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Effect of different chemical preservatives on storage of ash gourd juice

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Abstract
Ash gourd is an important vegetable crop for its long storage life and good scope in value addition. Juice based on ash gourd was prepared and stored by using different chemical preservatives. Therefore, the aim of the experiment was to compare the effect of different chemical additives namely Potassium metabisulfite (KMS), Sodium benzoate and their combination, on the physicochemical and antioxidant activity of ash gourd juice. The storage was done for 3 months at ambient temperature and the analysis was conducted at the interval of 0, 45 and 90 days. For the various chemical parameters like TSS, pH, acidity, ascorbic acid, reducing sugars, total sugars and antioxidants very slight changes was observed. Among different preservatives the juice preserved with KMS was found to stable during storage.

Keywords: ASH gourd juice, preservatives, KMS, sodium benzoate, storage

Introduction
Ash gourd is an under-exploited but important vegetable crop belonging to the family Cucurbitaceae grows widely in Uttar Pradesh, Madhya Pradesh, Maharashtra, Kerala, Karnataka, Andhra Pradesh, and Tamil Nadu as an annular trailing or climbing herb (Walters, 1989) [25]. Among cucurbitaceous crops ash gourd is important because it have a great potential for contributing to nutritive food requirement (Pandey *et al.*, 2015) [18]. It occurs as a native plant in Southern Asia. It is also known as winter melon, white gourd, white pumpkin, wax gourd etc. (Seshadri, 1993) [22]. According to place of origin ash gourd can be called as Petha (Hindi), Kundur (Malay), Bhuru Kolu or Safed Kolu (Gujarati), Kushmanda (Sanskrit), Donggua (Chinese) and Beligo (Indonesia) or winter melon/wax gourd (English) (Bimakar *et al.*, 2012) [2].
In North India, ash gourd is used for the preparation of petha as a sweet item, also famous as 'Agra ka Petha' and in South India, it is used as a fruit drink (Rashinkar, 2012) [21]. Ash gourd has several functional and medicinal properties especially in Ayurveda, it is recommended for treating peptic ulcer, urinary tract infections, diabetes mellitus, epilepsy and other nervous system disorders (Palamthodi and Lele, 2014) [16]. It can also prevent kidney damage (Pandey, 2008) [19]. Its fruits contain a relatively high level of K and low Na and from the index of nutritional quality value, it has been adjudged as a quality vegetable (Pandey, 2008) [19]. Ash gourd contains 67 % edible portion, 96.5 g moisture, 0.4 g protein, 0.1 g fat, 0.3 g minerals, 0.8 g fibre, 1.9 g carbohydrate, 10 kcal energy, 30 mg calcium, and 0.8 mg iron etc, respectively (Gopalan *et al.*, 2007) [5].
Ash gourd has several functional properties and used in traditional Chinese medicine to treat hypertension and inflammation (Haung *et al.*, 2004) [6]. A large number of cucurbitaceous species, which have not been exploited or are under-exploited, have a great potential for contributing to nutritive food requirement. Among them, ash gourd (*Benincasa hispida*) is an important (Pandey *et al.*, 2015) [18]. Due to the high pH level of vegetable juices, they are eminently prone to microbial contamination. Thus, it can be used as a value-added product in jam, ketchup, beverages, cakes and ice cream (Palamthodi *et al.*, 2019) [17].

Material and methods
Raw material
Well matured ash gourd fruits were procured from the local market of Hamirpur and brought to the laboratory of the Department of Food Science and Technology for studying various chemical parameters and product development.

Processing of juice

Ash gourd fruit was washed, peeled and cut into small pieces and extracted the juice by using screw type juice extractor (M/S Bajaj machinery Pvt. Ltd. Delhi) as shown in figure Figure 2.1. For conducting storage studies at an interval of 90 days the extracted juice was preserved by different methods viz, pasteurization, potassium meta-bisulphite (KMS) @2000 ppm, sodium benzoate @2000 ppm and their combinations in glass bottles at ambient temperature.

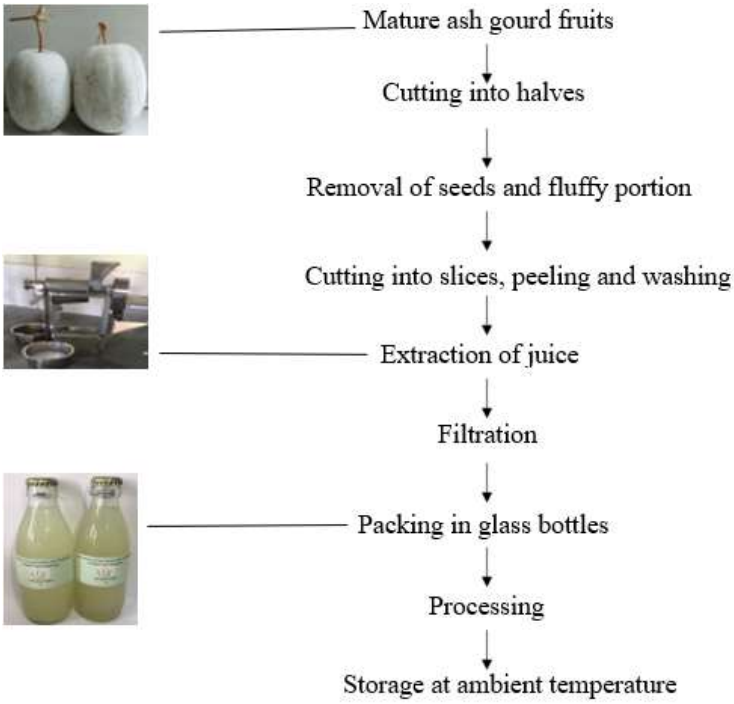

Fig 2.1: Extraction of ash gourd juice

2.3 Chemical analysis

2.3.1 Total Soluble Solids

Total soluble solids were determined by using hand refractometer (0-32 ºB). Total soluble solids of sample were measured by placing a drop of the juice on the prism of the refractometer and reading was expressed in the term of degree brix (Ranganna, 2014) [20].

2.3.2 Titratable Acidity

The titratable acidity was assessed by the standard method of Ranganna (2014) [20]. 10ml of sample was diluted with 100 ml distilled water. Further, 10ml of this aliquot was titrated against 0.1N NaOH solution to a pink end point using phenolphthalein as an indicator. The acidity as citric acid was calculated by the following expression:

$$\text{Titratable acidity (\%)} = \frac{\text{Titre value} \times \text{Normality of alkali} \times \text{Volume made} \times \text{Eq. weight of dominating acid}}{\text{Weight of sample for estimation} \times \text{Volume of aliquot} \times 1000} \times 100$$

2.3.3 pH

pH of ash gourd juice was determined using pH meter. Buffer solution of pH 7 was used for periodical calibration of pH meter.

2.3.4 Sugars

Total and reducing sugars of ash gourd juice were determined by Lane and Eynon (1923) [12] volumetric method as detailed by Ranganna (2014) [20]. The samples were prepared after using standardized method followed by titration against 10 ml of standardized Fehling's solution using methylene blue as an indicator to a brick red precipitate for determining the reducing and total sugars, respectively.

For estimation of reducing sugars, 25g sample was diluted to 100 ml with distilled water in 250 ml volumetric flask and neutralized with NaOH, using phenolphthalein as an indicator. To the neutralized sample, 2 ml of 45% lead acetate was added and kept for 10 min. Then, 2 ml of 22% potassium oxalate was added to remove excess of lead and the final volume was made up to 250 ml with distilled water and filtered. The aliquot was used for the estimation of sugars. For total sugars, 50 ml of the filtrate was hydrolysed by the addition of 1 ml HCl and was left for a period of 24 hours for inversion. The sample after inversion was neutralized with NaOH using phenolphthalein as an indicator and the volume was made to 250 ml with distilled water. The aliquot was used for titration against boiling 10 ml Fehling's solution using methylene blue as an indicator to a brick red precipitate for estimating total sugars. Total and reducing sugars expressed as percent were calculated using the following formula:

$$\text{\% Reducing sugars} = \frac{\text{mg of invert sugar x dilution}}{\text{Titre x wt. or volume of sample}} \text{ x } 100$$

1.

2. Total sugars as invert sugars (%) = calculated as in (1) making use of titre value as obtained in the determination of total sugars after inversion

2.3.5 Ascorbic acid

The ascorbic acid content was assessed by the method of Ranganna (2014) [20]. 10ml of sample (ash gourd juice) was mixed with 3% metaphosphoric acid to make 100ml by using with metaphosphoric acid followed by filtration through filter paper. 5ml of the aliquot was taken and titrated with standard dye (2, 6- dichlorophenol- indophenol dye) to a faint pink

colour end point. Ascorbic acid (mg/100ml) in the sample was calculated using following expression: -

$$\text{Ascorbic acid (mg/100g)} = \frac{\text{Titre value} \times \text{Dye factor} \times \text{Volume made}}{\text{Weight of sample taken for estimation} \times \text{volume of aliquot taken} \times 1000} \times 100$$

2.3.8 Antioxidant activity

Antioxidant activity (Free radical scavenging activity) was measured as per the method of Brand-Williams *et al.* (1995) [3]. DPPH (2, 2-diphenyl-1-picrylhydrazyl) was used as a source of free radical. A quantity of 3.9 ml of 6x10-5 mol/L DPPH in methanol was put into a cuvette with 0.1 ml of sample extract and the absorbance was measured at 515 nm after 30 min. Methanol was used as blank. Antioxidant activity was calculated using following equation: -

$$\text{Antioxidant activity (\%)} = \frac{\text{Ab(b)} - \text{Ab(s)}}{2\text{Ab (b)}} \times 100$$

Where,
Ab (B) = Absorbance of blank
Ab (S) = Absorbance of sample

2.4 Results and discussion

Ash gourd juice preserved with different chemical preservatives were evaluated for storage of 90 days at ambient temperature.

2.4.1 Effect on Total Soluble Solids (TSS)

Data presented in Table 2.1 shows that TSS content of ash gourd juice increases significantly (3.00 to 3.28 °B) in all treatments during storage period of 90 days. The mean maximum values (3.18 °B) were recorded in T1 while mean minimum (3.08 °B) in T2. The mean values range from 3.00 to 3.20 °B from 0 to 90 days**.** The interaction between different treatments and storage intervals was found to be significant (p≤0.05). An increase in TSS during storage may be due to breakdown of polysaccharides into monosaccharides. Similar increasing trend for TSS was noticed by li *et al.* (2009) [13] in longan juice, by Majumdar *et al.* (2011) [15] in bottle gourd-basil leaves juice, by Majumdar *et al.* (2012) [14] in ash gourd-mint leaves juice, by Kaur and Aggarwal (2014) [10] in bitter gourd juice preserved with different chemical preservatives.

Table 2.1 Effect of different methods of preservation on TSS of ash gourd juice

Treatment	0	45	90	Mean
T1	3.00	3.25	3.28	3.18
T2	3.00	3.09	3.14	3.08
T3	3.00	3.18	3.21	3.13
T4	3.00	3.12	3.16	3.09
Mean	3.00	3.16	3.20	
Where, T1: Pasteurization of juice at 90°C for 10 seconds followed by filling in Glass bottles T2: KMS (2000ppm) T3: Sodium benzoate (2000ppm) T4: Sodium benzoate (1000ppm) + KMS (1000ppm)				

2.4.2 Effect on titratable acidity

Data relating to change in acidity during storage are furnished in Table 2.2. which shows an increasing trend (0.13 to 0.17 %) with storage intervals from 0 to 90 days. Minimum changes were observed in treatment which was preserved with KMS while maximum in treatment T1 (pasteurization). The increase in titratable acidity might be due to the formation of organic acids by the degradation of ascorbic acid. Increase in acidity might be attributed due to breakdown of pectin into pectinic acid or due to formation of acid by the breakdown of polysaccharides or oxidation of reducing sugars. Majumdar *et al.* (2012) [14]; Kaur and Aggarwal (2014) [10] and Gajera and Joshi (2015) [4] have also observed increase in titratable acidity during storage in ash gourd-mint leaves juice, bitter gourd juice and bottle gourd based blend juice, respectively.

Table 2.2: Effect of different methods of preservation on pH of ash gourd juice

Treatment	0	45	90	Mean
T1	5.06	5.04	5.05	5.05
T2	5.06	5.03	5.02	5.04
T3	5.06	5.04	5.03	5.04
T4	5.06	5.03	5.03	5.04
Mean	5.06	5.04	5.03	
Where, T1: Pasteurization of juice at 90°C for 10 seconds followed by filling in Glass bottles T2: KMS (2000ppm) T3: Sodium benzoate (2000ppm) T4: Sodium benzoate (1000ppm) + KMS (1000ppm)				

2.4.3 Effect on pH

According to the results (Table 2.3), the interaction between different treatments as well as storage has no significant effect (p≤0.05) on pH of ash gourd juice. Among different treatments the pH values range between 5.06 to 5.02 during 90 days of storage intervals. The mean maximum values (5.05) were recorded in T1 while mean minimum (5.04) in rest of the treatments. The mean values range from 3.00 to 3.20 °B from 0 to 90 days of storage intervals. Similar decreasing trend in pH of ash gourd juice was observed by Gajera and Joshi (2015) [4] in bottle gourd based blend juice and by Majumdar *et al.* (2012) [14] in ash gourd-mint leaves juice.

Table 2.3: Effect of different methods of preservation on acidity of ash gourd juice

Treatment	0	45	90	Mean
T1	0.15	0.35	0.85	0.45
T2	0.15	0.25	0.30	0.23
T3	0.15	0.33	0.43	0.30
T4	0.15	0.27	0.33	0.25
Mean	0.15	0.30	0.48	

Where,
T1: Pasteurization of juice at 90°C for 10 seconds followed by filling in Glass bottles
T2: KMS (2000ppm)
T3: Sodium benzoate (2000ppm)
T4: Sodium benzoate (1000ppm) + KMS (1000ppm)

2.4.4 Effect on ascorbic acid content

A significant ($p \leq 0.05$) decrease in ascorbic acid content of ash gourd juice was recorded during storage period of 90 days. The mean values decrease from 7.72 to 6.67 mg/100g. The mean maximum value (7.72) was observed in treatment T2 (KMS) while minimum (6.68) in treatment T1(pasteurization). Slight decrease in ascorbic acid in juice might be due to the oxidation of irreversible conversion of L-ascorbic acid into dehydroascorbic acid oxidase caused by trapped or residual oxygen in the glass bottles (Jaiswal *et al.*, 2008) [8]. Similar trend has also been observed by Upale (2005) [24] during storage of jamun juice, by Babu (2015) in ripe pumpkin pulp and by Gajera and Joshi (2015) [4] in bottle gourd based blend juice storage.

Table 2.4: Effect of different methods of preservation on Ascorbic acid of ash gourd juice

Treatment	0	45	90	Mean
T1	8.13	6.89	4.98	6.67
T2	8.13	7.97	7.06	7.72
T3	8.13	7.87	6.47	7.49
T4	8.13	6.95	4.99	6.69
Mean	8.13	7.42	5.88	

Where,
T1: Pasteurization of juice at 90°C for 10 seconds followed by filling in Glass bottles
T2: KMS (2000ppm)
T3: Sodium benzoate (2000ppm)
T4: Sodium benzoate (1000ppm) + KMS (1000ppm)

2.4.5 Effect on reducing sugars

Reducing sugars of ash gourd juice preserved with different chemical preservatives (Table 2.5) showed a significant increase ($p \leq 0.05$) from 0.15 to 0.48 % during storage period. The mean highest value (0.45 %) was recorded in treatment T1 (pasteurization) while mean minimum (0.23%) was observed in T2 (KMS). The increase might be due to hydrolysis of starch into sugars as well as conversion of complex polysaccharides into simple sugars or inversion of non-reducing to reducing sugars. The increase in reducing sugars during storage might be due to the rapid hydrolysis of polysaccharide and their subsequent conversion to reducing sugars. Similarly, Majumdar *et al.* (2012) [14]; Babu (2015) [1] and Kadam and Lele (2016) [9] also reported an increase in reducing sugars of ash gourd-mint leaves juice, ripe pumpkin pulp and ash gourd-carrot juice during storage, respectively.

Table 2.5: Effect of different methods of preservation on reducing sugar of ash gourd juice

Treatment	0	45	90	Mean
T1	0.13	0.16	0.18	0.16
T2	0.13	0.13	0.16	0.14
T3	0.13	0.15	0.18	0.15
T4	0.13	0.14	0.16	0.14
Mean	0.13	0.14	0.17	

Where,
T1: Pasteurization of juice at 90°C for 10 seconds followed by filling in Glass bottles
T2: KMS (2000ppm)
T3: Sodium benzoate (2000ppm)
T4: Sodium benzoate (1000ppm) + KMS (1000ppm)

2.4.6 Effect on total sugars

As evinced by the data presented in Table 2.6 showed a significant increase ($p \leq 0.05$) in total sugars from 2.19 to 2.31 % of ash gourd juice during storage period of 90 days. The mean maximum value (2.34 %) was recorded in treatment T1(pasteurization) while minimum (2.27 %) in treatment T2. Increase in total sugars might be due to inversion of sucrose to glucose and fructose during storage. Similar increasing trend has been observed by Majumdar *et al.* (2011) [15] in bottle gourd-basil leaves juice and by Babu (2015) [1] in ripe pumpkin pulp storage.

Table 2.6: Effect of different methods of preservation on Total sugars of ash gourd juice

Treatment	0	45	90	Mean
T1	2.19	2.31	2.53	2.34
T2	2.19	2.29	2.34	2.27
T3	2.19	2.31	2.38	2.29
T4	2.19	2.30	2.34	2.28
Mean	2.19	2.30	2.40	

Where,
T1: Pasteurization of juice at 90°C for 10 seconds followed by filling in Glass bottles
T2: KMS (2000ppm)
T3: Sodium benzoate (2000ppm)
T4: Sodium benzoate (1000ppm) + KMS (1000ppm)

2.4.7 Effect on antioxidants

Data given in Table 2.7 reveal that antioxidants was significantly decreases ($p \leq 0.05$) in ash gourd juice (26.82 to 25.43 %) during storage period of 90 days. The mean maximum (25.64 %) changes were observed in treatment T1 (pasteurization) while minimum (26.36 %) in treatment T2 (KMS). Klimczak (2007) [11] reported that decrease in antioxidant activity during storage was attributed to a decrease in total phenolic content and vitamin C. Similar decreasing trend was noticed by Ibrahim *et al*. (2011) [7] in apple juice during storage and by Kadam and Lele (2016) [19] in ash gourd-carrot juice.

Table 2.7: Effect of different methods of preservation on Antioxidants of ash gourd juice

Treatment	0	45	90	Mean
T1	26.82	25.62	24.47	25.64
T2	26.82	26.57	25.69	26.36
T3	26.82	26.31	25.90	26.34
T4	26.82	26.56	25.67	26.35
Mean	26.82	26.27	25.43	

Where,
T1: Pasteurization of juice at 90°C for 10 seconds followed by filling in Glass bottles
T2: KMS (2000ppm)
T3: Sodium benzoate (2000ppm)
T4: Sodium benzoate (1000ppm) + KMS (1000ppm)

Conclusion

In this study, effect of different chemical preservatives on storage stability of ash gourd juice was evaluated. It is evident that ash gourd juice preserved with potassium meta-bisuphite (KMS) proved to be a better preservative than other and maintain good chemical characteristics upto 90 days of storage period.

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