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Standardization of Technology for Preparing of Ready to Serve Beverage from Pomegranate fruit

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ABSTRACT

The present investigation was undertaken to extract juices from pomegranate fruit, standardize the process for preparation of ready to serve beverage and to study the changes in quality during storage. The stored beverage was evaluated periodically at an interval of one month for chemical and sensory properties. The juice cum pulp extracted by four different methods showed that maximum recovery was obtained from TM3 (Screw type juice extractor) in respect of pomegranate Cv. Bhagwa (48.35 %) and Cv. Ganesh(49.93%). Extracted juice cum pulp was clarified by using four clarification methods and TC4 (Pectinase enzyme 2 % and incubation at 30 ºC for 4 hr and centrifugation at 10000 rpm for 15 minutes) was found best for recovery of clear juice. Pomegranate clarified juice recovery was observed in Cv. Bhagwa (91.97 %) and Cv. Ganesh (91.78% on w/w basis).

Keywords: Antioxidants, Beverage, Pomegranate, , Screw type pulper.

INTRODUCTION

Pomegranate (*Punica granatum* L) juice has shown a threefold higher antioxidant capacity than red wine or green tea (Gil *et al,* 2000) and 2, 6 and 8 fold higher capacity than those detected in grape/ cranberry, grapefruit and orange juices, respectively (Rosenblat and Aviram, 2006). Pomegranate has gained popularity because of human health building properties like anti-mutagenecity, anti-atherogenic, antioxidant, anti-hypertensive due to its high anthocyanin content (Basu and Penugonda, 2009).

The nutritive fortified drinks are now being referred as health or sport drinks. In view of medicinal and therapeutic properties of pomegranate, an attempt was made to utilize pomegranate fruit juice for preparation of ready-to-serve (RTS) beverage and to exploit its nutritive and medicinal value. Considering the increased cultivation and production of these fruits; methods of their processing technology and preservation methods were required to be developed in order to regulate the prices of produce during glut period and develop in efficient and practical methodology from pomegranate with the lowest possible cost. The present investigation was thus, carried out to study the physiochemical properties of pomegranate, to study the methods for extraction and clarification of juice and standardize the methods for the preparation of RTS beverage.

MATERIALS AND METHODS

The matured, healthy, uniform medium sized fruits of pomegranate Cv. Bhagwa and Ganesh were collected from the Department of Horticulture, Mahatma Phule Krishi Vidyapeeth, Rahuri.

Standardization of juice extraction methods

Extraction of juice

Fully matured fruits were thoroughly washed with tap water to remove surface dirt and microbial flora if any. The flow sheet for preparation of RTS beverage is given in Figure 1. Pomegranate fruits were surface sterilized with 100 ppm chlorine for 5 minutes and surface dried as suggested by Church and Parsons (1995). The pomegranate juice was extracted by four different methods of extraction such as TM1 (Manual separation), TM2 (Hydraulic basket press separator), TM3 (Screw type juice extractor) and TM4 (Brush type juice extractor).

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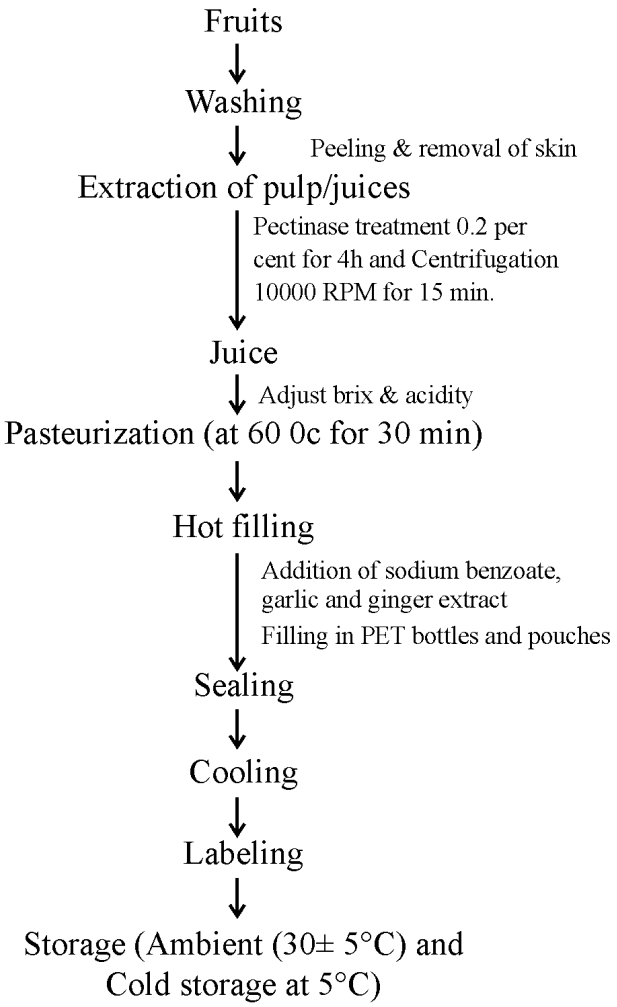
The juice recovery and clarification

The pulp /juices obtained from different extraction methods were evaluated for quality and best quality fruit juices/pulp samples obtained from one of the methods were used for further improvement of juice recovery and clarification by using different treatments such as TC1-Pectinase enzyme (0.2%) and incubation at 30ºC for 4 h and Centrifugation at 5000 rpm for 20 minutes, TC2 -Pectinase enzyme (0.2%) and incubation at 30ºC for 4 h and Centrifugation at 10000 rpm for 5 minutes, TC3 - Pectinase enzyme (0.2%) and incubation at 30ºC for 4 h and Centrifugation at 10000 rpm for 10 minutes, TC4 - Pectinase enzyme (0.2%) and incubation at 30ºC for 4 h and Centrifugation at 10000 rpm for 15 minutes. The juice recovery and clarification of fruit juices were performed as per the method suggested by Kotecha, *et al* (1995). Pectinase enzyme (0.2%) was mixed well with pomegranate juice. The clarified juice samples in the form of supernatant from two cultivars of pomegranate fruits and were used for further analysis and RTS beverage. The TC1 treatment was used as a control.

Chemical analysis of freshly prepared pomegranate fruit juice and RTS beverage

The treatments used for preparation of RTS beverage were T0- Control (Juice 15% + TSS 12°B + acidity 0.3% + 150 ppm sodium benzoate, use of chemical preservatives such as T1- Juice 15% +TSS14 °B + acidity 0.3% + 150 ppm sodium benzoate, T2-Juice 20% +TSS 14°B + acidity 0.3% + 150 ppm sodium benzoate are presented in table 3. Pomegranate juice and RTS beverage, T3 - Juice 15% +TSS 16°B + acidity 0.3 % + 150 ppm sodium benzoate, T4 - Juice 20% +TSS 16°B + acidity 0.3 % + 150 ppm sodium benzoate and natural preservatives such as T5-Juice 15%+TSS14°B +acidity 0.3 % + 50 ppm sodium benzoate)+ garlic 5ml + ginger 5ml (juice extract/lit RTS), T6-Juice 20%+TSS 14°B +acidity 0.3% + 50 ppm sodium benzoate + garlic 5ml + ginger 5ml (juice extract / lt.RTS), T7-Juice 15%+TSS 16°B +acidity 0.3% + 50 ppm sodium benzoate + garlic 5ml + ginger 5ml (juice extract / lt. RTS) and T8-Juice 20%+TSS 16°B +acidity 0.3% + 50 ppm sodium benzoate + garlic 5ml + ginger 5ml (juice extract/ lt. RTS) were analyzed for TSS, pH, acidity, total sugars, reducing sugars, anthocyanins and tannins, total antioxidant or scavenging activity (per cent) and micronutrients like Zn, Cu, Fe and Mn.

Fig 1. Flow sheet for preparation of ready-to-serve beverages



Statistical analysis

The data obtained in the present investigation were analyzed for the significance using Factorial Completely Randomized Design (FCRD) and procedure given by Panse and Sukhatme (1985).

RESULTS AND DISCUSSION

Physical characteristics of pomegranate fruits

The results of physical parameters of pomegranate fruit are presented in Table 1. It was observed that pomegranate fruits of Cv. Bhagwa were red in colour and that of Cv. Ganesh were yellowish pink in colour while both the varieties had round shaped fruits. The average weight, length and breadth of fruits of Cv. Bhagwa were 287.11 g, 6.43 cm and 7.31 cm while that of Cv. Ganesh was 322.50 g, 7.10 cm and 7.53 cm, respectively. Similar results were recorded by Dhamane *et al* (2014). The rind, seed and pulp cum juice recovery from fruits of Cv. Bhagwa was 42.21, 12.03 and 45.76 per cent while that of Cv. Ganesh was 40.38, 13.05 and 46.57 per cent, respectively. The juice of Cv. Bhagwa was very attractive in colour however the yield of juice obtained was slightly lower than Cv. Ganesh. The present findings were in accordance with the results of Bakshi *et al* (2014).

Chemical characteristics of pomegranate juice

The chemical composition of pomegranate juice is presented in Table 2. It showed that the TSS, pH, acidity, reducing sugars and total sugars of Cv. Bhagwa of pomegranate fruit were 14.30 °B, 3.32, 0.39, 9.36 and 12.27 per cent, whereas that of Cv. Ganesh were 14.10 °B, 3.10, 0.42, 9.63 and 12.13 per cent, respectively. Patil *et al* (2013) also reported the TSS of Cv. Ganesh 15 °B and acidity 0.3 per cent. The low pH of fruits helps in increasing the shelf life of product and good amount of sugar which provides better and quick source of energy. The total tannins, anthocyanin content and antioxidant activity of Cv. Bhagwa were 254.71, 85.77 mg/100 ml and 64.69 per cent where as in Cv. Ganesh, these were found to be 183.00, 81.22 mg/100 ml and 61.53 per cent, respectively. The tannins present in these fruits have proven to impart several good health benefits, for instance, they are helpful in preventing coronary heart diseases and cancer. The minerals, such as Mn, Fe, Cu and Zn in Cv. Bhagwa were found to be 0.11, 0.27, 0.07 and 0.32 mg/100 ml whereas in Cv. Ganesh these were 0.12, 0.29, 0.09 and 0.31 mg/100 ml, respectively. Results indicated that pomegranate fruits are good source of minerals and these minerals are helpful in maintaining and regulating various physiological processes in human body. These findings were in agreement with Opara *et al*(2009) and Fawole *et al* (2011) reported for pomegranate Cvs. Arkta, Ruby and Bhagwa.

Effect of methods of extraction on physico-chemical characteristics of pomegranate juice

Pomegranate is known to be a difficult fruit for processing due to the difficulty in its peeling as it is time consuming and irritating as the hands get stained due to tannins and oxidative enzymes. The basic method for extraction of juice involves the cut opening of the fruit, seed separation and pressing in

Table 1. Physical characteristics of pomegranate fruit.

Sr. No.	Parameter	Pomegranate fruit	
		Cv.Bhagwa	Cv. Ganesh
1	Rind colour of the fruits	Red	Yellow pink
2	Shape of the fruits	Round	Round
3	Average wt. of fruit (g)	287.11	322.50
4	Av.fruit length (cm)	6.43	7.10
5	Av.fruit breadth(cm)	7.31	7.53
6	Rind/Peel/pomace (%)	42.21	40.38
7	Seed (%)	12.03	13.05
8	Pulp cum juice recovery (%)	45.76	46.57

Table 2. Chemical parameters of pomegranate juice.

Sr. No.	Parameter	Pomegranate Cv	
		Bhagwa	Ganesh
1	TSS (° Brix)	14.30	14.10
2	pH	3.22	3.10
3	Acidity (%)	0.39	0.42
4	Reducing sugars (%)	9.36	9.63
5	Total sugars (%)	12.27	12.13
6	Tannins(mg/100ml)	254.71	183.00
7	Total anthocyanin content (mg/100ml)	85.77	81.22
8	Manganese(mg/100ml)	0.11	0.12
9	Iron (mg/100ml)	0.27	0.29
10	Copper (mg/100ml)	0.07	0.09
11	Zinc (mg/100ml)	0.32	0.31
12	Antioxidant activity (%)	64.69	61.53

screw press or basket press. The data on effect of various juice extraction methods on quality of juice is summarized in Table 3. The results showed that among four extraction methods TM3 (Screw type juice extractor) gave highest juice recovery on whole fruit weight basis (48.35%) and per cent aril weight basis (78.93) for Cv. Bhagwa and for Cv. Ganesh these values were 49.93 and 80.20, respectively. TM1 (manual separation) method resulted into lowest juice recovery on both whole fruit basis and aril basis. Notably, highest per cent pomace (18.25) was obtained on fruit weight basis by TM1 (manual separation) method for Cv. Bhagwa and 17.25 for Cv. Ganesh, however TM3 (Screw type juice extractor) method gave lowest per cent pomace on both weight and aril basis. Comparable results were shown by Dhamane *et al* (2014). Interestingly, the pomegranate fruit juice extraction method TM3 (screw type juice extraction) has better colour over the juice obtained from other three methods in terms of lightness L* value, redness a* value and blueness b* value which were 42.42, 19.25 and 0.47, respectively for Cv. Bhagwa, whereas for Cv. Ganesh these values were 52.14, 1.67 and 0.92, respectively. This may be due to release of more anthocyanins and other flavonoids in juice obtained by TM3 method as reported by Miguel *et al* (2004) in pomegranate juice.

The data presented in Table 4 indicated that the juice obtained by TM3 (Screw type juice extractor) method was nutritionally superior to juice obtained from other methods. It was found that juice obtained by TM3 (Screw type juice extractor) showed 14.60 °B TSS, 3.19 pH, 0.40, 10.43 and 12.50 per cent acidity, reducing sugars, total sugars, respectively and 242.3 and 44.41 mg/100ml tannins and total anthocyanin in cultivar Bhagwa, whereas 14.30 °B TSS, 3.12 pH, 0.44, 10.30 and 12.40 per cent acidity, reducing sugars, total sugars, respectively along with 175.40 and 75.40 mg/100ml tannins and total anthocyanin in cultivar Ganesh. Expectedly, the high total anthocyanin content was responsible to impart dark colour to the juice extracted from Cv. Ganesh, this observations was also reinforced by the enhanced colour parameters compared to Cv. Bhagwa. Patil (2009) clearly underline the great impact of extraction methods on physico-chemical parameters of fruit juice. The present findings were consistent with the results reported by Dhamane *et al* (2014).

Effect of clarification treatments on physico-chemical characteristics of pomegranate juice

Table 3. Effect of extraction methods on physical parameters of pomegranate juice.

Treatment	Juice recovery				Pomace/seeds obtained on fruit weight basis (%)		Juice colour					
	Whole fruit wt. basis (%)		Aril weight basis (%)				L* value		a* value		b* value	
	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG
TM1	39.43	42.72	62.72	65.70	18.25	17.25	41.46	51.11	18.08	1.50	0.43	0.83
TM2	41.20	44.60	72.60	74.50	14.21	13.90	41.58	51.32	18.21	1.53	0.42	0.87
TM3	48.35	49.93	78.93	80.20	13.35	13.10	42.42	52.14	19.25	1.67	0.47	0.92
TM4	46.40	48.10	75.10	78.43	13.80	13.50	42.14	51.98	18.92	1.54	0.43	0.89
SE +	2.11	1.64	3.46	3.76	1.13	0.95	0.23	0.25	0.28	0.04	0.01	0.02
CD at 5%	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 4. Effect of extraction methods on chemical parameters of pomegranate juice.

Treatment	TSS(°B)		pH		Acidity (%)		Reducing sugars (%)		Total sugars (%)		Tannins (mg/100g)		Total anthocyanins (mg/100g)	
	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG
TM1	14.30	14.10	3.17	3.10	0.35	0.42	10.42	10..28	12.47	12.20	240.2	162.2	82.46	70.72
TM2	14.50	14.30	3.15	3.11	0.37	0.44	10.46	10.25	12.50	12.30	241.1	172.4	83.20	73.44
TM3	14.60	14.30	3.19	3.12	0.40	0.44	10.43	10.30	12.50	12.40	242.3	175.4	84.41	75.40
TM4	14.50	14.20	3.13	3.10	0.39	0.40	10.56	10.25	12.40	12.40	215.4	146.8	81.20	71.43
SE +	0.06	0.05	0.05	0.04	0.02	0.04	0.03	0.02	0.02	0.05	6.46	6.45	1.20	1.06
CD at 5%	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Clarification or fining is one of the most important steps in fruit juice processing because it helps to remove active haze precursors and thus decrease the potential for haze formation during storage. Pomegranate juice contains only trace amount of pectin. Therefore, it can be filtered easily after pressing without clarification. However, clarification is necessary to prevent the formation of cloudy appearance during storage and also to improve the taste of the product. If the clarification is not employed, the product has bitter taste due to high tannin content. The data (Table 5) show that juice clarified by TC4 treatment (Enzyme 0.2% and incubated at 300C for 4 hr and centrifugation at 10000 rpm for 15 minutes) gave highest juice recovery (91.97 %) for Cv. Bhagwa and 91.78 per cent for Cv. Ganesh (on w/w basis). Notably, minimum 7.13 per cent sediment was obtained after clarification of juice by TC4 treatment for Cv. Bhagwa and 8.22 per cent for Cv. Ganesh among all treatments. Similar results were reported by Neifer *et al* (2009) and Song-nian *et al* (2011).The colour of juice obtained by treatment TC4 was superior in terms of lightness and L* value (35.42) for Cv. Bhagwa and 48.98 for Cv. Ganesh. Treatment TC3 was found superior in terms of redness among all methods and a* and blueness b* values obtained were 23.27and 0.40 for Cv. Bhagwa and 2.71 and 0.80, respectively for Cv. Ganesh. This could be due to reduced turbidity and viscosity of juice by addition of pectinase enzyme and its effects on pH of juice that reduces stability of anthocyanins. Pectinase enzyme plays important role in clarification of the fruit juice by depectination.

The data (Table 6) indicates that the juice obtained by clarification treatment TC1 (Enzyme 0.2% and incubated at 300C for 4 hr and centrifugation at 5000 rpm for 20 minutes), could not conserve nutritional value unlike to the juice obtained from clarification treatment TC2, TC3 and TC4. It was found that TSS (14.12 °B) was highest for Cv. Bhagwa by treatment TC2 whereas it was highest for Cv. Ganesh (14.2) by method TC3. The acidity was highest i. e. 0.39 per cent in clarified juice of for Cv. Bhagwa by treatment TC3 and for Cv. Ganesh it was 0.46 per cents. Reducing sugars were highest in juice clarified by treatment TC4 and total sugars were found highest 12.98 per cent by treatment TC2 for Cv. Bhagwa. Tannin and total anthocyanins content (mg/100ml) were 242.3 mg/100g and 82.4 mg/100g, respectively in the juice of treatment TC3 for Cv. Bhagwa, whereas for Cv. Ganesh it was 171.9 and 72.45, respectively. These results were in confirmation with the results presented by Patil *et al* (2013) and Dhamane *et al* (2014).

Standardization of ingredients levels for freshly prepared pomegranate RTS beverage

A separate trial was conducted to select optimum level of juice, TSS and acidity of the beverage. Based on sensory evaluation data for pomegranate juice by panel of semi-trained judges, the optimum level of juice, TSS and acidity were found to be 15 per cent, 16 °B and 0.30 per cent, respectively (Table 7). Various trials were made with juice levels 15 and 20 per cent juice and TSS having variation 14°B and 16 °B in either chemical or natural preservative, shows the superiority of treatments T3 and T7 with higher score for overall acceptability 8.0 and 8.1 for Cv. Bhagwa and 7.8 and 7.9 for Cv. Ganesh, respectively as compared to control which was 7.4 for Cv. Bhagwa and 7.3 for Cv. Ganesh. Based on sensory evaluation by panel of semi trained judges, the optimum level of juice, TSS and acidity were found to be 15 per cent, 16 °B and 0.30 per cent, respectively.

CONCLUSION

Thus it was concluded that the juice extracted by Screw type juice extractor was highest in pomegranate. Extracted juice clarified by Pectinase enzyme 2 per cent and incubation at 30 ºC for 4 hr and centrifugation at 10000 rpm for 15 minutes was best for recovery of clear juice. Based on sensory properties fresh RTS beverages from pomegranate juice containing 15%, TSS 14ºB and acidity 0.3% were found best in this research finding.

Table 5. Effect of clarification treatments on physical parameters of pomegranate juice.

Treatment	Juice Recovery after clarification on w/w basis (%)		Sediment obtained after clarification of juice (%)		Juice colour					
			L* value		a* value		b* value			
	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG
TC1	85.42	84.62	14.58	15.38	35.14	46.11	21.25	2.24	0.33	0.72
TC2	87.48	86.17	12.52	13.43	34.58	47.32	22.22	2.37	0.36	0.75
TC3	91.43	91.10	8.57	8.90	32.65	48.14	23.27	2.71	0.40	0.80
TC4	91.97	91.78	7.13	8.22	35.42	48.98	21.95	2.56	0.38	0.81
SE +	1.58	10.87	1.72	1.74	0.62	0.61	0.42	0.10	0.01	0.02
CD at 5 %	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 6. Effect of clarification treatments on chemical parameters of pomegranate juice

Treatment	TSS(°B)		pH		Acidity (%)		Reducing sugars (%)		Total sugars (%)		Tannin (mg/100g of fruit)		Total anthocyanins (mg/100g)	
	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG	CB	CG
TC1	14.10	14.0	3.25	3.11	0.38	0.40	10.22	10.20	12.97	12.20	220.2	160.40	78.43	69.42
TC2	14.12	14.1	3.22	3.17	0.39	0.41	10.27	10.10	12.98	12.80	241.1	168.47	82.20	70.12
TC3	14.05	14.2	3.10	3.14	0.39	0.46	10.32	10.05	12.90	12.00	242.3	171.90	82.41	72.45
TC4	13.90	14.0	3.12	3.10	0.35	0.42	10.37	10.65	12.40	12.40	235.4	164.8	79.20	72.36
SE +	0.09	0.05	0.07	0.02	0.01	0.01	0.03	0.14	0.14	0.17	5.08	6.45	1.02	0.77
CD at 5%	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 7. Sensory evaluation of freshly prepared RTS beverage from pomegranate juice.

Sr. No.	Treatment	Colour and appearance		Flavour		Taste		Overall acceptability	
		CB	CG	CB	CG	CB	CG	CB	CG
1	T0	7.5	7.4	7.3	7.2	7.3	7.2	7.4	7.3
2	T1	7.9	7.8	7.4	7.3	7.7	7.6	7.6	7.5
3	T2	7.9	7.8	7.6	7.3	7.7	7.6	7.7	7.5
4	T3	8.0	7.9	7.8	7.6	8.1	8.0	8.0	7.8
5	T4	7.9	7.8	7.7	7.5	7.7	7.6	7.7	7.6
6	T5	7.9	7.8	7.6	7.6	7.7	7.6	7.7	7.7
7	T6	7.9	7.8	7.7	7.3	7.7	7.6	7.7	7.7
8	T7	8.1	7.9	8.0	7.8	8.1	8.0	8.1	7.9
9	T8	7.9	7.8	7.8	7.4	7.7	7.6	7.6	7.6
	SE +	0.054	0.049	0.071	0.065	0.080	0.080	0.070	0.060
	CD at 5%	NS	NS	NS	NS	NS	NS	NS	NS

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