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RESEARCH ARTICLE

EFFECT OF BLENDING ON BIOCHEMICAL, FUNCTIONAL AND SENSORY QUALITY OF APPLE-JAMUN JUICE BLEND.

Vigya Mishra¹ and K D Sharma².

1. Amity International Centre for Post Harvest Technology and Cold Chain Management, Amity University Uttar Pradesh, Noida
2. Department of Food Science and Technology, College of Horticulture, (Dr. Y S Parmar University of Horticulture & Forestry, Nauni, Solan), Neri, Hamirpur (HP)

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Abstract

Antioxidant rich natural juice blends of apple fruit were formulated owing to its high functional value due to the presence of bioactive compounds. Suitability of the juice blends prepared by blending of Apple and Jamun juice was evaluated. Blending of Apple and Jamun juice in 90: 10 proportions gave highest antioxidant activity and sensory acceptability. The blends were filled in pre-sterilized glass bottles, processed and stored up to 6 months at 25±5°C and 4±2°C temperatures. Quality evaluation of juice blends showed significant changes in ascorbic acid, total phenolics, anthocyanins and antioxidant activity upon ambient storage. The results indicated that Apple-Jamunnatural juice blends rich in bioactive compounds having high antioxidant potential can replace the synthetic beverages besides, shall also make available a variety of natural fruit and vegetable juice choices at reasonable expenses to the domestic consumers and export purpose.

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Introduction:-

Fruits and vegetables are indeed important components in both modern and traditional healing modalities throughout the world (Krashy, 2006). It is invariably suggested to increase the intake of fruits and vegetables in human diet, as an excellent source of bioactive compounds besides fibers and minerals (Kris *et al.*, 2002). Diet rich in fruits and vegetables also provides important antioxidant phenolic phytochemicals which are supposed to exert positive effect on human health (Mikstacka *et al.*, 2010). Fruits, vegetables and their processed products in combination have synergistic effects on the antioxidant activities leading to greater reduction in risk of chronic diseases (Won and Pork, 2005). Fruit based juice beverages are one of the fastest growing segments within the beverage industry as these serve as dietary supplements and are rich in vitamins, minerals, vital micronutrients with many potential health benefits. Apple (*Malus domestica*) is an important fruit of family Rosaceae and is liked throughout the world by all classes of the people due to its established nutritional and economical significance (Chaudhary, 1994). Apple and apple juice have been reported to decrease the possibility of incidence of possibility of prostate cancer, anti influenza viral activity and the risk of chronic diseases such as cardiovascular disease and cancer (Boyer and Liu, 2004; Hamauzet *et al.*, 2005). Jamun (*Syzygiumcumini*Skeels.) is an underutilized fruit crop, gaining popularity among the consumers due to its high neutroclinical values in rural as well as in urban masses. In addition, the ripe berries are good source of anthocyanins, vitamins, minerals, iron and pectin with fair amount of ascorbic acid. It is

Corresponding Author:-Vigya Mishra.

Address:-Amity International Centre for Post Harvest Technology and Cold Chain Management, Amity University Uttar Pradesh, Noida.

used as an effective therapeutic medicine against diabetes, heart and liver trouble (Garande and Joshi, 1995). Jamun fruits are processed to make jam, jellies, squash, vinegar and ice cream for its pleasing and attractive purple colour due to presence of anthocyanins. Thus, flavonoids present in the fruit play dual role as attractive natural colorants which also add nutrition to the food and helps improve overall quality of food. In spite of high nutritive value not much break through seems to have been made in efficient utilization of Jamun fruit in the field of beverage industry (Kapoor and Ranote, 2015).

Consumption of 100 per cent pure fruit juices is an effective approach to meet the current dietary recommendations for fruit intake as well as provide essential nutrients and functional components including antioxidants therefore it can be a part of healthy active life style contrary to the synthetic beverages responsible for weight gain and chronic diseases in long run. The nutritional and therapeutic value of natural juices is far greater than that of synthetic drinks. Therefore, blending of fruit and vegetable juices could be an economic requisite to utilize profitably some underutilized fruits and vegetables for processing which may not otherwise have favourable characters such as colour, aroma and mouth feel. Blending of apple juice with astringent and highly nutritious fruits like jamun and bittergourd can provide health beverages with high medicinal and therapeutic value.

Therefore the present study was conceptualized to formulate the antioxidant rich natural juice blends of apple and jamun juice/pulp owing to their high functional value and high phytochemical content and to evaluate the functional and antioxidant potential of the developed blends.

Materials and Methods:-

Raw Materials:-

Mature fruits of apple (*Malus x domestica* Borkh.) cv. Spartan were procured from the Regional Horticultural Research Station, Seobagh (Kullu). Jamun (*Sizygium cumini* Skeels) fruits were procured from Solan Market and brought to the laboratory. Chemicals used in the experiment were of CDH brand while glassware used was of Borosil made.

Preparation of juice/pulp blends:-

Juice from apple was extracted by crushing the fruits in crusher followed by extraction using hydraulic press. Juice was heated at 82°C for 10 minutes. Jamun pulp was extracted by cooking the crushed fruits in pressure cooker for 5 min. at its own juice. Boiled fruits were passed through baby pulper and pulp was again passed through strainer. Juice and Pulp were filled hot in presterilized clean glass bottles separately and pasteurized at 90°C for 20 minutes followed by cooling to room temperature. The bottles were stored at 3-4°C till further use.

Preparation of blends:-

Apple juice and Jamun pulp were blended in proportions of 100:0, 95:5, 90:10, 85:15, 80:20, 75:25, 70:30, 65:35, 60:40 and 55:45. After sensory evaluation best blend was selected. Selected blend was prepared in required quantity and filled in pre-sterilized glass bottles (200 ml capacity) and pasteurized and stored. Final product was optimized on the basis of sensory evaluation score for different attributes and subjected to a storage study of 6 months at 25±5°C and 3±2°C.

Sensory evaluation:-

The blended beverages were evaluated for overall acceptability by a panel of 7 judges. Coded samples were served and the panelists were asked to evaluate the samples on the basis of colour (appearance), flavour, body and overall acceptability on a 9 – point Hedonic scale (Amerine et al. 1965).

Biochemical Composition:-

Total Soluble Solids:-

Total soluble solid content of fresh fruits and products was measured with the help of hand refractometer (ERMA) and was expressed as °B.

Titration Acidity:-

Titration acidity of samples was analyzed by titrating the sample against 0.1N NaOH using phenolphthalein as indicator (AOAC, 2004). Acidity was expressed in terms of per cent citric acid.

Sugars:-

Total and reducing sugars were analyzed as per the method of Lane and Eynon (1923). The total sugars content was measured as per cent.

Ascorbic acid:-

Ascorbic acid content was determined as per AOAC (1980) method by titrating the fruit extract in 3% HPO₃ up to pink endpoint, using 2, 6- dichlorophenol indophenol dye as indicator. Results were expressed as mg/100g of sample.

Total phenolics:-

The content of soluble phenols was measured using a modified Folin and Ciocalteu (1927) method using catechol as a standard. The absorbance was then measured at 660 nm using a Shimadzu UV-vis spectrophotometer (Kyoto, Japan). The results were expressed as mg/100ml, using a calibration curve over the range of 8-30µg/ml of catechol.

Free radical scavenging activity (FRSA):-

The antiradical efficiency was assessed, using the DPPH method, as described by Sánchez-Moreno *et al.* (1998). An aliquot (0.1 ml) of the sample extract was added to 3.9ml DPPH solution in methanol (6x10⁻⁵ mol/L) in a cuvette. The absorbance was measured at 515nm using the kinetic mode of the spectrophotometer at time zero and every 10s until the reaction reached the steady state plateau. The reference cuvette contained the DPPH solution only. The results of the assay were expressed in per cent. Juice blends were coloured and/or cloudy, thus for the spectrophotometric UV-vis measurements the background corrections for the absorbance are necessary.

Microbial evaluation:-

Total plate count using nutrient agar, potato dextrose agar and yeast malt extract agar media was done. Total plate count was carried out by aseptically inoculating 0.1 g of serially diluted samples in standard plate count agar medium prepared according to Ranganna (1997). An aliquot (0.1 mL) of the sample after serial dilution (10-2, 10-4, 10-6 and 10-8) was aseptically inoculated in pre-sterilized plates followed by pouring total plate count agar (10-15 mL) under sterilized environment of laminar air flow. The plates were then incubated at 37°C for 72 h prior to counting of microbes (Bacteria, yeasts and moulds). The results of the total plate count (TPC) were expressed as x 10² CFU/g of sample.

Statistical analysis:-

The data pertaining to physico-chemical characteristics of juice/blended beverage were analyzed statistically by following complete randomized block design (CRD) factorial (Cochran and Cox, 1967) while data on sensory quality of juice/blended beverage were analyzed by randomized block design (RBD) as described by Mahony (1985). Various experiments conducted in this study were replicated thrice and means were compared by using LSD (Least Significant Difference) test. For all the analyses the alpha error was set at 0.05.

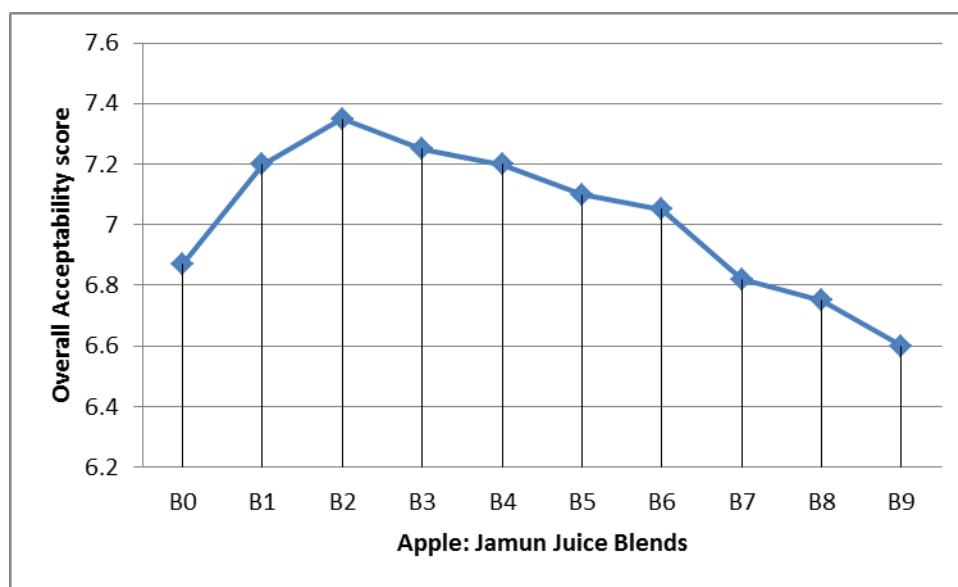
Results and Discussion:-**Physico-chemical characteristics of fresh fruits:-**

Physico-chemical characteristics of apple and Jamun fruits have been given in Table 1. TSS of apple fruits was recorded as 11.23°B. Similar results have been reported by Paul and Southgate (1978) in apple fruits. Total phenolic content of apple fruits was recorded as 365.67mg/100 g which was higher than that reported by Raj *et al.* (2011) in apple (295 mg/100 g). Antioxidant activity of apple fruits was recorded as 74.79 per cent which was quite lower than that reported by Oszmianski and Wojdylo (2009) in juices of apple fruits cultivar Champion (94.21%) and Idared (83.05%). This difference might be due to the difference in cultivar used or maturity stage of fruits. The maximum antioxidant activity and total phenolics content was recorded in Jamun fruits as 415.49mg/100g and 76.47%, respectively. The maximum total soluble solids and ascorbic acid content was recorded as 16.55°B and 22.74mg/100g, respectively in Jamun fruits. Total anthocyanins content in Jamun fruits was recorded as 157.99mg/100g. Contrary to our results Rai *et al.* (2011) reported that Jamun fruits contained 55.48 mg/100 g ascorbic acid, 1175.17 mg/100 g phenolics, 61.56% antioxidant activity and 7.25 mg total anthocyanins per 100 g of fruits. Higher content of ascorbic acid, total phenolics and presence of anthocyanins increased the free radical scavenging activity of Jamun higher than apple. Anthocyanin and phenolics rich fruits like aonla, jamun and bael are good source of dietary antioxidants. Antioxidant activity of the apple, blackcurrant and blended juices of both were strongly correlated with phenolic content by Oszmianski and Wojdylo (2009). Highest level of antioxidant activity was recorded in blackcurrant juice which was attributed to its high anthocyanins content.

Table1:-Physico-chemical properties of fresh apple and jamun fruits.

Parameters	Fruits	
	Apple	Jamun
	Fruits	
TSS (°B)	11.23± 0.37	16.55± 0.48
Acidity (%)	0.32± 0.05	0.96± 0.18
Total Sugars	10.44 ± 0.34	14.00 ± 1.23
Reducing Sugars	6.96 ± 0.30	6.81 ± 0.33
Ascorbic acid (mg/100g)	6.11± 0.61	22.74± 1.86
Total Phenolics (mg catechol/100g)	365.67± 3.45	415.49± 79.59
Antioxidant Activity (%)	74.70± 1.74	76.47± 2.99
Total anthocyanins (mg/100g)	-	157.99 ± 9.20

The data pertaining to the effect of supplementation of different proportions of Jamun juice/pulp on overall acceptability score of blended juice have been presented in Fig 1. During blending a decrease in overall acceptability scores was noticed with the increase in the proportion of Jamun pulp after a certain ratio which may be attributed to the high acidity and fibrous texture of the jamun pulp. During organoleptic test, blend containing Apple and Jamun juice/pulp in the proportion of 90:10 was adjudged best with a score of 7.35. During selection, blend having higher OAA along with higher Jamun pulp content was preferred owing to the higher nutritional and medicinal value of Jamun.

**Fig 1:-** Effect of blending of Jamun pulp on sensory scores of apple juice.

Effect of storage on quality of blends:-

TSS:-

The data pertaining to changes in quality characteristics of Apple-Jamun juice blends during storage under different storage conditions has been presented in Table 2. The data reveals that the total soluble solids increased during storage and increase was more under ambient conditions than refrigerated conditions, irrespective of treatments and storage intervals. The increase in total soluble solids may be attributed to solubilization of pulp constituents of Jamun during storage and degradation of starch into simpler sugars due to hydrolysis of polysaccharides (Jawanda *et al.* 1978) which may be higher at ambient temperature than at low temperature.

Titrateable Acidity:-

A gradual decrease was observed in titrateable acidity during storage but was statistically non-significant. The decrease in acidity might be due to chemical reactions taking place between organic acids and pigments by the action of enzymes and temperature (Kannan and Thirumarar 2001).

Table 2:- Effect of storage on quality characteristics of Apple-Jamun juice blends

Parameters	Storage Conditions	Juice/Blends (B)	Storage Interval (Days)			Mean	CD _(0.05)
			0	90	180		
TSS (°B)	AT	Control	10.9	11.13	11.21	11.08	B=1.16
		AJ	11.5	11.76	11.89	11.72	SC=NS
		Mean	11.2	11.445	11.55	11.4	
	RT	Control	10.9	10.93	11.16	11	SI=NS
		AJ	11.5	11.69	11.78	11.66	
		Mean	11.2	11.31	11.47	11.33	
Titratable Acidity (%)	AT	Control	0.3	0.31	0.33	0.31	B=NS
		AJ	0.4	0.41	0.42	0.41	SC=NS
		Mean	0.35	0.36	0.375	0.36	
	RT	Control	0.3	0.31	0.31	0.31	SI=NS
		AJ	0.4	0.4	0.41	0.4	
		Mean	0.35	0.36	0.36	0.36	
Reducing Sugars (%)	AT	Control	6.86	6.9	6.96	6.91	B=1.51
		AJ	7.77	7.83	7.87	7.82	SC=NS
		Mean	7.315	7.365	7.415	7.365	
	RT	Control	6.86	6.89	6.93	6.89	SI=NS
		AJ	7.77	7.8	7.84	7.8	
		Mean	7.315	7.345	7.385	7.345	
Total Sugars(%)	AT	Control	10.27	10.3	10.31	10.29	B=1.28
		AJ	9.87	9.91	9.96	9.91	SC=NS
		Mean	10.07	10.105	10.135	10.1	
	RT	Control	10.27	10.27	10.28	10.27	SI=NS
		AJ	9.87	9.89	9.93	9.9	
		Mean	7.315	7.345	7.385	7.345	
Ascorbic Acid (mg/100ml)	AT	Control	4.81	3.08	1.85	3.25	B=1.31
		AJ	10.59	9.25	5.14	9.92	SC=0.95
		Mean	7.7	6.165	3.495	6.585	
	RT	Control	4.81	3.68	2.73	3.74	SI= 1.14
		AJ	10.59	10.35	9.92	10.29	
		Mean	7.7	7.015	6.325	7.02	
Free Radical Scavenging Activity (FRSA)	AT	Control	72.15	71.78	70.56	71.5	B=NS
		AJ	75.15	73.73	72.28	73.72	SC=NS
		Mean	73.65	72.755	71.42	72.61	
	RT	Control	72.15	70.93	68.34	70.47	SI=1.95
		AJ	75.15	72.87	70.4	72.81	
		Mean	73.65	71.9	69.37		
Total Phenolics Content (mg/100ml)	AT	Control	198.25	173.85	131.59	167.9	B=7.24
		AJ	238.45	189.59	137.62	188.55	SC=5.12
		Mean	218.35	181.72	134.605	178.225	
	RT	Control	198.25	185.67	178.51	187.48	SI=6.27
		AJ	238.45	228.36	198.64	221.82	
		Mean	218.35	207.015	188.575	204.65	

Control: Apple juice (100%), AJ: Apple: Jamun Juice blend (90:10), B= Blends, SC= Storage Conditions, SI= Storage Intervals, AT= Ambient Temperature (25±2°C); RT= Refrigerated Temperature (2±2°C)

Sugars:-

Data shows that the total sugars decreased throughout the storage and the decrease in total sugars was more under ambient storage conditions than refrigerated, while the interaction among conditions, treatments and intervals was nonsignificant. The same trend was recorded in reducing sugars. The decrease in total sugars might be attributed to the involvement of sugars in browning reactions (Shaw *et al.*, 1977).

Ascorbic Acid:-

Blending imposed significant changes in the functional properties of blends (Table 2). An increase 220.16% was recorded after blending apple juice with jamun pulp in 90: 10 proportions. During storage a loss of 17.27% was recorded after 6 months of storage. Losses were more in blends stored under ambient storage conditions. A loss of vitamin C by 74% after 6 months of ambient storage in bottlegourd-basil leaves juice have also been reported by Majumdar *et al.* (2011). Similarly, 26.47% loss of vitamin C has been reported by Tiwari (2000) during 6 months storage of guava and papaya beverage at room temperature. Majumdar *et al.* (2009) also found remarkable loss of vitamin C (74%) during 6 months storage of cucumber-litchi-lemon juice at room temperature. Loss in ascorbic acid 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 (Deka *et al.*, 2004).

Antioxidant activity:-

Antioxidant activity (AA) of Apple: Jamun juice blends was evaluated in terms of free radical scavenging activity (FRSA). It was strongly correlated with ascorbic acid and total phenolics content. The highest antioxidant activity along with total phenolic content was recorded in AJ (Table 2). High AA of AJ blend may be attributed to its high phenolics and anthocyanins content as found in our results. Polyphenolics and anthocyanins have been reported to be the most important factors responsible for the antioxidant activity of fruits and vegetables (Oszmianski and Wojdylo, 2009; Rice-Evans *et al.*, 1997). However, nonphenolic components present in juices/pulps such as ascorbic acid might also contribute to the radical scavenging activity (Kapasakalidis *et al.*, 2006). A higher decrease in antioxidant activity was observed at refrigerated storage as compared to ambient storage. The degradation products formed during storage of juices at higher temperature may have influenced the AA of blends (Oszmianski and Wojdylo, 2009). During the processing of foods, various transformations of phenolics occur to produce yellowish to brownish pigments (Clifford, 2000). These chemical changes have been attributed to post-harvest treatments which could lead to the formation of various compounds having antioxidant and pro-oxidant properties and could exert complex effects on the antioxidant properties of phenolic compounds (Lee, 1992; Murakami *et al.*, 2002).

Total Phenolics content (TPC):-

A significant reduction in the phenolics content of juice blends was recorded during storage (Table 2). The highest phenolics content was recorded in AJ (238.45 mg/100 ml). Degradation of phenolics was higher at ambient storage as compared to refrigerated storage. In a similar study Zhang *et al.* (2008) reported no significant changes in total phenolics content of apple juice at 5 days of storage which later on decreased significantly during storage at ambient temperature. Miller *et al.* (1995) also reported the similar findings for apple juice stored at 4°C over 10 days of storage. Raj *et al.* (2010) reported a significant decrease in the phenolic content of sand pear and apple juice blends during six months of storage.

Sensory quality of blends:-

Overall acceptability of the blends was not affected significantly by storage period and conditions (Table 3). During storage lightness was recorded in the colour of AJ blend which may be due to the degradation of anthocyanins as well as sedimentation of Jamun pulp during the storage.

Microbial Examination of blends:-

During microbial examination of juice and blends, no apparent spoilage was seen. None of the isolates were found in nutrient agar, potato dextrose agar and yeast extract malt agar medium which shows that all the products were safe for consumption and maintained good microbial quality during the storage period of six month.

Table 3:-Effect of storage on overall acceptability score of Apple-Jamun juice blends.

Parameters	Storage Conditions (SC)	Juice/Blends (B)	Storage Interval (Days) (SI)			Mean	CD _(0.05)
			0	90	180		
Overall Acceptability	AT	Control	6.00	5.90	5.80	5.90	B= 0.53
		AJ	8.00	7.80	7.70	7.81	SC=NS
		Mean	7.00	6.85	6.75	6.86	
	RT	Control	6.00	5.90	5.90	5.93	SI=NS
		AJ	8.00	8.00	7.80	7.88	
		Mean	7	6.95	6.85	6.91	

Control: Apple juice (100%), AJ: Apple: Jamun Juice blend (90:10), AT= Ambient Temperature (25±2°C); RT= Refrigerated Temperature (2±°C)

Conclusion:-

Results showed that enrichment of fruit juices like apple which are deficient in many functional components, with others rich in the same components, results in products rich in antioxidants. The blending of apple and Jamun juices/pulp had a significant effect on antioxidant activity of juices. Therefore, these juices can be used as a good source of antioxidants in our diet and may be relevant in prevention of disease in which free radicals are implicated.

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