Storage Influence on Physico-Chemical Composition and Antioxidant Activity of Jamun Drink Prepared from Two Types of Pulp

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***Abstract*—The influence of different types of *Jamun* pulp was assessed by making drink in six combinations as an attempt to add value to underutilized fruit of Pakistan. pH and ascorbic acid (21.92%) decreased significantly along with phenolic contents (6.13-4.86 g of GAE/kg) and antioxidant activity (70.68-48.62 percent) till storage period of 60 days while significant increase in acidity, TSS, reducing sugars, total sugars and viscosity was observed. Statistically significant differences were determined among sensory parameters as a function of pulp type and concentration, while treatment T_5 (10% pulp with seed) was much liked by the consumer and obtained highest score (7.42+0.06).**

***Index Terms*—syzygium cumini, total phenolic contents, storage, physicochemical, antioxidant**

I. Introduction

Jamun (*Syzygium cumini* L.; family, Myrtaceae) is an underutilized fruit crop of Pakistan cultivated on area of 1338 hectare with total production of 7712 tons per annum [1]. Along with major constituents like water (83-85%), carbohydrates (12-14%), fat (0.15-0.3%) and protein (0.13-0.7%), *jamun* is also a rich source of ash, crude fiber, minerals (Ca, Mg, P, Fe, Na, Cu), thiamine, folic acid, vitamin A and C and chemo-protective nutrients [2]. *Jamun* lessens the blood glucose and hence play important role in the treatment of diabetes. The seeds of *jamun* encompass Glucoside, Jamboline and Ellagic acid, which has strong tendency to prohibit starch conversion into sugar when production of glucose in the body is in large amounts [3]. *Jamun* is highly perishable fruit, so expected storage stability is maximum 2 days when kept at ambient temperature. Typically losses of perishable fruits in developing countries like Pakistan are 10-25% due to inadequate harvesting, handling and processing [4]. External and internal conditions of fruits are responsible for post-harvest losses and affect the shelf life and quality parameters of fruits like weight loss, pH, juice contents, soluble solid contents and firmness [5].

The ripened *jamun* fruits can be used for the preparation of health drinks, preserves, squashes, jellies, nectars and wine [6]. In most countries fruit juices are known important commodities from trade point of view [7] because these provide beneficial nutrients, which are essential for human health. Moreover, nutrients are best absorbed in the form of beverages [8], consequently demand for juices is continuously increasing day by day.

Jamun fruits come in underutilized fruit category which is not commercially processed in Pakistan. The quality of *jamun* is greatly deteriorated between its harvesting and consumption so there is a considerable wastage of this fruit. To prevent this wastage and to make the *jamun* available round the year in a consumer convenient form, this practicable fruit can be converted into value added fruit drink with different types of pulp. The present research work was aimed to evaluate the best quality *jamun* drink on the basis of different pulp and seed concentration as a function of value addition. Effect of storage on the physico-chemical and phytochemical properties of *jamun* drink stored at room temperature were also measured to determine the consumer acceptability throughout the storage time.

II. Material and Methods

A. Procurement of Raw Material

Fresh *Jamun* fruit of variety *Ra* was purchased from local farm of Faisalabad, Pakistan. Fruit was divided in two lots for processing, one for juice and other for pulp preparation.

B. Pulp Preparation

The present research was carried out in National Institute of Food Science and Technology, University of Agriculture, Faisalabad-Pakistan. Quality *jamun* fruits were washed with tap water to remove dust+ and then blanched gently in boiling water for 3 minutes in order to inactivate enzymes and to soften the pulp. Two types of pulp were prepared *i.e.* pulp with seed and without seed. Seeds were removed manually for the preparation of seedless pulp while pulp with seed was obtained after grinding the whole *jamun* fruit. Both types of pulp were passed through fine pulper equipped with 0.5 mm sieve in order to remove coarse particle which may cause

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separation in drink. Obtained pulp of both types was pasteurized separately at temperature 90 ℃ for 60 seconds and filled hot in pre-sterilized glass bottles (500 mL). Caped tightly followed by processing in boiling water for 15 to 20 minutes and after properly cooling to room temperature stored at -18 ℃ for stabilization till further use.

C. Preparation of Jamun Drink

Jamun juice was prepared after manually removing the seeds from the fruit and by passing the seedless fruit from the juice extractor. Obtained juice was filtered through muslin cloth to remove suspended coarse particles in order to obtain cleared juice. Freeze pulp was used to prepare pulp supplemented *jamun* drink. *Jamun* juice after extraction was blended with pulp to prepare six different treatments *i.e.* T_1 (7% pulp without seed), T_2 (10% pulp without seed), T_3 (13% pulp without seed), T_4 (7% pulp with seed), T_5 (10% pulp with seed) and T_6 (13% pulp with seed). After pre-heating the drink at 60-70 ℃ it was homogenized and finally pasteurized at 85-90 ℃ for 60 seconds as described by [9]. Drink was hot filled at 65-75 ℃ in pre-sterilized glass bottles of 200 mL capacity and caped, which was further subjected to processing in boiling water for 15 minutes. Then bottles were cooled immediately to 20-25 ℃ to give heat shock and stored at room temperature (25-30 ℃) for further studies. Selection of final product was made on the basis of sensory evaluation for different quality aspects at regular intervals for 60 days storage.

D. Physico-Chemical Analyses

Viscosity of *jamun* drink supplemented by both types of pulp was measured at regular intervals for 2 months directly by Brookfield viscometer expressed in centipoise (cP) as described by [10]. Each sample was titrated against 0.1N NaOH till pH 8.3 to determine the titrateable acidity while pH was determined by using digital pH meter (Inolab 720, Germany) by following the method explained in [11]. Total soluble solid (TSS) contents were measured with hand refractometer (BS Eclipse 45-03, UK) by following [12] and results were directly expressed in °Brix at room temperature. The ascorbic acid contents of *jamun* drink were evaluated by using the detective dye, DCPIP (2, 6-dichlorophenolindophenol) in accordance with the prescribed method No. 967.12 of [11]. Determination of total sugar was carried out through Lane and Eynon Method as described by Rangana [12].

E. Preparation of Extract

Five milliliters of *jamun* drink was mixed with 5 mL of 80% methanol for 30 minutes at room temperature. Then transferred homogenized sample into a 100 mL volumetric flask and added 50% aqueous methanol up to the mark. The flask was covered with aluminum foil and placed in orbital shaker at 120 rpm for 45 minutes at 60 ℃. Then the solvent was removed from sample with the help of rotary evaporator (EYELA N-Series Aspirator A-3S) and filtered under suction till 1-2 mL of sample remained. Stored extracts at –20 ℃ until used.

F. Total Phenolic Contents (TPC) and FreeRadical Scavenging Activity (DPPH Assay)

Folin-Ciocalteau spectrophotometric method was used to determine TPC in samples [13] by measuring the absorbance at 765 nm with UV/Visible light spectrophotometer (Cecil CE-7200). Gallic acid was used as standard calibration curve and expressed in g of gallic acid equivalents (GAE)/per kg of sample. DPPH (1,1-diphenyl-2-picrylhydrazyl) free radical scavenging activity of *jamun* drink extract was measured by using the protocol of [14]. Antioxidant activity was calculated by using the following formula:

$$\% \text{ Inhibition (g/L) (DPPH)} = [(AB - AA) / AB] \times 100$$

AA = Absorbance of blank sample at time = 0 minutes

AA = Absorbance of tested extract solution at time = 15 minutes

G. Sensory Evaluation

Sensory evaluation of drink was carried out by train panel selected from the National Institute of Food Science and Technology, University of Agriculture Faisalabad, Pakistan using a 9-point hedonic scale (where 1=dislike extremely and 9=like extremely). Sensory attributes examined were included color, flavor, taste and overall acceptability [15]. Judges were also asked to provide additional comments based on their sensory observation.

H. Statistical Analyses

All analyses for each parameter were performed in triplicate and data presented as means $\pm$ standard deviation The results obtained for each parameter were subjected to statistical analysis, using two factor factorial analysis design at 0.05 level of significance [16].

III. Results and Discussion

A. Physico-Chemical Analyses

Mean values of viscosity varied from 2.74$\pm$0.08 to 2.85$\pm$0.01 centipoise (cP) within treatments (Table I) demonstrated that resistance to flow differed significantly by varying the concentration of both seed and without seed pulp. Maximum mean value of viscosity had been shown by the treatment T_6 (2.85$\pm$0.01 cP) and least value by T_1 (2.74$\pm$0.08 cP), which might be due to the increased fruit concentration along with seeds. The results of the present study correlated well with the findings of the storage study on the concentrated apple juice conducted by [17] who described that viscosity increased with the increase in solid contents of juices. Considerable increase in viscosity (2.80$\pm$0.04 to 2.96$\pm$0.07 cP) has been found during whole storage period as cleared from Table II.

TABLE I: EFFECT OF TREATMENT ON PHYSICO-CHEMICAL PARAMETERS OF JAMUN DRINK

Parameter	Treatments					
	T_1	T_2	T_3	T_4	T_5	T_6
pH	04.19±0.13	04.17±0.16	04.12±0.22	04.18±0.09	04.13±0.25	04.07±0.16
Acidity (%)	00.56±0.02	00.69±0.05	00.80±0.01	00.62±0.05	00.77±0.08	00.94±0.09
TSS (°B)	12.58±0.25	13.12±0.19	13.50±0.22	12.72±0.12	13.38±0.15	13.70±0.08
Viscosity (cP)	02.74±0.08	02.78±0.07	02.81±0.02	02.76±0.04	02.80±0.03	02.85±0.01

Values are expressed as mean ± standard deviation of each parameter

T_1 = 7% pulp without seed, **T_2** = 10% pulp without seed, **T_3** = 13% pulp without seed

T_4 = 7% pulp with seed, **T_5** = 10% pulp with seed, **T_6** = 13% pulp with seed

TABLE II: EFFECT OF STORAGE INTERVAL ON PHYSICO-CHEMICAL PARAMETERS OF JAMUN DRINK

Parameter	Storage (Days)				
	0	**15**	**30**	**45**	**60**
pH	05.26±0.19	05.07±0.23	04.92±0.11	04.75±0.14	04.19±0.09
Acidity (%)	00.67±0.05	00.75±0.01	00.89±0.02	01.03±0.03	01.17±0.01
TSS (°B)	12.70±0.29	12.90±0.25	13.08±0.15	13.40±0.19	13.75±0.27
Viscosity (cP)	02.80±0.04	02.82±0.11	02.86±0.03	02.93±0.02	02.97±0.07

Values are expressed as mean ± standard deviation of each parameter

Viscosity increases rapidly after 30 days storage interval which might be due to decreased juice contents as a result of hydrolytic reactions in drink utilizing dry matters such as starch, total titratable acids with the passage of time.

Data regarding pH and acidity values indicated that both theses parameters varied significantly within treatments and during storage as depicted in Table I and Table II. It was observed that with increase in pulp concentration within treatments pH decreases while acidity increased proving, acidic nature of fruit. pH and acidity values of treatments T_3 (4.12±0.22, 0.80±0.01%), T_5 (4.13±0.25, 0.77±0.16%) and T_6 (4.07±0.16, 0.94±0.09%) respectively were found statistically significantly ($p \leq 0.05$). Maximum acidity or lowest pH (Table I) was observed in T_6 due to increased concentration of pulp (13 %) with seed. pH of all six treatments decreased (5.26±0.19 to 4.19±0.09) during storage intervals of 2 months as shown in Table II with the corresponding increase in the acidity (0.67±0.05% to 1.17±0.01%). Reduction in pH value of drink is supposed to be due to increase in the acidity during storage. Increase in titratable acidity with the passage of time might be due to degradation of pectin (glacturonic acid) considerably present in drink as a fruit constituent. As a result of this degradation many acidic compounds have been formed which are responsible for the increased acidity of drink. Decrease in pH of strawberry drink had also been shown by [18], with maximum pH value 3.40 at day 0 and minimum value of 2.56 at day 90 of storage. Similar trend of acidity had been observed in literature in case of the apple and apricot blended juice [19].

Pulp concentration directly influenced the total soluble solid (TSS) contents of drink. As concentration of pulp (both without seed and with seed) increases, TSS within treatments increased correspondingly (Table I). Mean values regarding TSS showed the highest TSS (13.70+0.08 °B) of treatment T_6, while TSS contents of T_1 was found lowest *i.e.* 12.58±0.25 °B. The reason behind is that with the increase in concentration of both types of pulp, amount of suspended particles in drink increased. These particles may interfere with actual °Brix reading during TSS measurement as a result increasing trend was observed within treatments. Mean values of total soluble solids illustrated that this parameter also varied significantly during storage (12.70±0.29 to 13.75±0.27 °B). The main reason of °Brix enhancement is supposed to be due to the formation of mono and disaccharides from the break-down of polysaccharides (carbohydrates) as a result of sugar fermentation of juice or due to microbial degradation of fruit juice with the passage of time as stated by author [20]. Increasing trend in the total soluble solids from 13.00 to 13.87 °B had also been reported during storage of strawberry drink [18].

Ascorbic acid contents were significantly influenced by both *i.e.*, pulp type and its concentration as presented in Fig. 1. Highest value of ascorbic acid contents had been shown by treatment T_6 at zero day (11.59 mg/100 mL) due to high pulp with seed concentration. Comparatively the ascorbic acid contents were higher for the treatments having pulp with seed, owing to ascorbic acid contents of the seed. A decrease of 21.92% in ascorbic acid contents was observed during whole storage period (Fig. 1), due to its vulnerability to the oxidative mechanism resulting from the presence of not just oxygen, but also due to exposure to light, heat and enzymes along with storage temperature. Similar results were obtained in thermally and pasteurized pineapple juice during storage [21], [22]. Loss in ascorbic acid content of 25.65 % for hot fill and 26.74 % for aseptic method was reported during shelf life of 350 days in apple juice [23].

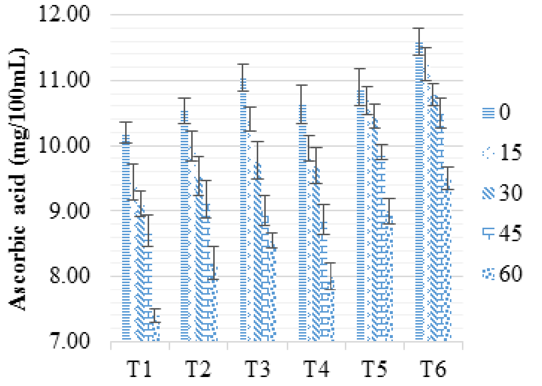

Figure 1. Effect of treatments and storage on ascorbic acid contents (mg/100mL) of *Jamun* drink during storage (25-30°C).

Total sugars varied significantly within treatments from 11.55$\pm$0.8 to 11.78$\pm$0.05g/100mL as demonstrated in Fig. 2. Minimum mean value of total sugars *i.e.* 11.45$\pm$0.1 g/100 mL (Fig. 2) had been shown by treatment T_1 and maximum by T_6 (11.71$\pm$0.2 g/100 mL respectively) as a function of increase in concentration of pulp and seed in *jamun* drink. According to mean values of storage intervals gradual increase in total sugars (11.58$\pm$0.06 to 11.83$\pm$0.04 g/100 mL) was found in *jamun* drink. This increase in total sugars could be due to conversion of oligosaccharides and other carbohydrates in di and mono-saccharides. This might also be associated with conversion of sucrose to its components as a result of high storage temperature, increased catalytic oxidation and hydrolysis of sugars at lower pH with the passage of time. Similar trend had been found in fruit based functional drinks with the passage of time [24], [25].

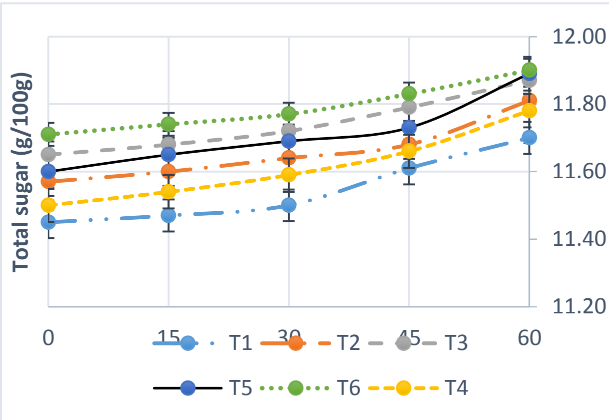

Figure 2. Effect of treatments and storage on total sugar contents (g/100mL) of *Jamun* drink during storage (25-30°C)

B. *Phytochemical Study*

Concentration of bioactive compounds such as total phenolic content (TPC) and DPPH assay (anti-oxidant activity) of supplemented *jamun* drink highly significantly affected by both types of pulp as revealed in Fig. 3 and 4. TPC gradually increased from T_1 to T_6 within treatments at zero days as well as during whole storage period (Fig. 3). Comparing the treatments with seed and without seed pulp it was clearly evident that the treatments T_3 (13% pulp without seed), T_5 (10% pulp with seed) and T_6 (13% pulp with seed) showed greater percentage of anti-oxidant activity up to 15 days storage interval (Fig. 4). After 15 days T_3 replaced by T_4 until 60 days storage period that might be associated with increased pulp concentration along with total phenolic contents of the seeds. Statistically significant ($p \leq 0.05$) decreasing trend was observed during storage from 6.13$\pm$0.33 to 4.86$\pm$0.28 g of GAE/kg and 70.68$\pm$2.83 to 48.62$\pm$1.25 percent for total phenolic contents and antioxidant activity respectively of *Jamun* drink. Treatment T_2 showed maximum changes (30.64%) in TPC followed by T_3 (27.10%), T_1 (22.36%), T_4 (21.36%), T_6 (18.33%) and T_5 (12.28%) during storage, while percent changes in antioxidant activity were found in following decreasing order: $T_1 > T_2 > T_3 > T_6 > T_4 > T_5$ within treatments.

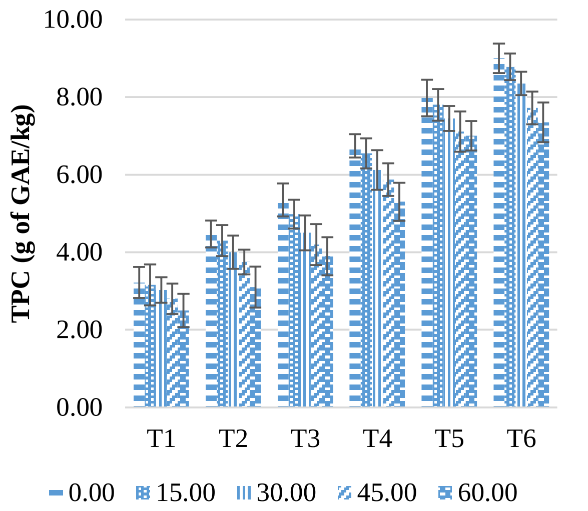

Figure 3. Effect of treatments and storage on TPC (g of GAE/kg) of *Jamun* drink during storage (25-30°C)

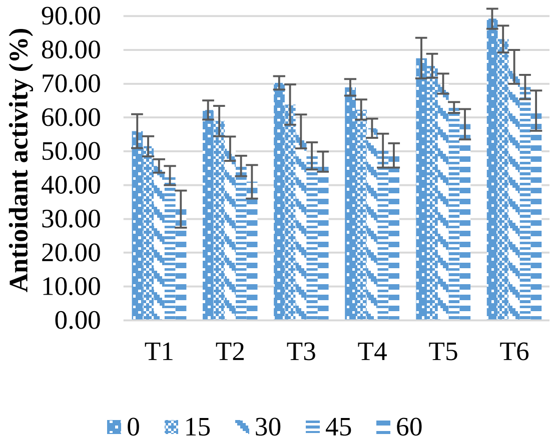

Figure 4. Effect of treatments and storage on antioxidant activity (%) of *Jamun* drink during storage (25-30°C)

The findings of the current study are in line with the findings of [26] while studying the effect of temperature and storage on the phenolic profile and antioxidant activity of grape juice. A decrease in total phenolic contents from 323.3 to 287.1 mg of GAE/100g was observed in grape juice stored at 4℃ during the storage period of two weeks. Similar findings about antioxidant activity had also been reported in literature by different authors who confirmed decrease in antioxidant activity with storage [27], [28]. A reduction of 7-35% in antioxidant activity was found during storage of pasteurized black chokeberry juice concentrates [29].

C. *Sensory Evaluation*

Sensory score for each of the following parameters like color, flavor, taste and overall acceptability in terms of treatments is shown in Fig. 5 while effect of storage at each time interval is graphically represented in Fig. 6. It is cleared from the Fig. 5 that first three treatments (T1, T2 and T3) showed increasing almost linear trend in case of each sensory parameter with the highest scores were obtained by flavor parameter. Suddenly the graph turned into zig-zag pattern after treatment T3 showing that the sensory scores for treatment T3 and T6 decreased while treatment T5 containing 10% pulp with seed got the highest points. The scores for overall acceptability was found in the following decreasing order: T5 (7.42+0.06) > T3 (7.35+0.09) T2 (7.16+0.05) > T6 (7.12+0.08) > T1 (7.10+0.10) > T4 (7.09+0.07).

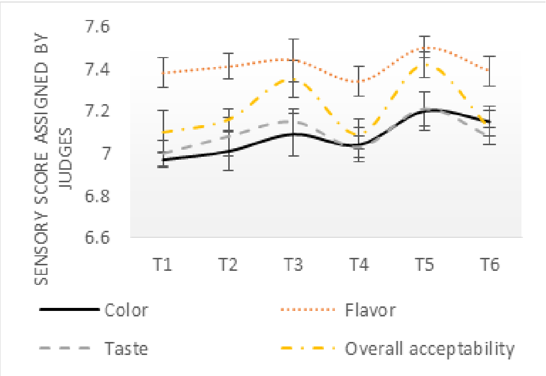

Figure 5. Effect of treatments on sensory parameters of Jamun drink

During storage the score for color, flavor, taste and overall acceptability varied from 7.35±0.10 to 6.84±0.06, 7.28±0.06 to 6.88±0.04, 7.63±0.07 to 7.22±0.05 and 7.43±0.04 to 6.97±0.09 respectively as shown in Fig. 6. The treatments T_5 (10% pulp with seed) gained maximum sore for all above mentioned parameters. This treatment remained acceptable throughout the storage period and no signs of spoilage appeared during that period. An overall significantly decreasing trend was observed among all the treatments during storage. Based on judges remarks, slight separation on treatments containing seed of *jamun* added negative impact on color, although consumer much liked their taste and flavor comparative to treatments containing pulp without seed. A diminution in sensory score may be accredited to the increase in acidity and browning reaction between reducing sugars and amino acids during storage of drinks. In a similar study author reported that the loss of sensory parameters may be due to the degradation of ascorbic acid and furfural production during storage [24], [30].

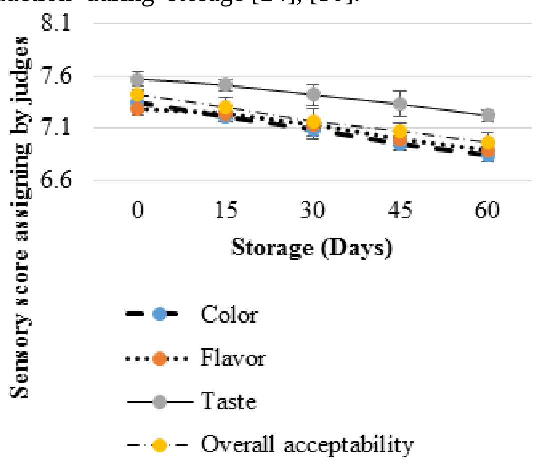

Figure 6. Effect of storage on sensory parameters of Jamun drink

IV. Conclusion

The present study was an attempt to preserve this nutrient rich and underutilized fruit of Pakistan and to make consumer acceptable drink by utilizing its non-edible seed as well. Apart from changes in physicochemical and phytochemicals, sensory evaluation suggested that T_5 (10% pulp with seed) was best because of acceptable astringic taste given by *jamun* seed. For the first time in Pakistan at pilot scale, this study showed an alternate mean to preserve this minor and neglected fruit as an attempt to preserve in the form of consumer acceptable drink. Nonetheless, further research is needed to explore the functional and health benefits of *jamun* drink with storage to win the consumer confidence.

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