

## **Standardization of preservation method and their combination for safe storage of pomegranate juice at room temperature**

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### **ABSTRACT**

The present investigation "Standardization of preservation method and their combination for safe storage of pomegranate juice at room temperature" was carried in the P.G. Research Laboratory, Department of Horticulture, Allahabad Agricultural Institute-Deemed University, Allahabad during the year 2006-2007. The experiment was laid out in 4 x 4 factorial with three replications. There were sixteen treatments comprising two factors with each of four levels. The treated juice was kept for 60 days of storage and physico-chemical characters were recorded from 0 days to 60 days of storage. In various levels of pasteurization minimum changes in T.S.S., acidity, pH, total sugar, reducing sugar and tannin were recorded with T<sub>2</sub>-70°C pasteurization while among the various levels of preservative the minimum changes were observed in P<sub>2</sub>-sodium benzoate at 500 ppm and in case of combination minimum changes were observed with T<sub>2</sub>P<sub>2</sub>-70°C pasteurization + sodium benzoate at 500 ppm. Overall minimum changes were observed in chemical preservative than pasteurization.

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**P**omegranate (*Punica granatum* L.) a small fruit tree belongs to family punicaceae. It is one of the favourite table fruits of tropical and subtropical regions where it has enjoyed the consumers patronage for its healthy dietetic and medicinal properties. It is considered a symbol of plenty and a basket of pomegranate was chosen as a symbol of plenty for the 18<sup>th</sup> International Horticulture Congress, held in 1970 (Singh, 1985). At present, pomegranate fruits are mainly used for table purpose, the freshly extracted juice of Ganesh pomegranate is light pink to pink coloured, mildly flavoured, sweet with refreshing and agreeable taste. The juice can be processed such as possibly into squash, syrup, nectar, jelly and such other products.

Preservation of fruit juice by heat is the most popular method. The method consists of essentially in heating the juice to 100°C or slightly below for a sufficient time to kill micro-organisms, which cause spoilage. While, pasteurization temperatures do not kill all the micro-organisms present in the juice. Some spores and spore bearing bacteria like *Bacillus subtilis* and *Bacillus mesentericus* can survive and multiply later. Pasteurized juice and squashes have a cooked flavour. After the container is opened they ferment and spoil within a short period, particularly in tropical climate. To avoid this, chemical preservatives are used. Chemically preserved juice can be kept for a fairly long time even after opening of the seal of the container. Sodium benzoate is a salt of benzoic acid and is used in the preservation of fruit juices

and squashes. The preservative action of benzoic acid increases in the presence of dioxide. A typical example is that of *Bacillus subtilis* which can not survive in benzoic acid solution in the presence of carbon dioxide, sodium benzoate is more effective against bacteria, yeast and moulds. The present investigation was, therefore, undertaken with a view to find out the most suitable combination of preservative for long storage of pomegranate juice at room temperature.

### **MATERIALS AND METHODS**

An experiment was carried out in P.G. Research Laboratory, Department of Horticulture, Allahabad Agricultural Institute-Deemed University, Allahabad during the year 2006-2007 for standardizing the effective method of preservation and their combination for safe storage of pomegranate juice. Pomegranate fruits of Mrig bahar were obtained from instructional-cum research orchard of the Department of Horticulture, Allahabad Agricultural Institute-Deemed University, Allahabad. The fresh fruits obtained from the garden were subjected to physico-chemical analysis as per the procedure. These fruits were employed for the juice extraction and extracted juice was treated with various levels of pasteurization viz. T<sub>0</sub>- without pasteurization, T<sub>1</sub>-60°C pasteurization, T<sub>2</sub>-70°C pasteurization, T<sub>3</sub>-80°C pasteurization and preservative viz. P<sub>0</sub>-sodium benzoate at 0 ppm, P<sub>1</sub>-sodium benzoate at 400 ppm, P<sub>2</sub>-Sodium benzoate at 500 ppm and, P<sub>3</sub>-Sodium benzoate at 600 ppm. The design of

experiment was 4 x 4 factorial design. The efficiency of the preservation method and their combination was evaluated on the basis of change in total soluble solid (T.S.S.), acidity, pH, total sugar, reducing sugar and tannin content. The initial readings were taken just after preparation of juice *i.e.* at 0 days of storage and change in various constituent of juice was recorded at various interval upto 60 days. The data recorded during the course of investigation were subjected to statistical analysis as per the method of analysis of variance technique. The significant differences between the treatment means were tested at 5% level of probability.

## RESULTS AND DISCUSSION

### *Effect on total soluble solid (%):*

The data on effect of pasteurization temperature, preservative concentration and their interaction on T.S.S. content of juice stored at room temperature found statistically significant at 30 and 60 days of storage, except 0 days of storage.

Initial T.S.S. content of juice was 15.50 per cent in various levels of pasteurization and preservative concentration. While it showed a increasing trend in all the treatments during 30 and 60 days of storage. At 60 days of storage as shown in Table 1 among the various levels of pasteurization the minimum changes were observed with  $T_2$ -70°C pasteurization and maximum changes were observed with  $T_0$ -without pasteurization. In various levels of preservative the minimum changes were observed with  $P_2$ - sodium benzoate at 500 ppm and maximum changes were observed with  $P_1$ - sodium benzoate at 400 ppm. While the juice without preservative *i.e.*  $P_0$ - sodium benzoate at 0 ppm get spoiled at 60 days of storage. Among the interaction effect minimum changes were observed in  $T_2P_2$ - 70°C pasteurization + sodium benzoate at 500 ppm followed by  $T_3P_2$ - 80°C pasteurization + sodium benzoate at 500 ppm and maximum changes were observed in  $T_0P_1$ - without pasteurization + sodium benzoate at 400 ppm. Overall minimum changes were observed in preservative method as compared to the pasteurization method.

Increase in total soluble solids was found to be associated with increase in sugars. These results are in agreement with those reported by Panlaniswami and Muthukrishnan (1974). They observed an increase in total soluble solids in lemon juice and squash during storage and Deshmukh (1991) also observed an increase in total soluble solids in pomegranate juice during storage.

### *Effect on acidity (%):*

The data on effect of pasteurization temperature,

preservative concentration and their interaction on acidity content of juice stored at room temperature were found statistically significant at 30 and 60 days of storage while initial acidity was 0.56 per cent *i.e.* non significant at 0 days of storage.

The data regarding acidity content of pomegranate juice at 60 days of storage as shown in Table 2 indicated that among the various levels of pasteurization the minimum changes observed with  $T_2$ - 70°C pasteurization and maximum changes were observed with  $T_0$ -without pasteurization. In the various levels of preservative the minimum changes were observed with  $P_2$ - sodium benzoate at 500 ppm and maximum changes were observed with  $P_1$ -sodium benzoate at 400 ppm. Among the interaction effect the minimum changes were observed in  $T_2P_2$ - 70°C pasteurization + sodium benzoate at 500 ppm followed by  $T_3P_2$ -80°C pasteurization + sodium benzoate at 500 ppm and maximum changes were observed in  $T_0P_1$ -without pasteurization + sodium benzoate at 400 ppm. Overall minimum changes were observed in preservative method as compared to the pasteurization method. The acidity content of pomegranate juice showed decreasing trend in all pasteurization temperature, preservative levels and their interaction during the storage. The decrease in acidity could be ascribed to chemical interactions between organic constituents of the juice, particularly the organic acids and anthocyanins. These findings are in agreement with those given by Panlaniswami and Muthukrishnan (1974) in lemon juice, Khurdiya and Anand (1981) in case of phalsa juice.

### *Effect on pH:*

The initial pH of the juice was 3.3, while data regarding pH of juice at 60 days of storage as shown in Table 3 indicated that among the various levels of pasteurization the minimum changes were observed with  $T_2$ -70°C pasteurization and maximum changes were observed with  $T_0$ -without pasteurization. In various levels of preservative the minimum changes were observed with  $P_2$ -sodium benzoate at 500 ppm and maximum changes were observed with  $P_1$ -sodium benzoate at 400 ppm. The interaction effect was statistically non-significant at 60 days of storage.

There was negligible change in pH of the juice during storage. Similar results were reported by Chobe (1991) in case of pomegranate juice and Patil and Jadhav (2001) in case of sweet orange juice.

### *Effect on total sugar:*

Total sugar showed an increasing trend in all the treatments. The initial total sugar was 13.50 per cent.

The data presented in Table 4 indicated that among the various levels of pasteurization the minimum changes were observed with  $T_2$ -70°C pasteurization and maximum changes were observed with  $T_0$ -without pasteurization. Among the various levels of preservative the minimum changes were observed with  $P_2$ -sodium benzoate at 500 ppm and maximum changes were observed with  $P_1$ -

sodium benzoate at 400 ppm and among the interaction effect the minimum changes were observed with  $T_2P_2$ -70°C pasteurization + sodium benzoate at 500 ppm followed by in  $T_3P_2$ -80°C pasteurization + sodium benzoate at 500 ppm and maximum changes were observed in  $T_0P_1$  – without pasteurization + sodium benzoate at 400 ppm. Overall minimum changes were observed in preservative

**Table 1 : Effect of pasteurization, preservative and their interaction on T.S.S. content (%) of pomegranate juice at 60 days of storage (Room temperature)**

Levels of Pasteurization (T) (°C)	60 DAYS				Mean (T)
	Levels of preservative (P) (Sodium Benzoate)				
	P <sub>0</sub> (0 ppm)	P <sub>1</sub> (400 ppm)	P <sub>2</sub> (500 ppm)	P <sub>3</sub> (600 ppm)	
T <sub>0</sub> (-)	0.00	16.50	16.08	16.24	16.28
T <sub>1</sub> (60°C)	0.00	16.43	15.95	16.18	16.19
T <sub>2</sub> (70°C)	0.00	16.28	15.85	16.05	16.06
T <sub>3</sub> (80°C)	0.00	16.35	15.90	16.12	16.12
Mean (P)	0.00	16.39	15.94	16.15	
		F-test	S. E. ±	C. D. (P=0.05)	
Pasteurization (T)		S	0.023	0.047	
Preservative (P)		S	0.023	0.047	
Interaction (T x P)		S	0.046	0.094	

**Table 2 : Effect of pasteurization, preservative and their interaction on acidity (%) of pomegranate juice at 60 days of storage (Room temperature)**

Levels of Pasteurization (T) (°C)	60 DAYS				Mean (T)
	Levels of Preservative (P) (Sodium Benzoate)				
	P <sub>0</sub> (0 ppm)	P <sub>1</sub> (400 ppm)	P <sub>2</sub> (500 ppm)	P <sub>3</sub> (600 ppm)	
T <sub>0</sub> (-)	0.00	0.40	0.48	0.44	0.44
T <sub>1</sub> (60°C)	0.00	0.41	0.49	0.45	0.45
T <sub>2</sub> (70°C)	0.00	0.43	0.51	0.47	0.47
T <sub>3</sub> (80°C)	0.00	0.42	0.50	0.46	0.46
Mean (P)	0.00	0.42	0.50	0.46	
		F-test	S.E.±	C. D. (P=0.05)	
Pasteurization (T)		S	0.003	0.006	
Preservative (P)		S	0.003	0.006	
Interaction (T x P)		S	0.006	0.012	

**Table 3 : Effect of pasteurization, preservative and their interaction on pH of pomegranate juice at 60 days of storage (Room temperature)**

Levels of Pasteurization (T) (°C)	60 DAYS				Mean (T)
	Levels of Preservative (P) (Sodium Benzoate)				
	P <sub>0</sub> (0 ppm)	P <sub>1</sub> (400 ppm)	P <sub>2</sub> (500 ppm)	P <sub>3</sub> (600 ppm)	
T <sub>0</sub> (-)	0.000	3.395	3.367	3.375	3.379
T <sub>1</sub> (60°C)	0.000	3.386	3.362	3.372	3.373
T <sub>2</sub> (70°C)	0.000	3.375	3.350	3.363	3.362
T <sub>3</sub> (80°C)	0.000	3.382	3.355	3.365	3.367
Mean (P)	0.000	3.385	3.359	3.369	
		F-test	S.E. ±	C. D. (P=0.05)	
Pasteurization (T)		S	0.0044	0.0091	
Preservative (P)		S	0.0044	0.0091	
Interaction (T x P)		NS	-	-	

method as compared to the pasteurization method.

Total sugar content of pomegranate juice was showed increasing trend in all pasteurization temperature, preservative concentration and their interaction during storage, increase in total sugar was found to be associated with increasing in reducing and non-reducing sugar. Similar results were obtained by Chobe (1991) in pomegranate juice and Patil and Jadhav (2001) in sweet orange juice.

#### *Effect on Reducing sugars (%):*

The data on effect of pasteurization temperature, preservative concentration and their interaction on reducing sugar content of juice stored at room temperature found statistically significant at 30 and 60 days of storage except 0 days of storage and the initial reducing sugar content of juice was 12.50 per cent.

Among the various levels of pasteurization the minimum changes were observed with T<sub>2</sub>-70°C

**Table 4 : Effect of pasteurization, preservative and their interaction on total sugar content (%) of pomegranate juice at 60 days of storage (Room temperature)**

Levels of Pasteurization (T) (°C)	60 DAYS				Mean (T)
	Levels of Preservative (P) (Sodium Benzoate)				
	P <sub>0</sub> (0 ppm)	P <sub>1</sub> (400 ppm)	P <sub>2</sub> (500 ppm)	P <sub>3</sub> (600 ppm)	
T <sub>0</sub> (-)	0.00	14.20	13.72	13.96	13.96
T <sub>1</sub> (60°C)	0.00	14.15	13.68	13.85	13.89
T <sub>2</sub> (70°C)	0.00	14.07	13.60	13.73	13.80
T <sub>3</sub> (80°C)	0.00	14.12	13.64	13.81	13.86
Mean (P)	0.00	14.14	13.66	13.84	
		F-test	S.E. (±)	C. D. (P=0.05)	
Pasteurization (T)		S	0.019	0.038	
Preservative (P)		S	0.019	0.038	
Interaction (T x P)		S	0.037	0.076	

**Table 5 : Effect of pasteurization, preservative and their interaction on reducing sugar content (%) of Pomegranate juice at 60 days of storage (Room temperature)**

Levels of Pasteurization (T) (°C)	60 DAYS				Mean (T)
	Levels of Preservative (P) (Sodium Benzoate)				
	P <sub>0</sub> (0 ppm)	P <sub>1</sub> (400 ppm)	P <sub>2</sub> (500 ppm)	P <sub>3</sub> (600 ppm)	
T <sub>0</sub> (-)	0.00	13.50	13.02	13.23	13.25
T <sub>1</sub> (60°C)	0.00	13.44	12.95	13.17	13.19
T <sub>2</sub> (70°C)	0.00	13.32	12.85	13.05	13.07
T <sub>3</sub> (80°C)	0.00	13.38	12.90	13.12	13.13
Mean (P)	0.00	13.41	12.93	13.14	
		F-test	S.E. ±	C. D. (P=0.05)	
Pasteurization (T)		S	0.018	0.036	
Preservative (P)		S	0.018	0.036	
Interaction (T x P)		S	0.036	0.073	

**Table 6 : Effect of pasteurization, preservative and their interaction on tannin content (%) of pomegranate juice at 60 days of storage (Room temperature)**

Levels of Pasteurization (T) (°C)	60 DAYS				Mean (T)
	Levels of Preservative (P) (Sodium Benzoate)				
	P <sub>0</sub> (0 ppm)	P <sub>1</sub> (400 ppm)	P <sub>2</sub> (500 ppm)	P <sub>3</sub> (600 ppm)	
T <sub>0</sub> (-)	0.000	0.220	0.264	0.238	0.241
T <sub>1</sub> (60°C)	0.000	0.225	0.265	0.245	0.245
T <sub>2</sub> (70°C)	0.000	0.238	0.275	0.255	0.256
T <sub>3</sub> (80°C)	0.000	0.233	0.270	0.250	0.251
Mean (P)	0.000	0.229	0.269	0.247	
		F-test	S.E. ±	C. D. (P=0.05)	
Pasteurization (T)		S	0.002	0.004	
Preservative (P)		S	0.002	0.004	
Interaction (T x P)		S	0.003	0.007	

pasteurization and maximum changes were observed with T<sub>0</sub>-without pasteurization (Table 5). Among the various levels of preservative the minimum changes were observed with P<sub>2</sub>-sodium benzoate at 500 ppm and maximum changes were observed with P<sub>1</sub>-sodium benzoate at 400 ppm and among the interaction effect the minimum changes were observed with T<sub>2</sub>P<sub>2</sub>-70°C pasteurization + sodium benzoate at 500 ppm followed by T<sub>3</sub>P<sub>2</sub>-80°C pasteurization + sodium benzoate at 500 ppm and maximum changes were T<sub>0</sub>P<sub>1</sub>-without pasteurization + sodium benzoate at 400 ppm.

The increase in reducing sugar content during storage could be attributed to gradual inversion of non reducing sugar to reducing sugar by hydrolysis. Panlaniswami and Muthukrishnan (1974) noticed similar behaviour of reducing sugar in lemon juice and squash. Zaric and Sanic (1979) reported similar results in pear and peach juice, they observed decrease in sucrose content and increase in reducing sugars during storage. Waskar and Khurdiya (1987) also reported similar trends of increasing reducing sugars in phalsa beverage during storage.

#### **Effect on tannin content (%):**

Tannin content showed a decreasing trend in all the treatments. The initial tannin was 0.295 per cent, while after 60 days of storage among the various levels of pasteurization the minimum changes were observed with T<sub>2</sub>-70°C pasteurization and maximum changes were observed with T<sub>0</sub>-without pasteurization (Table 6). Among the various levels of preservative the minimum changes were observed with P<sub>2</sub> sodium benzoate at 500 ppm and maximum changes were observed with P<sub>1</sub>-sodium benzoate at 400 ppm and among the interaction effect the minimum changes were observed in T<sub>2</sub>P<sub>2</sub>-70°C pasteurization + sodium benzoate at 500 ppm followed by in T<sub>3</sub>P<sub>2</sub>-80°C pasteurization + sodium benzoate at 500 ppm and maximum changes were observed in T<sub>0</sub>P<sub>1</sub>-without pasteurization + sodium benzoate at 400 ppm.

The reduction in tannin content of the juice with increasing storage period were observed in all the treatments. These findings are in agreement with those given by Bhatia *et al.* (1959) and Waskar and Khurdiya (1987).

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