NUTRACEUTICAL VALUES OF CITRUS FRUIT SPREAD FORTIFIED WITH RASAYANA HERBS

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ABSTRACT

The results of studies made on three different types of spreads namely, Citrus fruit spread Balya (fortified with Withania somnifera and Chlorophyrum borivilianum), Citrus fruit spread Medhya (fortified with Bacopa monnieri and Convolvulus pluricaulis) and Citrus fruit spread Hridya (fortified with Terminalia arjuna and Boerhaavia diffusa) were reported. The general physico-chemical parameters and nutritive values were estimated by relative standardized methods. The results of physico-chemical parameters and assays were found to be satisfactory. Significant nutritive values prove that they can be used as dietary supplements. HPTLC fingerprinting of individual Citrus fruit spread shows the presence of respective Rasayana herbs. This study proves that these fortified spreads can be used as dietary supplements with significant therapeutic value.

KEYWORDS: Citrus fruit spread, Rasayana herbs, Physico-chemical parameters, Nutritive values, dietary supplements.

INTRODUCTION

Today’s world is extremely competitive and everyone is exposed to a fierce and continuous struggle for existence. The third millennium demands speedy actions at every step of life. This, in turn, has deeply affected our food habits. Processed, packed, readily available yet nutritious foods have become an essential part of our diet. In today’s fast and money-oriented lifestyle, people prefer “Ready to eat” instant food, over home-made food. One of such instant food items, consumed commonly, by almost all age groups and economical classes, are “Jams, Jellies and Marmalades.” Marmalades or Spreads are products, prepared from citrus fruit ingredients with one or more carbohydrate sweeteners1.

This paper involves the study of a product named “Citrus fruit spread”, which is a sort of fruit spread. Citrus medica (Bada nimbu) pulp is used as a base in this spread. Citrus medica Linn. (Family Rutaceae) is a garden plant, chiefly cultivated for its valuable fruit, in South-west and Northern India. The fruit and its juice is a stimulant, an excellent aromatic, stomachic, refrigerant, astringent and digestive. Its juice is a refrigerant drink that checks bilious vomiting and fevers. The fruit is used for making pickles and candies. The peel and rind are made into excellent refrigerant, astrigent and digestive.

Citrus fruit spread is also fortified with Rasayana herbs, to enhance its nutritive value. Rasayana, means “Rejuvenation” Therapy, which is a unique contribution of Ayurveda, which provides multi-dimensional benefits. It has the power to nourish all the Dhatus (tissues) properly and restore basic homeostatic balance of the body1. It improves the overall physical, mental and moral qualities. The herbs which are used in this therapy are called “Rasayana Herbs.”

Citrus fruit spread Balya is fortified with Balya Rasayana herbs namely, Withania somnifera (Ashwagandha) and Chlorophyrum borivilianum (Safed musli). Both these herbs act as tonic. They bestow longevity and age stabilization upon the user, as well as, retone...
youthfulness and maintain strength of all organs, optimally. They also increase the immunity of the body.

Citrus fruit spread Medhya is fortified with *Bacopa monnieri* (Brahmi) and *Convolvulus pluricaulis* (Shankhapushpi). “Medhya” means intellect. These herbs strengthen memory and intelligence, by Prabhava (specific action). Shankhapushpi as Medhya Rasayana was studied by Hetal et al. Also, studies on psychotropic effects of the Medhya Rasayana drug, Shankhapushpi were carried out by Singh et al. The role of Brahmi in brain function and therapy was also studied by Shinomol et al.

Citrus fruit spread Hridya is fortified with *Terminalia arjuna* (Arjuna) and *Boerhaavia diffusa* (Punarnava). These herbs improve the functioning of specific organs like the heart and kidneys. They also increase digestive power.

Thus, consumption of these fortified spreads, prevents early signs of old age, improves complexion, physical strength and immunity, and gives happiness to one’s self and life.

The general physico-chemical and nutritive analysis of the three spreads, was carried out, to evaluate whether they can be used as “Dietary Supplements” and presence of the respective Rasayana herbs used for fortification, was confirmed by well-versed HPTLC technique.

**MATERIALS AND METHODS**

**Physico-chemical Parameters**

Description, Loss On Drying at 105°C, Solubility in Water, total ash content pH of 1%w/v solution in water and acidity were carried out by previously established standard methods.

**Brix Reading and Refractive Index:**

Direct readings for Brix and Refractive Index were recorded, by gently spreading minimal quantity of sample, on the prism of a previously calibrated Abbe’s refractometer.

**Content of Tannins:**

Appropriate dilutions of sample and tannic acid standard were prepared, and color development was carried out, as given under the colorimetric procedure by S. Ranganna. Optical densities of sample and standard, were recorded at 760nm and content of tannin was calculated.

**Content of Flavonoids:**

About 2g sample was accurately weighed and 60ml methanol was added to it. This solution was frequently shaken and kept overnight. Next day, the methanol was decanted and two more washings, with 25ml methanol each, were given to the soaked sample. All the three methanolic extracts were collected and filtered through absorbent cotton. The filtrate was then concentrated to 10ml. The concentrated methanolic extract was then precipitated with 100ml of solvent ether, shaking it vigorously and keeping it for 30 minutes. Then, the ether layer was filtered through cotton and evaporated in a clean, pre-weighed beaker and dried to constant weight at 105°C, for 4 hours. The percentage of Flavonoids was then calculated.

\[
\text{% w/w of flavonoids} = \frac{\text{Weight of residue in g}}{\text{Weight of sample in g}} \times 100
\]

**HPTLC Fingerprinting:**

**Conditions:** Stationary phase - HPTLC Aluminium sheets silica gel 60 F 254, Solvent System - chloroform : methanol (9:1), Sample Volume - 15 microlitre, Spotting level – 1cm, Saturation time and Run Time - 20 minutes, Length of Run - 8cm, Evaluation - Peak height and area, Detection - After spraying with vanillin sulphuric acid reagent.

Preparation of Sample and Extracts used as Reference Standard for spotting

Citrus fruit spread (Balya, Medhya and Hridya): About 2.0 g of sample was dissolved in 10 ml of distilled water and thoroughly extracted with 10 ml of petroleum ether. Petroleum ether layer was collected and evaporated gently, on waterbath. The residue obtained was reconstituted in 5 ml of methanol and the clear solution was used as a sample solution.

Corresponding Extracts: 1g of each extract was gently extracted in 10ml methanol by sonicating and warming, if necessary, and filtered through Whatman Filter Paper No.1, and the clear filtrate was used as reference standard solution.

**Microbiology:** As per USP – 2010 – 33 Guidelines.

**Nutritive Values**

**Fats:**

Accurately weighed 4g to 5g of the sample was hydrolysed and extracted thoroughly, with solvent ether. The ether layer was collected and washed with water and dried gently on waterbath. The residual fat obtained, was dried at 105°C to constant weight and the fats were calculated as follows. Calculations:

\[
\text{Fats (w/w)} = \frac{[(C – B) \times 100]}{A}
\]

Where,

- \(A\) = Weight of the sample in g,
- \(B\) = Weight of empty conical flask in g,
- \(C\) = Weight of conical flask with the residue, dried at 105°C

**Proteins:**

Accurately weighed, 2g of the sample was digested by heating the same with a mixture of 10g potassium sulphate, 1.5 g of copper sulphate and 20ml concentrated sulphuric acid till it turned green. After the contents were cooled, they were transferred to ammonia
distillation flask. Cooled 40% Sodium hydroxide solution was added into it to make it alkaline. These contents were then distilled immediately, and liberated gas was gently collected in 50ml of 0.1M hydrochloric acid. After completion of distillation, the solution was titrated against 0.1M sodium hydroxide using methyl red as indicator, the end point being red to yellow(C). Similarly, blank was carried out by titrating 50ml of 0.1M hydrochloric acid against 0.1M sodium hydroxide, using methyl red as indicator (B).

Calculations: Proteins (%w/w) = \[(B – C) \times 0.0014 \times 6.25 \times 100 \times MF\] / A
Where,
A = Weight of sample in g,
B = Blank (Burette) Reading in ml,
C = Sample (Burette) Reading in ml,
MF = Molarity Factor of 0.1M Sodium hydroxide. 6.25 = Conversion factor of nitrogen to protein.

Fibre:
About 2g of sample was accurately weighed and then exhausted with ether. 200ml of boiling dilute sulphuric acid was added to the ether-exhausted marc, and refluxed accurately for 30 minutes, filtered, and the residue on the filter paper was washed to neutral. The residue was rinsed back into the flask with 200ml of boiling sodium hydroxide solution, refluxed accurately for 30 minutes, and then rapidly filtered through a tared filter paper. The residue on the filter paper was washed with boiling water, until the last washing was neutral, and rinsed with boiling sodium hydroxide solution, refluxed accurately for 30 minutes, and then rapidly filtered through a tared filter paper. The residue was incinerated, ignited to constant weight, cooled in a dessicator and the ash was weighed. The difference between the weight obtained by drying at 110°C and that of the ash represented the weight of the fibre. Percentage of fibre was calculated by applying the following mentioned formula.
Calculations: Fibre (%w/w) = [(C – B) – (E – D) X 100] / A
Where,
A = Weight of the sample in g,
B = Weight of empty dry filter paper in g,
C = Weight of filter paper with residue, after drying at 110°C, in g,
D = Weight of empty crucible in g,
E = Weight of crucible with residue, after ignition, in g.

Carbohydrates: Carbohydrates (%w/w) = 100 - (%Moisture + %Ash + %Protein + %Fats + %Fibre)

Sugars:
Sugar was analyzed by using the Lane and Eynon’s Method in 2g of accurately weighed homogeneous sample. (Sugar by Lane Eynon Method 923.09, 920.183b).

Glucose:
Glucose %w/w = (Blank reading in ml – Sample reading in ml) X 0.0045 X (100 / Weight of sample in g) X (100 / 50) X Normality Factor of 0.05N sodium thiosulphate.

Energy (Calorific Value):
4(%) Carbohydrates) + 4(%) Proteins) + 9(%) Fats) = Calorific Value in kilocalories.

Content of Iron:
Calibration of the spectrophotometer was done by standard method using potassium dichromate solution. Iron was estimated by carrying out colorimetric method on the ash of 5g of the homogeneous sample.

Content of Vitamin C:
Vitamin C was estimated by titration method using Indophenol in 2g of accurately weighed homogeneous sample. (Analysis of vitamin C by 2, 6-dichloroindophenol titration AOAC Method 967.21, 45.1.14 (2).)

RESULTS AND DISCUSSIONS:

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<th>Table 1: Physico-chemical parameters of “Citrus fruit spread” – Barya, Medhya and Hridya</th>
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The above, Table 1 summarizes physico-chemical parameters of all three types of Citrus fruit spread – Balya, Medhya and Hridya.

Descriptions of all samples show variation in color from light brown to reddish brown as per the color of rasayana herbs added in the original Citrus fruit spread. Loss on Drying shows variations from 29.769% to 31.171%. Solubility in water for Balya and Medhya is found to be around 68%, whereas in Hridya it is 70.83%. Sahi et al. have reported TSS in pineapple, apple, plum, orange and mango fruit as 9.00, 12.00, 16.00, 10.00, and 20.00 percent respectively. Degree brix gives idea about the soluble solids. Balya and Medhya give same brix reading that is 69.50 degree brix, however, Hridya shows bit higher value, around 72.50 degree brix. Refractive index also is found to be same in Balya and Medhya and higher in Hridya as recorded in Table 1. Total ash ranges from 0.361% to 0.383% as per the inorganic materials present in the respective samples.

The above, Table 2 illustrates microbiological status of Citrus fruit spread. Since all the three types of Citrus fruit spread are fortified with different rasayana herbs, it is necessary to check the microbiological load in individual lot. Table 2 shows the detailed microbiological evaluation on quantitative basis.

The above, Table 3 focuses on nutritive values in Citrus fruit spread, including iron content and vitamin C content. Considerable amount of fats, proteins, fibres, carbohydrates, sugars, glucose and calories in all three types of Citrus fruit spread impart dietary value to them. Also iron content along with vitamin make the product more valuable. Hridya Citrus fruit spread which is likely to be considered useful for nourishing the heart is found to contain more iron compared to other two types. Vitamin C in the sample, helps in absorbing the iron. Hence both the ingredients are valuable. The above Table 4 gives qualitative assurance for the presence of added rasayana herbs in Balya, Medhya and Hridya Citrus fruit spread. HPTLC, the sophisticated and more reliable technique is used for the same.

The overlapping of spectra cited in the above, Fig 1 proves the data given in Table 4, more effectively. Fig 2
supports the data of Ashwagandha in Balya Citrus fruit spread and Arjuna in Hridya Citrus fruit spread, as the overlapping spectra were obtained after derivatisation.

CONCLUSION
Perfect desired quality status, significant nutritive values and presence of medicinally useful rasayana herbs in Balya, Medhya and Hridya Citrus fruit spread, make them extremely valuable as dietary supplements. Therefore, we can use them in our breakfast, to make it more delicious and healthy.

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