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Effect of extraction method and thermal processing on retention of bioacvtive compounds of pomegrante (*Punica granatum*) (cv. Bhagwa) juice

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ABSTRACT

The effect of juice extraction methods (from arils and halved fruits), temperature (70, 80, 90 and 100℃) and thermal processing time (2, 5 and 10 minutes) on quality of the pomegranate (*Punica granatum* L.) juice has been studied. The quality parameters studied included biochemical parameters such as ascorbic acid, total anthocyanin content, total phenolic compounds, antioxidant capacity, TSS, and acidity. The color, microbial, and sensory quality was also evaluated. The results revealed that thermal treatment reduces important bioactive compound with increase in temperature and time of processing. The thermal processing at 80℃ for 5 min. found to be suitable for reducing microbial load to below detectable limits and maintaining sensory quality up to 7.91 on ten point hedonic scale. The juice extracted from halved fruits was found to be nutritionally rich than juice extracted from arils in terms of antioxidant capacity, ascorbic acid, total phenolic compounds, total anthocyanin and color value a*. The retention of antioxidant capacity, total phenolic compounds,total anthocyanin content, ascorbic acid and color for thermal processing at 80℃ for 5 minutes for juice extracted from halved fruits was 22.68 mg/100ml AAE, 2189 mg/l GAE, 3.18 mg/100 ml, 18.33 mg/100 ml and 37.05 respectively.

Key words: Bioactive compounds, Juice, Pomegranate, Shelf life, Thermal treatment

Pomegranate (*Punica granatum* L.) juice has wide acceptability among the consumers because of nutraceutical value, attractive color, sweet sour refreshing taste, and difficulty involved in consumption of fresh fruits. The pomegranate juice contains sugars, mainly fructose and glucose, and pectin. It also contains organic acids such as ascorbic acid, citric acid, and malic acid, and bioactive compounds such as phenolics and flavonoids, with anthocyanin's being one of the most important, especially the 3-glucosides and 3, 5-diglucosides of delphinidin, cyanidin, and pelargonidin (Aviram *et al*. 2000, Tezcan *et al*. 2009 and Gil 2000).

Pomegranate juice possess a 3-fold higher antioxidant activity than that of red wine or green tea (Gil *et al*. 2000), and two, six and eight fold higher levels than those detected in grape/cranberry, grapefruit, and orange juice; respectively (Rosenblat and Aviram 2006). Pomegranate provides 12% of the Daily Value (DV) for vitamin C and 16% DV for vitamin-K per 100g serving. The pomegranate juice contains minerals such as Na, K, Fe, Cr and Cu (Heyn 1990). The antioxidant activity of the pomegranate juice can be correlated to the phenolic composition. The phytochemistry and pharmacological actions of pomegranate components such as anthocyanin's, ascorbic acid and other phenolic compounds of pomegranate suggest a wide range of clinical applications for the treatment and prevention of cancer, diseases where chronic inflammation is believed to play an essential etiologic role (Lansky and Newman 2007), cardiovascular diseases such as atherosclerosis (Avirama *et al*. 2004) and reduces systolic blood pressure (Stowe 2011, Mohan *et al*. 2010). It has been demonstrated that the consumption of this juice decreases the susceptibility of low-density lipoprotein to aggregation and retention.

The ever changing lifestyle of people and attempts to counter effects of the stressful lifestyle, and preference towards healthy and balanced eating habits has made people include fruit juices in their daily diet. India is one of the largest producer of pomegranate in world with production of 17.89 lakh tonnes in year 2014-15 (NHB). This increased production of fruits and demand for nutritious pomegranate juice has led to surge in new small scale processing units for juice. In small scale pomegranate processing unit's juice

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is extracted either from pressing arils or halved fruits and pasteurization carried in the tubular pasteurizer's (Gaikwad *et al*. 2014). Thermal processing is the most commonly used preservation technique to extend the shelf-life of juices. Thermal processing, alone or in combination with chemical or biochemical preservation techniques, is the most effective method for inactivation of microorganisms, enzymes and to increase the shelf-life in the food industry (Alighourchi *et al*. 2013). However, the bioactive compounds and nutritional quality of the juice is affected by method of juice extraction and thermal treatment.

The present research work was performed with objective to evaluate the effect of the thermal processing parameters such as temperature, time and method of juice extraction either from aril or halved fruits on the pomegranate juice quality.

MATERIALS AND METHODS

The fully matured pomegranates fruits of cv. Bhagawa grown in mrig bahar were harvested in month of December 2015 from fields of ICAR-National Research Centre on Pomegranate, Solapur, India. Only fruits of uniform size, good quality and free from pest and diseases were chosen for experimentation. Fruits were transported on the same day to the cold room in laboratory and stored at 5 °C until used for experimentation.

The healthy fruits of uniform size and appearance were selected and washed with chlorine water (200ppm sodium hypochlorite) followed by fresh water. The fruits were then separated in to two batches for juice extraction from halved fruits (designated by H) and from arils (designated by A). The fruits from first batch were cut into two halves with a sharp knife juice extracted by hydraulic press. In second batch fruits were peeled manually, arils separated and pressed for juice extraction. The extracted juice samples were immediately stored at 5°C in the dark for 24 h for settlement of sediments. The separated top portion of juice was filtered through muslin cloth and is used for further thermal treatment.

The pomegranate juice samples were heat treated at 70, 80, 90, and 100 °C ± 1°C for 2, 5, and 10 min. The untreated samples from both methods of extraction were considered as control. The temperature was depicted by first suffix (0: control, 1:70°C, 2:80°C, 3:90°C, and 4: 100°C) and treatment time was depicted by second suffix (0: control, 1: 2 min., 2: 5 min. and, 3:10 min). The juice was then packaged (crown corked) in sterilized glass bottles while still hot. The bottles were allowed to cool down to ambient temperature and stored in a cold room (5°C) until used for analysis.

The total soluble solids (TSS) of juice samples was measured using digital pocket refractometer values corrected to 20°C expressed as °Brix. Titrable acidity was measured by titrating against 0.1 N NaOH AOAC (1984) and is expressed as per cent citric acid.

Total sugar, reducing sugar of pomegranate fruits juice batches were measured using Lane and Eynon method as described by Ranganna (2000).

Ascorbic acid in pomegranate fruits juice was determined quantitatively according to the AOAC (2000) based on the reduction of 2,6 di-chlorophenol indophenol by L-ascorbic acid (Harris and Ray 1935, Sadasivam and Balasubraminan 1987).

Total phenolic compounds were determined by Folin-Ciocalteu (FC) colorimetric method which is based on chemical reduction of a mixture of tungsten and molybdenum oxides (Singleton and Rossi 1965). This method relies on the transfer of electrons in alkaline medium from phenolic compounds to a mixture of phosphomolybdic and phosphotungstic acids to form blue complexes readable by a spectrophotometer (Ainsworth and Gillespie 2007).

Total anthocyanin contents were estimated with a pH-differential method using two buffer systems: 0.025 M potassium chloride buffer at pH 1.0 and 0.4 M sodium acetate buffer at pH 4.5 (Wrolstad *et al*. 2005). The absorbance of samples was recorded at 510 and 700 nm according to the following equation.

$$A=(Abs510-Abs700)pH1-(Abs510-Abs700)pH4.5$$

The results were expressed as cyanidin-3-glucoside equivalents for pomegranate juice using a molar absorptive coefficient (ε) of 26900 L/mol.cm, molecular weight (MW) of 449.2 g/mol, dilution factor (DF), and absorption value (A), according to the following equation.

$$\text{Total anthocyanin content (mg/100ml)} = (A * MW * DF * 100)/(\varepsilon*1)$$

The FRAP method as described by Benzie and Strain (1996) was used to determine antioxidant capacity.150 µl of juice sample was added with 4.5 ml of freshly prepared FRAP (ferric reducing ability of plasma) reagent. The mix is incubated at room temperature for 30 min. and the absorbance was read at 593 nm. Standard curve developed using different concentrations of ascorbic acid (20-100µg/ml). The results were expressed as ascorbic acid equivalent antioxidant capacity (AEAC).

Aliquots of 25 ml of pomegranate juice sample were transferred into sample cup which was, covered and the color parameters were observed using a LabScan XE colorimeter according to Shwartz *et al*. (2009 a, 2009 b), and expressed in dimensions of L*, a*, b*. The average values of triplicate readings were reported for each sample. Values of L*- indicate darkness and L*+ indicate lightness of sample color, while a-indicates green color and a*+ indicates red color. The b*+ indicates a yellow color and b*- indicates blue color.

Total aerobic plate count (TPC) and total yeasts and molds(YMC) were determined on plate count agar (Patricia 1995, Vanderzant *et al*. 1992, FDA 1992) and potato dextrose agar (Vanderzant *et al*. 1992; FDA 1992), respectively using a100-µL surface plating method. PCA plates were incubated at 37°C for 48 h, and PDA plates at 25°C for 2-5 days. Results were expressed as colony-

forming units per milliliter (cfu/ml).

The juice samples were evaluated by a twenty member panel of judges on nine point hedonic scale as per the standard methods (Ranganna 2000). The parameters used for evaluation were color, flavor, and taste. The score card that represented grading of samples by the judges for different characteristics was as follows: Like extremely=9, Like very much=8, Like moderately=7, Like slightly=6, Neither like nor dislike=5, Dislike slightly=4, Dislike moderately=3, Dislike very much=2, Dislike extremely=1.

The design of experiment was laid out using factorial RBD with three replications. To find out significance of treatment ANOVA was carried out using GLM procedure of SAS 9.3. The multiple comparisons were performed using Tukey-Kramer at (P< 5%) level of significance.

RESULTS AND DISCUSSION

Effect of extraction method on biochemical constituents of fresh pomegranate juice

The biochemical constituents of fresh pomegranate juice extracted from arils and halved fruits were presented (Table 1).The method of juice extraction has significant effect on all biochemical parameters studied except on TSS which although shown higher values for juice extracted from halved fruits than arils but was not statistically significant. TSS was 16.50 and 16.63°B respectively for juice extracted from arils and halved fruits.Besides sugars, vitamins and minerals contributes to the total soluble solids content in juice. Therefore, the juice extracted from halved fruits might have contribution of minerals and vitamins from peel pressed while extracting juice. The reducing sugars were major parts 92.28% and 95.51% of total sugars present in juice extracted from arils and halved fruits respectively. Total sugars are the major soluble solid constituents of fresh pomegranate fruits juice contributing 88.72 and 81.05% of total soluble solids for juice from arils and halved fruits respectively. This also suggest that peel extract does not contribute significantly in terms of sugars to juice and thus juice from arils has higher total sugars. These results are in nearly agreement with those found by El-Nemr *et al*. (1992), Melgarejo *et al*. (2000), Al-Maiman and Ahmed (2002), Al-Said *et al*. (2009) and Janbi and Al Said (2014) with some slight variations as result of the difference of the pomegranate variety, stage of ripening, the location and climatic conditions of growing area.

The low values of titrable acidity 0.31% in juice extracted from arils were found when compared with 0.38% from halved fruits. The increase in total anthocyanin content, total phenol content, ascorbic acid, and total antioxidant capacity was 33.96, 20.70, 22.49 and 65.54 per cent respectively. The higher values of total phenol content, anthocyanin content and ascorbic acid content in juice extracted from halved fruits might be due to contribution from pressed peel. This in turn must have resulted in higher value of antioxidant capacity also. The peel as source of phenolic compounds, anthocyanin and ascorbic acid has been reported by Barros *et al.* (2014), Shiban *et al* (2012), Janbi and Al Said (2014) and Li *et al*. (2006).

Table 1 Biochemical constituent, color and microbial parameters of fresh pomegranate juice extracted from arils and halved fruits

Biochemical parameter	Juice extracted from arils (Mean±SE)	Juice extracted from halved fruits (Mean±SE)
TSS (°B)	16.50 ±0.05	16.63±0.09
Acidity (%)	0.38±0.004	0.31±0.00
Reducing sugar (%)	13.51±0.08	12.93±0.11
Non reducing sugar (%)	1.13±0.08	0.91±0.11
Total sugar (%)	14.64±0.003	13.84±0.006
Total Anthocyanin content (mg/100ml)	4.24±0.00	5.68±0.006
Total phenolic compounds (mg/l of GAE)	1826±0.33	2204±0.00
Ascorbic acid content (mg/100ml)	16.67±0.42	20.42±0.42
Total antioxidant capacity (mg/100 ml of AA)	14.80±0.011	24.50±0.00
Color L*	40.30 ±0.021	38.13±0.0265
Color a*	42.22±0.006	47.36±0.0252
Color b*	21.06±0.030	17.36±0.0100
Yeasts and molds count (log CFU/ml)	0.60±0.11	0.56±0.03
Total aerobic plate count (log CFU/ml)	2.93±0.02	2.60±0.03

Effect of extraction method and thermal treatment on biochemical parameters

The juice extracted from arils and halved fruits when subjected to different temperature and time combinations had effects on different biochemical constituents.

TSS, acidity and sugars

The pasteurization temperature and time has significant effect on TSS, reducing and non-reducing sugars. TSS, reducing sugars, non-reducing sugars and total sugar found to be increased with increase in pasteurization temperature and time (Table 2). The TSS of the juice in thermal processing has increased steadily from 16.50 and 16.63 to 18.57 and 18.67 respectively for juice from arils and halved fruits. The increase in TSS might be due to evaporation of water during thermal treatment leading to concentration of juice to some extent. The similar opinions were put forth by Dar *et al*. (1992) for apple juice, Pareek *et al*. (2011) for Nagpur mandarin juice. The titrable acidity of juice sample is significantly affected with temperature however processing time does not have significant effect on it. The reducing, non-reducing and total sugars were increased significantly from 13.51, 1.13, 14.64 and 12.93, 0.91, 13.84 for untreated juice from arils and halved fruits respectively to 16.89, 5.33, 22.22 and 16.11, 3.98, 20.09 for 100 °C and 10 min. The

Table 2 Effectof thermal treatment on biochemical constituent of pomegranate juice extracted by different means

Method of juice extraction	Pasteurization temperature (°C)	Time (min.)	Treatment	Total soluble solids (°B)	Acidity (%)	Reducing sugar (%)	Non-reducing sugar (%)	Total sugar (%)
Arils	Control	Control	A00	16.50±0.05^{n}	0.38±0.004^{a}	13.51±0.08^{fg}	1.13±0.08^{h}	14.64±0.003^{ij}
	70	2	A11	16.53±0.09^{nm}	0.38±0.004^{a}	13.89±0.09^{ef}	1.65±0.08^{ghi}	15.54±0.006^{ghij}
		5	A12	16.80±0.06^{ijkl}	0.35±0.007^{ab}	14.02±0.13^{ef}	1.85±0.20^{fghi}	15.87±0.336^{ghi}
		10	A13	16.97±0.03^{ijkl}	0.32±0.004^{c}	14.29±0.05^{cde}	2.26±0.04^{defghi}	16.55±0.003^{efgh}
	80	2	A21	16.77±0.14^{klm}	0.35±0.003^{ab}	14.02±0.13^{ef}	1.85±0.2^{fghi}	15.87±0.336^{ghi}
		5	A22	17.13±0.07^{hij}	0.35±0.004^{ab}	14.29±0.24^{cde}	2.30±0.3^{defghi}	16.59±0.372^{efgh}
		10	A23	17.60±0.10^{ef}	0.32±0.002^{c}	14.71±0.10^{cd}	3.43±0.4^{bcdef}	18.14±0.433^{cde}
	90	2	A31	17.50±0.00^{fg}	0.33±0.004^{bc}	14.29±0.05^{cde}	2.64±0.3^{cdefgh}	16.93±0.378^{defg}
		5	A32	17.97±0.03^{cd}	0.32±0.002^{c}	14.85±0.15^{c}	3.29±0.5^{bcdefg}	18.14±0.430^{cde}
		10	A33	18.20±0.11^{bc}	0.32±0.004^{bc}	15.63±0.11^{b}	4.53±0.5^{ab}	20.16±0.381^{ab}
	100	2	A41	18.00±0.06^{cd}	0.32±0.007^{bc}	14.85±0.15^{c}	3.72±0.4^{abcde}	18.57±0.438^{bcd}
		5	A42	18.21±0.03^{bc}	0.35±0.002^{bc}	15.63±0.05^{b}	3.93±0.5^{a}	19.56±0.505^{bc}
		10	A43	18.57±0.03^{a}	0.33±0.007^{bc}	16.89±0.18^{a}	5.33±0.4^{a}	22.22±0.607^{a}
Halved fruits	Control	Control	H00	16.63±0.09^{lmn}	0.31±0.00^{c}	12.93±0.11^{g}	0.91±0.11^{i}	13.84±0.006^{j}
	70	2	H11	16.69±0.007^{klmn}	0.31±0.00^{c}	13.16±0.0^{g}	1.47±0.00^{h}	14.63±0.00^{ij}
		5	H12	16.87±0.03^{jklm}	0.32±0.00^{bc}	13.51±0.0^{fg}	1.42±0.29^{h}	14.93±0.291^{hij}
		10	H13	17.13±0.03^{hij}	0.35±0.00^{abc}	14.02±0.13^{ef}	1.85±0.20^{fghi}	15.87±0.336^{ghi}
	80	2	H21	16.73±0.07^{klmn}	0.31±0.00^{c}	13.51±0.0^{fg}	1.13±0.00^{hi}	14.64±0.00^{ij}
		5	H22	17.00±0.0^{ijk}	0.32±0.00^{bc}	14.15±0.13^{de}	1.40±0.15^{h}	15.55±0.007^{ghij}
		10	H23	17.40±0.1^{fgh}	0.35±0.00^{abc}	14.43±0.14^{cde}	1.80±0.29^{fghi}	16.23±0.333^{fghi}
	90	2	H31	17.23±0.03^{ghi}	0.32±0.00^{bc}	14.29±0.0^{cde}	1.27±0.00^{hi}	15.56±0.00^{ghij}
		5	H32	17.70±0.1^{def}	0.31±0.00^{c}	14.71±0.0^{cd}	1.86±0.00^{fghi}	16.57±0.00^{efgh}
		10	H33	18.00±0.0^{cd}	0.32±0.00^{bc}	14.85±0.15^{c}	3.29±0.5^{bcdefg}	18.14±0.431^{cde}
	100	2	H41	17.90±0.0^{cde}	0.32±0.00^{bc}	14.71±0.0^{cd}	1.66±0.20^{ghi}	16.37±0.203^{fghi}
		5	H42	18.50±0.0^{ab}	0.33±0.00^{bc}	15.63±0.0^{b}	2.12±0.00^{fghi}	17.75±0.00^{efd}
		10	H43	18.67±0.03^{a}	0.33±0.00^{bc}	16.11±0.02^{ab}	3.98±0.49^{abc}	20.09±0.508^{ab}
CD (P=0.05) (n=3)				0.052	0.005	0.085	0.242	0.250

Values followed by different letters within a same column are significantly (P<0.05) different. (Where, A:juice extracted from arils, H: Juice extracted from halved fruits, the temperature was depicted by first suffix 0: control, 1:70°C, 2:80°C, 3:90°C, and 4: 100°C and treatment time was depicted by second suffix 0: control, 1: 2 min., 2: 5 min and, 3:10 min).

Table 3 Effect of method of juice extraction and thermal treatment on bioactive compounds of pomegranate juice

Method of juice extraction	Pasteurization temperature (°C)	Time (min.)	Treatment	Antioxidant capacity (mg/100 ml of AAE)	Total phenol content (mg/L of GAE)	Total anthocyanin (mg/100ml)	Ascorbic acid content (mg/100ml)
Arils	Control	Control	A00	14.80±0.01^{j}	1826.00±0.33^{g}	4.24±0.00^{bc}	16.67±0.42^{defg}
	70	2	A11	14.76±0.01^{j}	1817.00±1.15^{h}	4.07±0.00^{bcd}	16.00±0.72^{efghi}
		5	A12	14.64±0.02^{jk}	1813.00±3.33^{h}	3.60±0.01^{efd}	15.33±0.16^{ghijk}
		10	A13	14.36±0.01^{ml}	1801.00±0.66^{j}	2.87±0.003^{gh}	15.00±0.29^{ghijkl}
	80	2	A21	14.54±0.01^{kl}	1815.00±0.57^{h}	3.76±0.003^{cde}	15.83±0.417^{fghij}
		5	A22	14.24±0.01^{m}	1810.00±0.33^{i}	2.73±0.006^{ghi}	15.00±0.28^{ghijkl}
		10	A23	12.72±0.01^{o}	1798.00±0.57^{j}	2.33±0.007^{hij}	13.75±0.14^{klmn}
	90	2	A31	13.88±0.11^{n}	1808.00±1.15^{i}	3.16±0.003^{fg}	14.50±0.08^{ijklm}
		5	A32	12.64±0.006^{o}	1791.00±0.33^{k}	2.75±0.006^{ghi}	14.00±0.25^{klmn}
		10	A33	11.04±0.01^{q}	1778.00±0.33^{l}	1.82±0.00^{jk}	12.92±0.42^{mno}
	100	2	A41	11.70±0.007^{p}	1790.00±0.33^{k}	2.31±0.010^{hij}	13.33±0.42^{mnl}

Contd.

Table 3. (*Concluded*)

Method of juice extraction	Pasteurization temperature (°C)	Time (min.)	Treatment	Antioxidant capacity (mg/100 ml of AAE)	Total phenol content (mg/L of GAE)	Total anthocyanin (mg/100ml)	Ascorbic acid content (mg/100ml)
		5	A42	10.92±0.02^{q}	1768.00±0.57^{m}	1.74±0.003^{jk}	12.50±0.28^{no}
		10	A43	10.56±0.01^{r}	1712.00±0.33^{n}	0.12±0.003^{l}	11.25±0.43^{o}
Halved fruits	Control	Control	H00	24.50±0.00a	2204.00±0.00a	5.68±0.0056a	20.42±0.42a
	70	2	H11	23.78±0.00^{b}	2190.67±0.33^{b}	4.41±0.00^{b}	19.58±0.42^{ab}
		5	H12	23.73±0.02^{b}	2188.33±0.67^{bc}	4.10±0.0056^{bcd}	18.50±0.25^{bc}
		10	H13	21.56±0.02^{g}	2187.00±0.00^{bc}	3.67±0.005^{cdef}	17.67±0.17^{cde}
	80	2	H21	23.77±0.007^{b}	2189.67±0.33^{b}	4.12±0.005^{bcd}	18.75±0.00^{abc}
		5	H22	22.68±0.00^{d}	2189.00±0.00^{b}	3.18±0.005^{efg}	18.33±0.42^{bcd}
		10	H23	21.30±0.00^{h}	2184.00±0.00^{cd}	2.79±0.010^{ghi}	17.50±0.00^{cdef}
	90	2	H31	22.90±0.041^{c}	2190.00±0.00^{b}	3.73±0.005^{cdef}	17.50±0.00^{cdef}
		5	H32	22.25±0.03^{e}	2187.67±0.33^{bc}	3.16±0.005^{fg}	16.25±0.00^{efgh}
		10	H33	21.42±0.00^{gh}	2181.00±1.15^{d}	2.25±0.005^{ij}	15.00±0.00^{ghijkl}
	100	2	H41	22.06±0.00^{f}	2188.00±0.00^{bc}	2.84±0.050^{ghi}	14.58±0.42^{hijklm}
		5	H42	21.31±0.02^{h}	2162.67±0.88^{e}	2.30±0.00^{hij}	14.17±0.42^{jklmn}
		10	H43	20.85±0.08^{i}	2121.67±1.33^{f}	1.24±0.551^{k}	12.50±0.00^{no}
CD (P=0.05) (n=3)				0.027	0.699	0.085	0.255

Values followed by the different letters within a same column are significantly (P<0.05) different. (Where, A:juice extracted from arils, H: Juice extracted from halved fruits, the temperature was depicted by first suffix 0: control, 1:70ºC, 2:80ºC, 3:90ºC, and 4:100ºC and treatment time was depicted by second suffix 0: control, 1:2 min, 2:5 min and, 3:10 min).

Table 4 Effect of method of juice extraction and thermal treatment on sensory scores

Method of juice extraction	Pasteurization temperature (°C)	Time (min.)	Treatment	Taste	Flavor	Color	Overall acceptability
Arils	Control	Control	A00	9.00^{a}	9.00^{a}	8.00^{bc}	8.66^{a}
	70	2	A11	9.00^{a}	9.00^{a}	8.00^{bc}	8.66^{a}
		5	A12	8.50^{b}	9.00^{a}	8.00^{bc}	8.50^{ab}
		10	A13	8.50^{b}	9.00^{a}	8.00^{bc}	8.50^{ab}
	80	2	A21	8.50^{b}	8.50^{b}	8.00^{bc}	8.33^{ab}
		5	A22	8.50^{b}	8.25^{bc}	8.00^{bc}	8.25^{ab}
		10	A23	7.00^{e}	7.00^{e}	7.50^{dc}	7.16^{c}
	90	2	A31	7.00^{e}	7.00^{e}	7.50^{dc}	7.16^{c}
		5	A32	7.00^{e}	6.50^{f}	7.00^{de}	6.83^{cd}
		10	A33	6.50^{f}	6.00^{g}	7.00^{de}	6.50^{de}
	100	2	A41	6.00^{g}	6.00^{g}	6.50^{ef}	6.16^{ef}
		5	A42	6.025^{fg}	6.00^{g}	6.00^{f}	6.00^{ef}
		10	A43	5.00^{h}	5.00^{i}	5.00^{g}	5.00^{g}
Halved fruits	Control	Control	H00	8.00^{c}	8.00^{c}	9.00^{a}	8.33^{ab}
	70	2	H11	8.00^{c}	8.00^{c}	9.00^{a}	8.33^{ab}
		5	H12	8.00^{c}	8.00^{c}	9.00^{a}	8.33^{ab}
		10	H13	8.00^{c}	7.50^{d}	8.50^{ab}	8.00^{b}
	80	2	H21	8.00^{c}	7.50^{d}	8.50^{ab}	8.00^{b}
		5	H22	8.00^{c}	7.50^{d}	8.25^{b}	7.91^{b}
		10	H23	7.50^{d}	6.50^{f}	8.00^{bc}	7.33^{c}
	90	2	H31	7.00^{e}	7.00^{e}	7.50^{dc}	7.16^{cd}
		5	H32	6.50^{f}	6.00^{g}	7.00^{de}	6.50^{de}
		10	H33	6.00^{g}	6.00^{g}	6.50^{ef}	6.16^{ef}
	100	2	H41	6.00^{g}	5.50^{h}	7.00^{de}	6.16^{ef}
		5	H42	5.00^{h}	5.50^{h}	7.00^{de}	5.83^{f}
		10	H43	4.25^{i}	4.25^{h}	6.00^{f}	4.83^{g}

Values followed by different letters withing a same column are significantly (P<0.05) different.

effect of thermal treatment on increase in total sugars has been reported by Paul and Ghosh (2012). The values of total sugars at 90 and 100 ℃ at 10 min. for both methods of extraction had shown higher values than that of TSS. This might be because of conversion of polysaccharides into sugars. The substantial increase in sugars levels in heat processed juices during storage was also observed by Ghorai and Khurdiya (1998) which they have attributed to the inactivation of enzymes, which might play an important part in the reactions responsible for decreasing acidity and conversion of polysaccharides into simple sugars.

Antioxidant capacity, total phenol compounds, total anthocyanin and ascorbic acid content

Antioxidant capacity, total phenol compounds, total anthocyanin and ascorbic acid content have significantly reduced with temperature and time (Table 3). The antioxidant capacity has been reduced from 14.80 and 24.50 mg/100 ml AAE for raw juice from arils and halved fruits to 10.56 and 20.85 mg/100 ml AAE when pasteurized for 100 °C for 10 min. respectively (Table 3). The reduction in antioxidant capacity and total phenolic compounds found to be significant for pasteurization temperature of 80 °C and 5 min. onwards for juice from arils as well as halved fruits. Reduction in total anthocyanin and ascorbic acid has been observed over control samples with increase in temperature and time of processing. The similar observations for reduction in bioactive component with thermal treatment have been made by Paul and Ghosh (2012) and Alper *et al*. (2005).The antioxidant capacity of pomegranate juice has been positively correlated to its phenolic compounds, ascorbic acid content and anthocyanin.

Color values L, a* and b**

The color is most important quality attribute in pomegranate juice which affects purchase decision. The method extraction had significant effect on color values L*, a* and b* (Table 1). The a* values which shows redness of juice were significantly higher (47.36) for juice extracted from pressing halved fruits than that from arils (42.22) showing better color. However the L* which represents lightness was higher for juice extracted from arils than halved fruits indicates comparative darkness in juice from halved fruits due to addition of peel extract. The b* for juice from halved fruits were higher showing blueness than that from arils.

The effect of pasteurization temperature and time on juice extracted from arils and halved fruits on the color characteristics were depicted in (Fig 1). Color parameters (L*, a* and b*) shown significant degradation in values with increase in juice pasteurization temperature and time. However, the sharp colour degradation in terms of L*, a* and b* representing lightness, redness and yellowness was observed after two minutes of pasteurization time for all pasteurization temperatures. This reduction in colour values is due to degradation of color pigments in the pomegranate juice.

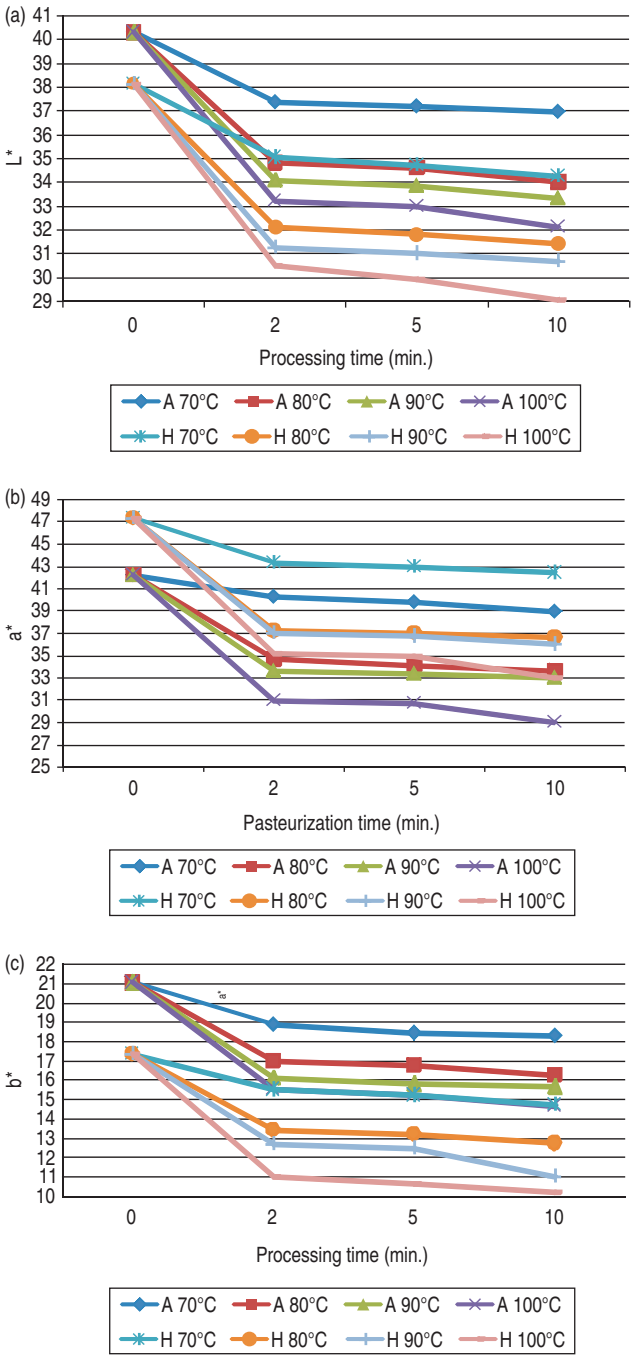

Fig 1 Changes in color value(a)L*, (b)a* and(c) b* with method of juice extraction, pasteurization temperature and time (A: Juice extracted from arils H: Juice extracted from halved fruits)

Microbial quality

The juice extracted from arils had higher bacterial and yeasts molds count than that from halved fruits pressed in press (Table 1). This might be due to higher contamination during handling required for aril extraction process. The total aerobic bacteria (TPC) and total yeasts and molds (YMC) were reduced with temperature and time. The juice extracted from arils as well as halved fruits treated at 80°C and 5 min. shown total aerobic bacterial and total yeasts and mold

count below detectable limits. Acceptable limits of total plate count and yeasts and mold count for fruit beverages is 1.69 and 0.30 log cfu/ml respectively (FSSAI 2011).

Sensory evaluation

The results revealed that the sensory score for juice extracted from arils had higher score in terms taste, flavor and overall acceptability than that from halved fruits which has higher sensory score for color (Table 4). The sensory score decreased in respect of all parameters with increase in temperature and time of thermal processing. The sensory score were higher at 80 ℃ for 5 min. for both methods of juice extraction which significantly reduced with higher time and temperature over this.

The pomegranate juice extracted from halved fruits found rich in phenols, anthocyanin, and ascorbic acid and consequently in antioxidant activity when compared with juice extracted from arils. This juice rich in bioactive compound with high antioxidant capacity will be an effective scavenger of several reactive oxygen species. The small scale pomegranate processers can go for juice extraction with halved fruits by pressing in hydraulic type of press instead of juice extraction from separated arils. The pasteurization of juice at 80 ℃ for 5 min. found to be suitable for reducing microbial load, maintaining sensory quality within acceptable limits and retention of antioxidant activity and total phenol compounds. The research findings will be useful to eschew use of steam boilers for thermal treatment at high temperature using steam instead the hot water can be used at 80°C for 5 min.

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