures.

On the basis of fractional analysis (Table 4), it can be concluded that the water extract contains sodium, potassium, calcium and traces of copper, cobalt and ferrous iron. The water insoluble but acid soluble portion contains ferric iron, ferrous iron, copper and cobalt, the water extract is seen to contain less copper and cobalt than the acid soluble filtrate. This indicates that cobalt and copper are present in the form of their oxides which is insoluble in the aqueous medium. Sodium and potassium in general are supposed to have higher aqueous solubility and hence it is extracted in the aqueous medium.

Ferrous and the ferric iron are assumed to be in the oxide form. Ferrous oxide is practically insoluble in water and alkalis, readily soluble in acids, while ferric oxide is only soluble in dilute hydrochloric acid. Fractional analysis of *Lauha Bhasma* indicates that silicious matter present is 1% w/w (Table 4), which may come from the earthen pots used during processing.

The present work details the chemical analysis and evaluation of *Lauha Bhasma*. This helps us to ascertain the quality of the product, standards or specifications which must be assigned so as to get the product of uniform standard, immaterial of the variations in a process, raw materials or other factors.

### Table 4
**Fractional Analysis of Lauha Bhasma**

| S.No | Weight of Sample (mg) | Weight of Water insoluble matter (mg) | Weight of Water soluble (mg) | Weight of Water insoluble matter (mg) |
|------|----------------------|--------------------------------------|----------------------------|--------------------------------------|
|      |                      |                                      |                            | Acid soluble                        | Acid Insoluble                      |
| 1.   | 100.2                | 95.244                               | 4.956                      | 94.098                              | 1.126                                |
| 2.   | 100.1                | 95.210                               | 4.890                      | 94.113                              | 1.097                                |
| 3.   | 99.9                 | 94.960                               | 4.931                      | 93.971                              | 0.998                                |

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