A comparative study between the moist heat versus microwave method for the determination of vitamin C content in amla

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ABSTRACT

Amla/Indian gooseberry has a wide range of biological effects including antioxidant, antiviral, antimicrobial, antitumor and antibacterial activities. The fruits of the plant have been used in Ayurveda as a potent rasayana which is used to promote health and longevity. Amla is a rich source of polyphenols, minerals and vitamin C. This study is based on the investigations carried out to assess the loss of vitamin C (ascorbic acid) in Amla during moist heat and microwave oven method. The vitamin C content in moist heat and Microwave oven treated was noted to be about 888 mg/100 g and 881 mg/100g respectively. The yield of vitamin C estimated by titration showed that the yield enhanced from 983 to 1283mg to support the fact that the vitamin C levels have gradually increased with increase in power in microwave oven extraction method when compared to the moist heat extraction method. Thus the outcome is of translational value to the beverages and nutraceutical industry for shelf life improvement of the Vitamin C based health drinks.

Keywords: Amla, ascorbic acid, nutraceutical, microwave and moist heat

INTRODUCTION

Emblica officinalis, commonly known as amla/Indian gooseberry, belongs to the family of Euphorbiaceae. India ranks first in the area of amla crop production in the world. It has wide range of biological effects including antioxidant, antiviral, antimicrobial, antitumor and antibacterial activities. The fruits of the plant have been used in Ayurveda as a potent rasayana which is used to promote health and enable increasing the lifespan by boosting immune system, arresting the ageing process and revitalizing the body in debilitating conditions. Amla is a well known rich source of polyphenols, minerals and vitamin C. The content of vitamin C is ranging from 200-900 mg per 100 g of the edible portion [1,2,3]. Vitamin C is regarded as the first line of natural antioxidant defense in plasma and a powerful inhibitor of lipid peroxidation. It also regenerates the major antioxidant tocopherol (vitamin E) in lipoproteins and cell membranes. Intracellular mechanisms exist which can regenerate vitamin C (ascorbate) from its inactive metabolite dehydroascorbate by reduced glutathione [4]. The active ingredient of Amla comprises of gallic acid or ellagic acid structures attached to vitamin C. The leaves and bark are rich in tannin. The pH value of 10% w/v of aqueous solution is acidic [5,6]. TSS, pH, reducing sugars, total sugars and browning of the preserves increases during storage irrespective of different methods employed for preparation of the preserves. Moisture, ascorbic acid, tannin and titratable acidity of the preserves decreased during storage. The preserves at sugar syrup of 70°Bricks was found most effective in retention of ascorbic acid and tannins and showed low microbial load [7].
The amla fruit extract is also a good source of polyphenols, flavones, tannins and other bioactive substances. These substances are also strong antioxidants and might contribute to improved health benefits. Thus it is important to characterize different types of medicinal plants for their antioxidant and antimicrobial property. *E. officinalis* alone or in combination with other herbs has been useful in the treatment of cold, warts, skin afflictions, influenza, anemia, diabetes, lung conditions and elevated cholesterol as an immune restorative in cancer conditions. It is one of the best natural anti-ageing remedies used in acne and other skin problems and also effective in the treatment of acidity and peptic ulcers. Regular use of amla improves immunity, fights cancers and rejuvenates the body. It fights chronic diseases like hypertension, high cholesterol, Diabetes, AIDS, influenza, chronic infections like cough and cold, fatigue and inflammatory conditions. The amla seeds and leaves are used in asthma, bronchitis, biliousness, conjunctivitis, inflammation, dyspepsia and dysentery. Liquid fermented from amla fruit is good for indigestion, anemia, jaundice, heart complaints and for promoting urination [8]. Ayurveda describes amla as one of the best cure for diabetes, bleeding disorders, strength and stamina promoter. Clinical studies on patients with pulmonary tuberculosis showed that vitamins of *E. officinalis* was better assimilated than synthetic vitamin C [9]. The amla extracts showed significant protection to DNA against oxidative damage as found by migration of DNA in agarose gel. Amla inhibits radiation induced lipid peroxidation (LPO) in microsomes and superoxide dismutase (SOD) in mitochondria. As amla fruits are perishable in nature, its shelf life during storage is very limited. The contents of ascorbic acid decreases with increase in storage period. Its decay loss can be minimized by storing in regulated conditions. It was found that decay loss was minimum in modified storage condition whereas it was maximum in zero energy chamber. The chemical changes occur during processing and storage after 135 days. It was also observed that total soluble content, the level of vitamin C, tannin, etc., are decreased as in fresh fruits but they are rich in calcium, sugar and further increased acceptability after storage of upto 135 days [10,11]. A mature amla can tolerate freezing as well as high temperature of 46°C. Amla being water soluble, it may scavenge the free radicals responsible to initiate lipid peroxidation. However, ascorbic acid alone is not responsible for antioxidant activity. Other polyphenolic compounds capable of scavenging oxidizing radicals also contribute to it. Ascorbic acid in amla itself does not provide high protection even at elevated concentration. Therefore, ascorbic acid and other polyphenols present in natural formulation of amla have higher antioxidant activity than equivalent amount of ascorbic acid present in synthetic form. Present study is unique in a way that it investigates to improve the yield of vitamin C (ascorbic acid) from amla by comparing the methodologies of moist heat and microwave oven methods.

**MATERIALS AND METHODS**

**a. Titration method**
The vitamin C (ascorbic acid) content in moist heat and microwave oven heat treated amla were estimated by titration using iodine solution. Briefly, 10 ml of filtrate (sample) was taken in 100ml conical flask and 10 drops of 1% starch indicator solution was added. Iodine solution was added using burette into the conical flask containing the sample. The end point was noted by the appearance of blue-black complex.

**b. Preparation of fruit pulp**
Amla fruits were cut into small pieces and seeds were removed. About 200 grams of pulp alone was weighed. It was crushed by using mortar and pestle by slowly adding 200 ml of sterile distilled water. Two sets of preparations were made separately as mentioned above for moist heat (MH) and microwave oven (MO) methods.

**c. Moist Heat method (MH)**
About 50 ml of MH sample was filtered through Whatman No.1 filter paper and filtrate was collected in separate conical flask (MHC). Remaining 50 ml of MH sample was subjected to autoclave at 121°C for 15 minutes and then filtered as mentioned above (MHF). Both the samples were titrated for vitamin C content in duplicate as per the titration protocol mentioned above.

**d. Microwave Oven Method (MO)**
The MO sample was divided into four parts each weighing 30 grams and equal volume of sterile distilled water was added to all of them. The first part was kept in microwave oven (IFB 20SC2) at power 60 (60%) for 240 seconds (MO60). The second part was subjected to heat at power 80 (80%) for 140 seconds (MO80), and third part was treated at power 100 (100%) for 75 seconds (MO100). The final part of sample was not kept at microwave oven (MOC). The time noted in all experiments was based on the boiling point of the particular sample. The raise in above time durations causes boiling and leads to spillage and wastage of the sample. The samples were filtered using...
Whatman No.1 filter paper and collected the filtrate in separate conical flask. Then the samples were titrated for vitamin C content in duplicate as per the titration protocol mentioned.

RESULTS AND DISCUSSION

The vitamin C content in MHF and MHC was noted to be about 888 mg/100 g and 881 mg/100g respectively (Table 1). This shows that the effect of heat does not have any noteworthy increase in Vitamin C content as also corroborated with a report that the ascorbic acid content of fresh amla fruit itself exhibited the value of 950mg/100 gm [12]. The moist heat does not have a great impact on the vitamin C content in the amla pulp. However, vitamin C in amla was lost during cooking more in open pan than pressure cooking [13]. In our study, when compared to the moist heat exposure, the microwave oven treated samples yielded higher quantity of vitamin C and the values have gradually increased from 983mg/100g to 1283 mg/100g with increasing power (Table 2 & Fig.1). Vitamin C content of amla increases in the sun dried samples (for example 100 grams of fresh amla gives out 600 mg of Vitamin C), its content increasing from 1500 to 1600 mg [14]. The moist heat treated filtrate sample was turned into brownish colour after autoclave, whereas no colour change was observed in sample during the microwave oven treatment. Browning of amla occurs in heating at higher temperature compared to lower temperature [15]. This shows that the colour changes attributed to the decrease in vitamin C content. Hence this method will not be ideal for packaging and storing of nutraceuticals where the shelf life of the Vitamin C content has to be maintained over a period of several months.

Table 1: Vitamin C content in moist heat treated samples

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>Temperature</th>
<th>Time in minutes</th>
<th>Average Vitamin C per 100g</th>
</tr>
</thead>
<tbody>
<tr>
<td>MHC</td>
<td>NIL</td>
<td>NIL</td>
<td>881 mg</td>
</tr>
<tr>
<td>MHF</td>
<td>121°C</td>
<td>15</td>
<td>888 mg</td>
</tr>
</tbody>
</table>

Table 2: Vitamin C content in microwave oven treated samples

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>Power</th>
<th>Time in seconds</th>
<th>Average Vitamin C per 100g</th>
</tr>
</thead>
<tbody>
<tr>
<td>MOC</td>
<td>NIL</td>
<td>NIL</td>
<td>849 mg</td>
</tr>
<tr>
<td>MO60</td>
<td>60 (60%)</td>
<td>240</td>
<td>983 mg</td>
</tr>
<tr>
<td>MO80</td>
<td>80 (80%)</td>
<td>140</td>
<td>1275 mg</td>
</tr>
<tr>
<td>MO100</td>
<td>100 (100%)</td>
<td>75</td>
<td>1283 mg</td>
</tr>
</tbody>
</table>

Fig1: Increasing vitamin C content during microwave oven treatment
CONCLUSION

In conclusion, the results are encouraging to support the fact the vitamin C levels have gradually increased when heated in microwave oven corresponding to the increase in power, compared to the moist heat extraction. Hence the microwave method is more suited to extract the vitamin C. The outcome could be of translational value to the beverages and nutraceutical industry where Vitamin C levels are very crucial and should be maintained at high levels to bring the required impact in the individual as immune booster through their products.

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REFERENCES