

## Storage Effects on Vitamin C, Anthocyanins, Total Phenolics and Antioxidant Capacity of Processed Jamun Fruit Products

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### ABSTRACT

*Jamun (Syzygium cumini L.) is an indigenous minor fruit of India. This belongs to family Myrtaceae. Processed products of jamun fruits gaining popularity among the consumers due to nutritional and antioxidant properties of fruits. The retention of antioxidant and phenolic properties of the processed products were studied at room and refrigeration temperature. Loss of vitamin C, anthocyanins, total phenolics and antioxidant activity was observed in processed products of jamun fruits like jam, squash and nectar of different variations; during storage period of 90 days at room and refrigeration temperature. Retention of vitamin C, total anthocyanins, total phenolics and antioxidant activity was observed in refrigerated temperature than at room temperature. Higher values of vitamin C was noticed in nectar (9.86-9.8mg/100g) followed by jam (7.7- 7.3mg/100g) and squash (3.65-3.59mg/100g). Better retention of anthocyanin content was observed in jam (132-128 mg/100g) followed by squash (128-120 mg/100g) and nectar (97-85 mg/100g); Squash had higher values for total phenolic content (300-289mg/100g GAE) followed by nectar (260-252mg/100g GAE) and jam (211mg/100g GAE). Significant decline in antioxidant activity of jam (73-23mg/100g of vitamin C equivalents), squash (82-28mg/100g of vitamin C equivalents) and nectar (85-32mg/100g of vitamin C equivalents) were observed at room temperature. Results of the storage study indicate that reduction of antioxidant activity, total phenolic content, total anthocyanins and vitamin C content, during storage at room temperature can be prevented by storing the jamun fruit products at refrigeration temperature.*

**Key words:** Minor fruit, processed products, antioxidant capacity, anthocyanins, vitamin C.

### INTRODUCTION

Jamun (*Syzygium cumini* L.) is an indigenous minor fruit of India. Which belongs to family Myrtaceae commonly known as jamun, jaman, duhat (Hindi) and black plum, Indian black berry, jambolan, jambolan plum, javaplum, Malabar plum, Portuguese plum in English<sup>1</sup>.

The edible pulp of plant forms 75 per cent of the whole fruit. Various minerals and vitamins were reported like Ca, Mg, P, Fe, Na, K, Cu, S, Cl, vitamins C, vitamin A, riboflavin, nicotinic acid, choline and folic acid.

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Glucose and fructose are the principle sources of sweeteners in ripe fruit with no trace of sucrose. Maleic acid is the major acid (0.59% of the weight of fruit). Small quantity of oxalic acid has been reported. Tannins mainly gallic acid is responsible for the astringency activity is due to efficiency to combine with tissues and proteins and precipitate them. Tannins are also efficient for gastro protective and antiulcerogenic activity. The purple colour of the fruit is due to presence of one or two cyanidin diglycosides. The cyanidin glycoside belongs to the category of anthocyanidins and their glycosides<sup>2</sup>.

*Syzygium cumini* fruit has proved medicinal properties against a number of diseases; it has a high economic value for the application in the food processing industries. Different types of products available in the Asian market are jam, jellies, seed powder, pulp powder, vinegar, wine, ice cream, juice, squash.

Jambolan it is also used as antioxidant, anti-inflammatory, neuropsychopharmacological, anti-microbial, anti-bacterial, anti-HIV, antileishmarial and antifungal, nitric oxide scavenging, antidiarrheal, antifertility, anorexigenic, gastro protective and antiulcerogenic, behavioural effects and radio protective activities<sup>3</sup>.

It is getting importance due to its nutritional and health benefits. The fruit is reported to be a good source of minerals, vitamin C, sugars, phenolic compounds (Gallic acid, tannins, flavonoids, anthocyanins) and other antioxidant components<sup>4,5,6</sup>.

Vitamin C is one of the most important in human diet and plays a large role in hundreds of the body's functions. The most plentiful tissue in the body is collagen, which is a connective tissue. The primary role of Vitamin C is to help this connective tissue. Because collagen is the defense mechanism against disease and infection and because Vitamin C helps build collagen, it makes sense that it is also a remedy for scurvy by contributes to haemoglobin production. Even in small amounts, vitamin C can protect indispensable molecules in the body, such as

proteins, lipids (fats), carbohydrates and nucleic acids (DNA and RNA) from damage by free radicals and reactive oxygen species that can be generated during normal metabolism as well as through exposure to toxins and pollutants (e.g. smoking). Vitamin C may also be able to regenerate other antioxidants such as vitamin E<sup>7,8</sup>. Anthocyanins act as phytochemical antioxidants with potential health related benefits. Some positive therapeutic effects of Anthocyanins are more or less related to the antioxidant mechanisms.

Antioxidants are molecules whose main function is to scavenge the free radicals. They can scavenge the free radicals even before they generated. Antioxidants actually tries to upgrade defense mechanism of the body the most common natural source of antioxidants are plants, mainly present in fruits and vegetables in the form of polyphenolics compounds which are mainly flavonoids. It is particularly important for those people who do not consume enough fruits and vegetables in their daily diet.

Phytochemicals have recently been studied for their positive health benefits and has attracted great attention researchers and consumers<sup>9</sup>. These compounds play a crucial role in preventing chronic diseases<sup>10</sup>. Foods containing high concentration of antioxidants are effective in prevention of cardiovascular diseases, cancers<sup>11</sup> and neurodegenerative diseases<sup>12</sup> as well as inflammation and problems caused by cell and cutaneous aging<sup>13</sup>.

The present study was aimed to observe the changes in vitamin C, anthocyanins, total phenolics and antioxidant activity of processed products of jamun fruits which were stored for period of 90 days under room and refrigeration temperature.

## MATERIAL AND METHODS

Pulp was prepared by different processing methods to prepare diversified products.

**Procurement of jamun fruits:** Jamun fruits were procured from the local market of the Bangalore, which have good quality; fruits of

uniform size and shape, free from bruises, injuries and damages were selected. Harvested fruits were procured for the experiment.

### Preparation for the development of products

Ingredients for the development of the products were procured, namely sugar and citric acid. Jamun fruits were thoroughly washed in clean water. Pulp was separated from seeds by squeezing the fruits manually, which is made into paste. Ingredients were weighed with the help of an electronic balance.

### Extraction of pulp

The squeezed pulp was filtered by squeezing through muslin cloth to get juice; the extracted juice was used for preparation of squash and nectar. Squeezed pulp was made into paste and used for the preparation of jam.

### Processing of fruit pulp to different products

Development and standardization of jamun fruit based value added products were carried out with different percentage of jamun fruit pulp content. Fresh jamun fruits were used for preparation of squash, nectar and jam<sup>14</sup>.

**Table 1. Ingredients used for the preparation of jam (per 100g)**

Ingredients	Quantity
Jamun fruit pulp	45%
Sugar	55%
Citric acid	0.5-0.6%

([www.agritech.tnau.ac.in/postharvest/fpo\\_spec.html](http://www.agritech.tnau.ac.in/postharvest/fpo_spec.html)<sup>17</sup>.)

### Preparation of jam

#### Selection and preparation of fruit

Selected good quality ripe fruits Washed in cold water, peel the fruits and remove the stones and corers present. Cut the peeled fruit into small pieces with a stainless steel knife. Pulp was prepared.

#### Addition of sugar and acid

#### Cooking

Cook the mixture slowly with occasional stirring. The fruit pulp should be crushed with a laddle during cooking. Continue cooking till the temperature of the mass reaches 105.5°C. A small portion of jam is taken out during boiling in a spoon or wooden laddle and cooled slightly. It is then allowed to drop. If the product falls off in the form of a sheet (or) flakes instead of flowing in a continuous stream (or) syrup, it means that the end point has been reached and the product is ready.

Otherwise boiling is continued till the sheet test is positive.

#### Packaging

Fill the hot jam into clean dry sterilized jars. Allow the jam to cool and fix the sterilized lid to the jar. Stored in a cool place

#### Standardization of the jam

Standardization of the jam was carried out with the different percentage of pulp concentration. Four variations of jam were prepared.

- J1T1- 45 per cent of fruit pulp with 68.5° Brix.
- J2T2- 50 per cent of fruit pulp with 68.5° Brix.
- J3T3- 55 per cent of fruit pulp with 68.5° Brix.
- J4T4- 60 per cent of fruit pulp with 68.5° Brix.

**Table 2. Ingredients used for the preparation of squash (per 100ml)**

Ingredients	Quantity
Jamun fruit pulp	25ml
Sugar	150g
Citric acid	0.8g
Water	75ml
Sodium benzoate	0.3g

([www.agritech.tnau.ac.in/postharvest/fpo\\_spec.html](http://www.agritech.tnau.ac.in/postharvest/fpo_spec.html)<sup>17</sup>.)

### Preparation of squash

As per standard procedure squash was prepared, 25 per cent fruit juice or pulp and 40 to 50 per cent total soluble solids, commercially. It also contains about 1.0 per cent acid and 350 ppm sulphur dioxide or 600 ppm sodium benzoate. It is diluted before serving.

#### Steps in squash making

- Selection of fruit- select fresh, juicy, fully riped, firm wholesome and fresh fruits.
- To soften the fruits, keep them in fresh lukewarm water for ten minutes.
- Extract the juice and strain it.
- Calculate the amount of sugar, water and preservative to be added.
- Prepare the syrup with sugar, citric acid and water; cool it.

- Add strained fruit juice in the cooled prepared syrup.
- Add preservative dissolved in small quantity of cooled squash.
- Pour in dry sterilized bottle through a funnel leaving 2.5cm head space.
- Close the bottle and seal it.

#### Standardization of squash

Standardization of the squash was carried out with the different percentage of pulp concentration. Four variations of squash were prepared.

- S1T1- 25 per cent of fruit pulp with 40° Brix
- S2T2- 30 per cent of fruit pulp with 40° Brix
- S3T3- 35 per cent of fruit pulp with 40° Brix
- S4T4- 40 per cent of fruit pulp with 40° Brix

**Table 3. Ingredients used for the preparation of nectar (per 100ml)**

Ingredients	Quantity
Jamun fruit pulp	20%
Sugar	75%
Citric acid	0.3%
Water	330ml
Sodium benzoate	600ppm

([www.agritech.tnau.ac.in/postharvest/fpo\\_spech.tml](http://www.agritech.tnau.ac.in/postharvest/fpo_spech.tml)<sup>17</sup>.)

### Preparation of nectar

This type of fruit beverage contains at least 20 per cent fruit juice / pulp and 15 per cent total soluble solids and also about 0.3 per cent acid. It is not diluted before serving.

#### Steps in nectar making

- Select fully ripened fruits, wash and remove the peels and seeds.
- Cut into small pieces and mash properly.
- Pass through a strainer to get a smooth puree.
- Heat puree and boil for one to two minutes and then quickly cool down to room temperature by keeping the pan in water and stirring the puree quickly.
- Calculate the amount of sugar, water and preservative to be added.
- Prepare the syrup with sugar, citric acid and water; cool it.

- Mix all the ingredients properly. Heat the beverage for 3-5minutes and pour it to hot sterilized bottles and seal immediately. Pasteurize the bottles at 62.8° C for 30 minutes, cool immediately.

#### Standardization of nectar

Nectar was standardized with the different percentage of pulp concentration. Four variations of nectar were prepared.

- N1T1- 20 per cent of fruit pulp with 15° Brix
- N2T2- 25 per cent of fruit pulp with 15° Brix
- N3T3- 30 per cent of fruit pulp with 15° Brix
- N4T4- 35 per cent of fruit pulp with 15° Brix

#### Estimation of Vitamin C<sup>15</sup>

Ascorbic acid was determined colorimetrically. Ascorbic acid was first

dehydrogenated by bromination. The dehydroascorbic acid was then reacted with 2, 4 dinitrophenyl hydrazine to form osazone and dissolved in sulphuric acid to give an orange-red colour solution which was measured at 540nm.

#### Estimation of total phenolic content

Polyphenols were determined colorimetrically. Phenols reacted with phosphomolybdic acid in Folin- Ciocalteu reagent in alkaline medium

Calculations:

$$\text{Total O.D/100g} = \frac{\text{O.D X Volume made up X 100}}{\text{Weight of the sample}}$$

$$\text{Total anthocyanin (mg/100g)} = \frac{\text{Total O.D/ 100g}}{98.2}$$

#### Estimation of antioxidant activity

The antioxidant activity was expressed in terms of ascorbic acid equivalents; so ascorbic acid is taken as standard. Various concentrations of ascorbic acid were prepared and added to DPPH solution. The decrease in O.D is plotted against concentration of ascorbic acid. The concentration of sample was calculated using the standard curve.

#### Statistical analysis<sup>16</sup>

Factorial Complete Randomized Design (FCRD) analysis variance was applied and the data obtained for shelf life study was subjected to determine the level significance. One way analysis of variance was applied for sensory scores.

## RESULTS

#### Processing of fruit pulp to different products from jamun fruits

Jamun fruits are rich sources of phytochemicals and traditionally different types of fermented and non-fermented products were developed and consumed during off-seasons. The juice colour of jamun is more thermostable in comparison to other coloured fruit juices. So, there is immense potential to develop varied processed product of jamun fruits. Products like jam, squash and nectar

and produced blue coloured complex (molybdenum blue).

#### Estimation of Anthocyanins

Sample of 10 g was blended with 10ml of Ethanolic HCl, transferred to a 100ml volumetric flask and made upto volume. Stored overnight in refrigerator at 4°C. Filter through Whatman No. 1 filter paper and record O.D of the filtrate at 535nm.

were processed and standardized using standard procedure. After processing into different products alters the content of vitamin C, anthocyanins, total phenolic content and antioxidant activity during storage period both in refrigeration and also in room temperature.

#### Changes in Vitamin C during storage on products

Vitamin C of jam, squash and nectar during storage period of 90 days are presented in Table 4 (fig 1). As the storage period increases loss of vitamin C was observed at room temperature than the refrigerated temperature. Retention of vitamin C was found in refrigerated temperature; higher values of vitamin C was found in nectar (9.86-9.8mg/100g) followed by jam (7.7-7.3mg/100g) and squash (3.65-3.59mg/100g).

#### Changes in anthocyanins during storage on products

Anthocyanins of jam, squash and nectar during storage period of 90 days were presented in Table 5(fig 2). As the storage period increases significant loss of anthocyanins were observed at both the room and refrigerated temperature. Better retention of anthocyanin content was observed in jam (132-128 mg/100g) followed by squash (128-120 mg/100g) and nectar (97-85 mg/100g), at refrigeration temperature. At

room temperature significant loss was observed in jam (132-78mg/100g) squash (128-79mg/100g) and nectar (97-64mg/100g).

#### **Changes in total phenolic content during storage on products**

Total phenolic content of jam, squash and nectar during storage period of 90 days are presented in Table 6(fig 3). As the storage period increases significant loss of total phenolic content were observed at both the temperature. Compare to refrigerated temperature, at room temperature significant loss of total phenolic content of jam (218-185mg/100g GAE), squash (300-280mg/100g GAE) and nectar (260-238 mg/100g GAE) were noticed. Squash had higher values for total phenolic content (300-289mg/100g GAE) followed by nectar (260-252mg/100g GAE) and jam (211mg/100g GAE) at refrigeration temperature.

#### **Changes in antioxidant activity during storage on products**

Antioxidant activity of jam, squash and nectar during storage period of 90 days were presented in Table 7(fig 4). As the storage period increases the decline in the antioxidant activity was observed at both room and refrigerated temperature. At the end of 90<sup>th</sup> day of the storage period higher antioxidant activity was observed in squash (78mg/100g vitamin C equivalents) followed by nectar (69mg/100g vitamin C equivalents) and jam (62mg/100g vitamin C equivalents) at refrigerated temperature. Significant decline in antioxidant activity of jam (73-23mg/100g of vitamin C equivalents), squash (82-28mg/100g of vitamin C equivalents) and nectar (85-32mg/100g of vitamin C equivalents) were observed at room temperature during the storage period of 90 days.

## **DISCUSSION**

### **Development of the products from jamun fruits**

The products namely jam, squash and nectar was prepared and standardized using standard procedure<sup>17,14</sup>.

### **Change in Vitamin C during storage on products**

The vitamin C of the jam, squash and nectar are predicted in Table 14. As the storage period increased the loss of vitamin C content was found to be significant irrespective of temperature and irrespective of jamun fruit products. During the storage period of 90 days loss of vitamin C content was found to be significant between the room and refrigerated temperature. Degradation of vitamin C content was found to be significant between the products. As the storage days increases the vitamin C decreases at both the temperature. The rate of degradation of vitamin C depends on both the temperature; significant reduction in the ascorbic acid content was observed in all the three products namely jam, squash and nectar. The reduction of ascorbic acid content in refrigerated temperature was less than that of room temperature. This could be due to the storage at refrigerated temperature reduces the rate of oxidation of ascorbic acid to dehydroascorbic acid. Citric acid has been reported to have protective effect on L-ascorbic acid and tends to slow its oxidation due to its metal sequestering properties. Being a photosensitive vitamin, vitamin C degradation during the storage period of 90 days might be accelerated by the presence of light at room temperature. Similar findings are in line with the Jawaheer<sup>18</sup> *et al.*

### **Change in total anthocyanins during storage on products.**

The anthocyanins of the jam, squash and nectar are depicted in Table 18. At room temperature reduction of anthocyanin content was high compare to refrigeration temperature for all the products. The loss of anthocyanin content was high due to degradation. These results are in agreement with Gimenez<sup>19</sup> *et al.*. Degradation of anthocyanins in all the products might be due to the oxidation. Ascorbic acid radicals can be regenerated to ascorbic acid by oxidising one molecule of anthocyanin into its well established radical form; so degradation of anthocyanins occur<sup>20</sup>. As the storage period increased the loss of anthocyanins was found to be significant between the room and refrigeration temperature. During the storage period of 90

days the loss of anthocyanins was found to be significant between the products of jamun fruits. As the storage period increased the loss of anthocyanin content was found to be significant irrespective of temperature and irrespective of jamun fruit products.

#### Change in total phenolic content during storage on products.

The Total phenolic content of the jam, squash and nectar are depicted in Table 19. Squash had higher values for total phenolic content at room and refrigerated temperature followed by nectar and jam, during the storage period of 90 days. A significant loss was noticed at room temperature than the refrigerated temperature for all the products. The decreasing trend of total phenols in jamun squash during storage period at room temperature might be due to the sensitivity of the phenolic components to oxidation at storage conditions<sup>21,22</sup>.

As the storage period increased loss of total phenolic content between the jamun fruit products were found to be significant. During the storage period of 90 days loss of total phenolic content of jam, squash and nectar

were found to be significant between the room and refrigeration temperature. As the storage period increased loss of total phenolic content was found to be significant irrespective of temperature and irrespective of jamun fruit products.

#### Change in antioxidant activity during storage on products.

The antioxidant activity of the jam, squash and nectar are described in Table 20. As the storage period increased loss of antioxidant activity was found to be significant irrespective of temperature and irrespective of jamun fruit products. During the storage period of 90 days loss of antioxidant activity of jam, squash and nectar were found to be significant between the room and refrigerated temperature. As the storage period increased loss of antioxidant activity was found to be significant between the jamun fruit products. The results revealed that nectar had higher antioxidant activity followed by squash and jam. These results are in agreement with the Muhammad and Sheikh<sup>23</sup>.

**Table 4. Changes in Vitamin C (mg/100g) during storage on products**

Storage Period (No of days)	Room temperature			Mean Values of Different jamun products	Refrigerated temperature			Mean Values of Different jamun products	Products			Combined Mean values for storage period
	Jam	Squash	Nectar		Jam	Squash	Nectar		Jam	Squash	Nectar	
Initial	7.7	3.65	9.86	7.07	7.7	3.65	9.86	7.07	7.7	3.65	9.86	7.07
30	6.5	2.81	8.83	6.38	7.6	3.64	9.84	7.02	7.55	3.22	9.33	6.70
60	5.9	2.7	7.19	5.26	7.5	3.6	9.81	6.97	6.7	3.15	8.5	6.11
90	5.1	2.6	6.69	4.79	7.3	3.59	9.8	6.8	6.2	3.09	8.2	5.84
Mean of individual products	6.3	2.94	8.14	5.875	7.525	3.62	9.827	6.965	7.037	3.277	8.972	6.43

Comparison of mean values	A	B	C	AB	BC	AC	ABC
SEm±	0.163	0.200	0.231	0.283	0.401	0.327	0.567
CD @ 5 %	0.467	0.572	0.661	0.810	1.145	0.935	1.620

Note: A-Temperature (A1, A2), B-Products (B1, B2, B3), C-Storage period (C1, C2, C3, C4)

**Table 5: Changes in Anthocyanin content (mg/100g) during storage on products**

Storage Period (No. of days)	Room temperature			Mean Values of Different jamun products	Refrigerated temperature			Mean Values of Different jamun products	Products			Combined Mean values for storage period
	Jam	Squash	Nectar		Jam	Squash	Nectar		Jam	Squash	Nectar	
Initial	132	128	97	119	132	128	97	119	132	128	97	119
30	125	112	88	108.33	131	125	93	116.33	128	118.5	90.5	112.33
60	94	98	77	89.66	130	123	89	114	112	110.5	83	101.83
90	78	79	64	73.66	128	120	85	111	103	99.5	74.5	92.33
Mean of individual products	107.25	104.25	81.5	97.66	130.25	124	91	115.08	118.75	114.12	86.25	106.37

Comparison of mean values	A	B	C	AB	BC	AC	ABC
SEm±	0.170	0.208	0.240	0.294	0.417	0.340	0.589
CD @ 5 %	0.486	0.595	0.687	0.842	1.191	0.972	1.684

Note: A-Temperature (A1, A2), B-Products (B1, B2, B3), C-Storage period (C1, C2, C3, C4)

**Table 6: Changes in Total phenolic content (mg/100g GAE) during storage on products**

Storage Period (No of days)	Room temperature			Mean Values of Different jamun products	Refrigerated temperature			Mean Values of Different jamun products	Products			Combined Mean values for storage period
	Jam	Squash	Nectar		Jam	Squash	Nectar		Jam	Squash	Nectar	
Initial	218	300	260	259.3	218	300	260	259.3	218	300	260	259.33
30	207	292	251	254	215	298	259	257.3	211	295	255	250
60	192	289	245	249	213	295	257	255	202.5	292	251	242
90	185	280	238	243	211	289	252	250.6	198	285	245	234.33
Mean of individual products	200.5	290.25	248.5	251.3	214.3	295.5	257	255.55	207.4	293	252.75	246.41

Comparison of mean values	A	B	C	AB	BC	AC	ABC
SEM±	0.169	0.207	0.239	0.293	0.415	0.339	0.587
CD @ 5 %	0.484	0.593	0.685	0.839	1.185	0.969	1.678

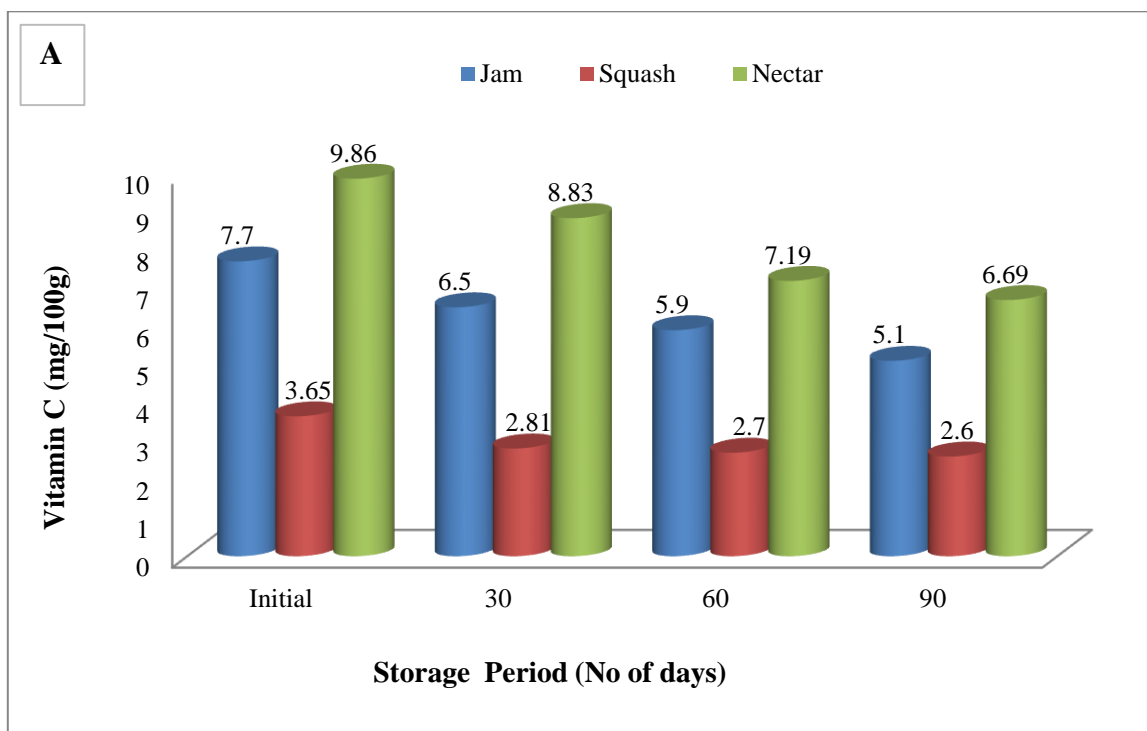
Note: A-Temperature (A1, A2), B-Products (B1, B2, B3), C-Storage period (C1, C2, C3, C4)

**Table 7: Changes in Antioxidant activity (mg/100g vitamin C equivalents) during storage on products**

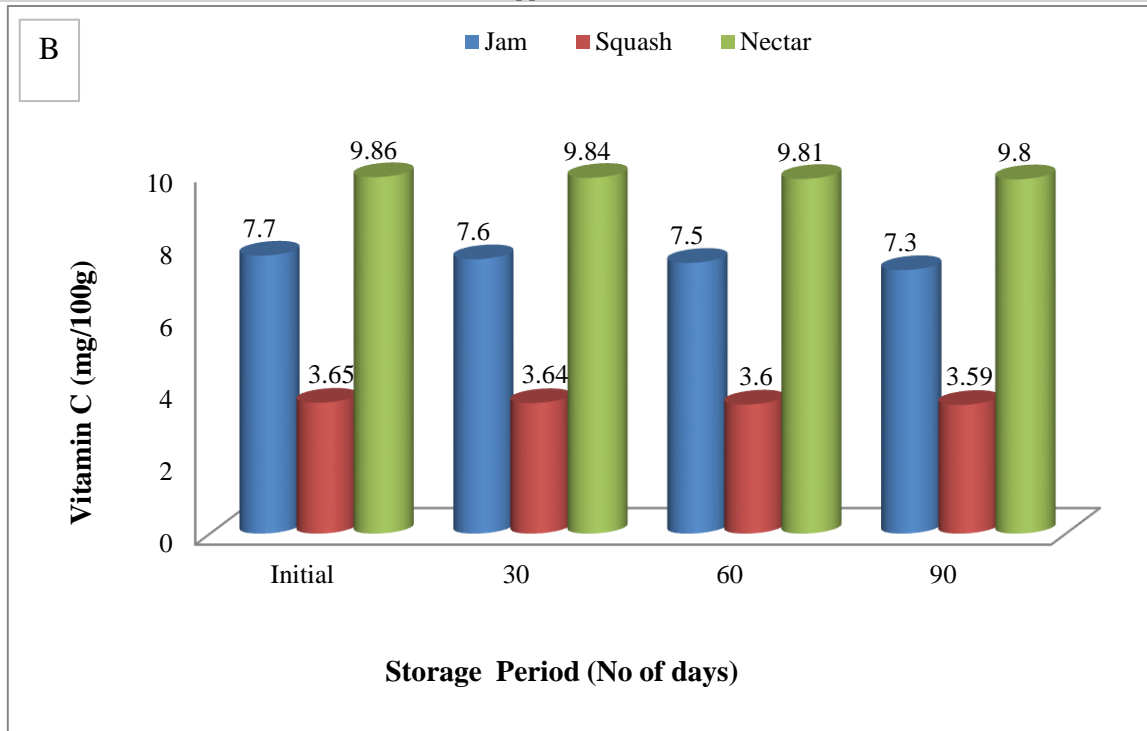
Storage Period (No of days)	Room temperature			Mean Values of Different jamun products	Refrigerated temperature			Mean Values of Different jamun products	Products			Combined Mean values for storage period
	Jam	Squash	Nectar		Jam	Squash	Nectar		Jam	Squash	Nectar	
Initial	73	82	85	80	73	82	85	80	73	82	85	80
30	65	58	67	63.33	70	80	83	77.66	67.5	69	75	70.5
60	48	47	59	51.33	68	79	70	72.33	58	63	64.5	61.83
90	23	28	32	27.66	62	78	69	69.66	42.5	53	50.5	48.66
Mean of individual products	53.5	53.75	60.75	55.58	68.25	79.75	76.75	60.25	60.5	66.75	68.75	77.42

Comparison of mean values	A	B	C	AB	BC	AC	ABC
SEM±	0.158	0.194	0.224	0.274	0.388	0.317	0.549
CD @ 5 %	0.453	0.554	0.640	0.784	1.109	0.906	1.569

Note: A-Temperature (A1, A2), B-Products (B1, B2, B3), C-Storage period (C1, C2, C3, C4)



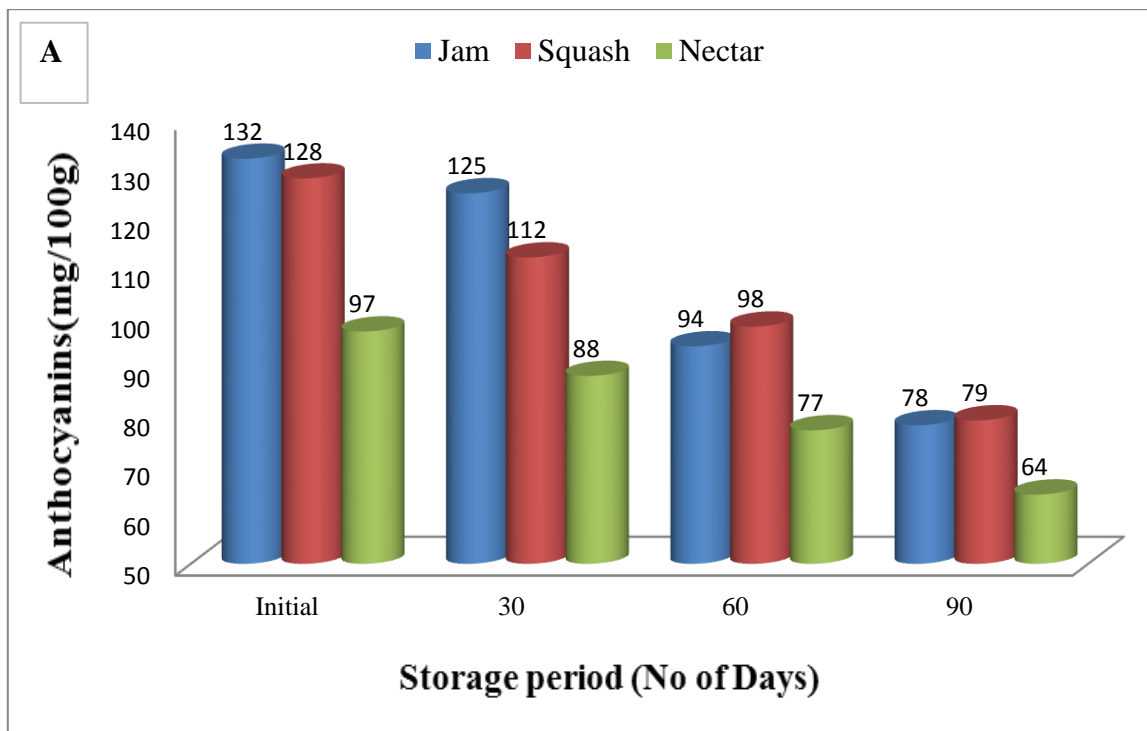


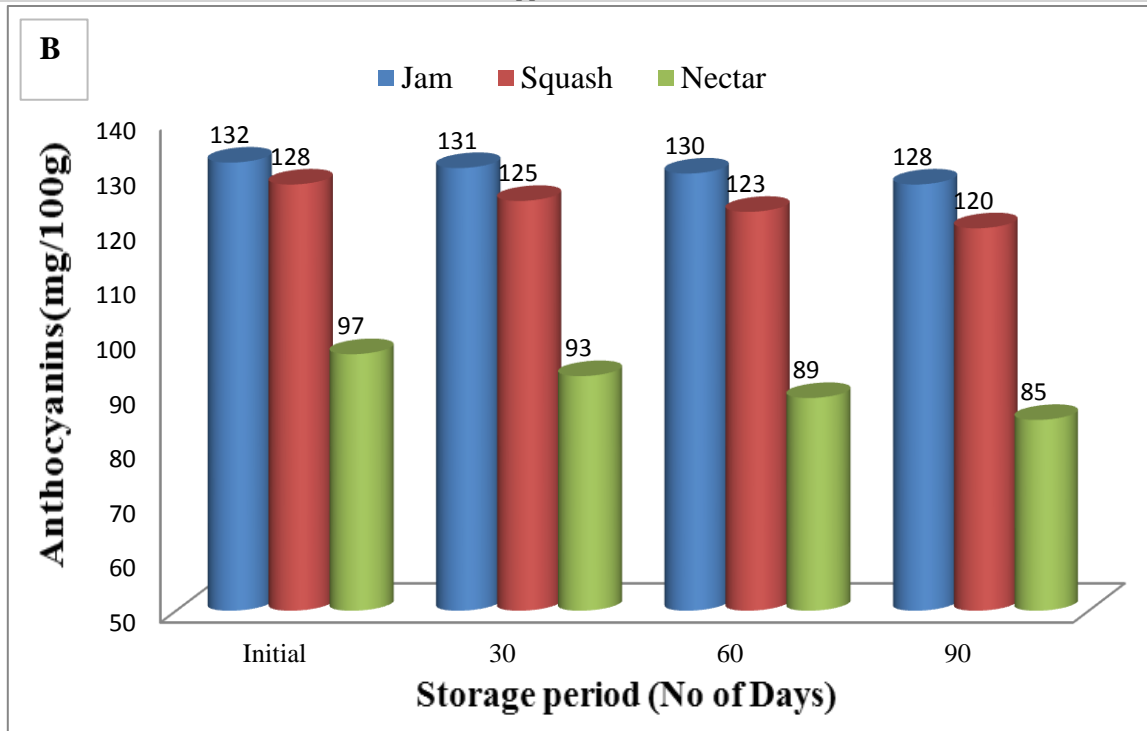


Note: A- Room temperature

B- Refrigerated temperature

Fig 1: Change in Vitamin C during storage on products

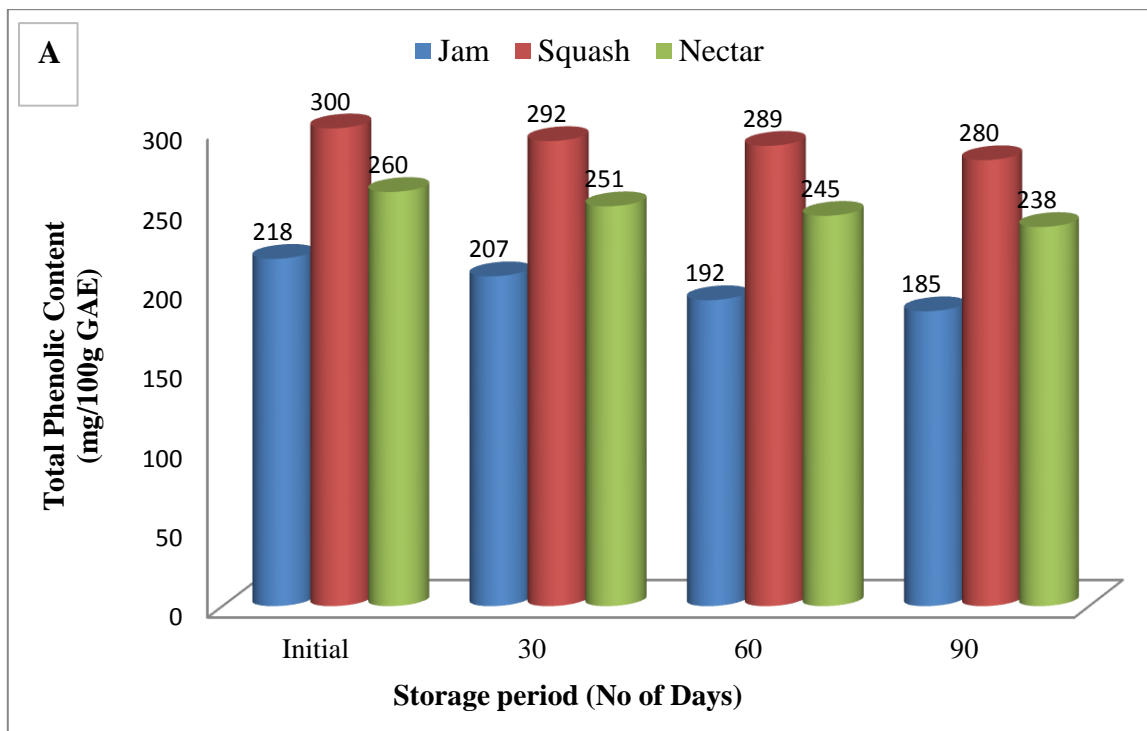


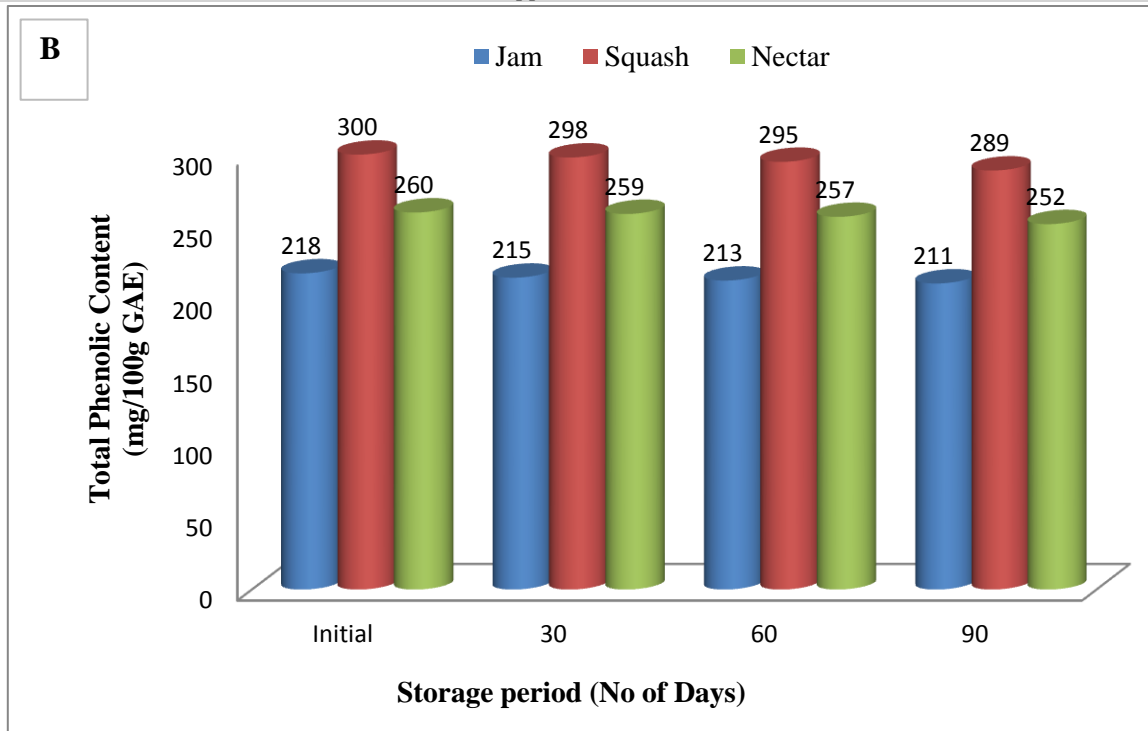


Note: A- Room temperature

B- Refrigerated temperature

Fig 2: Change in anthocyanins during storage on products

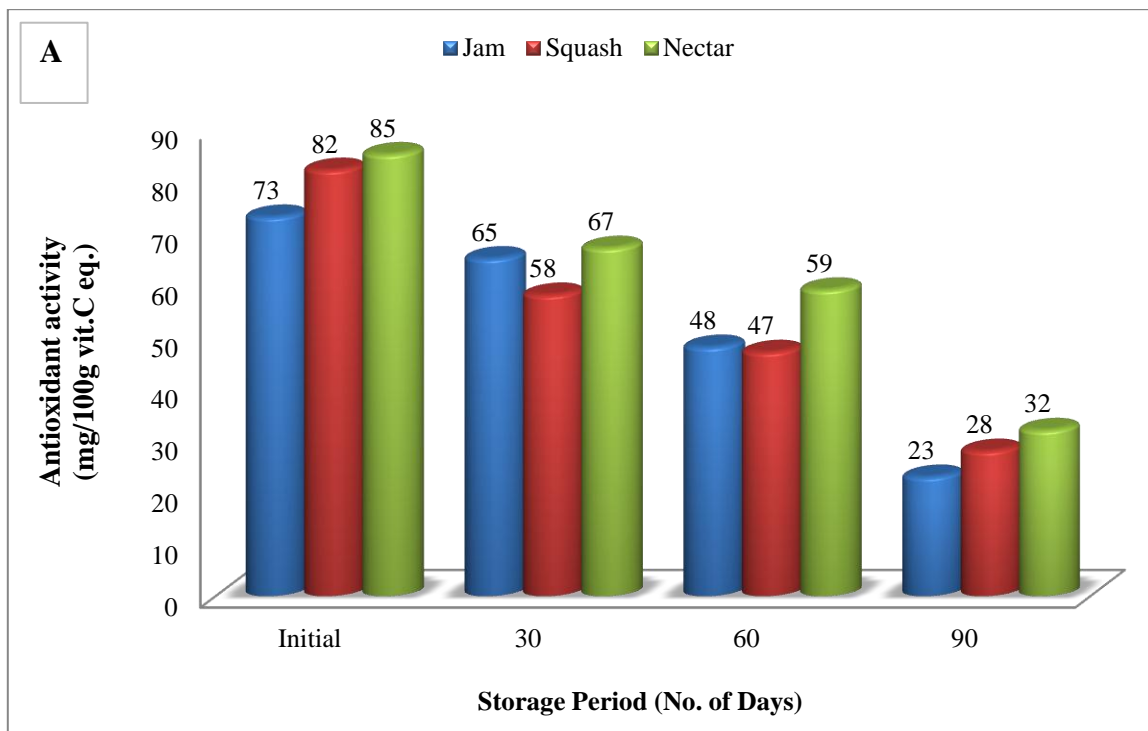


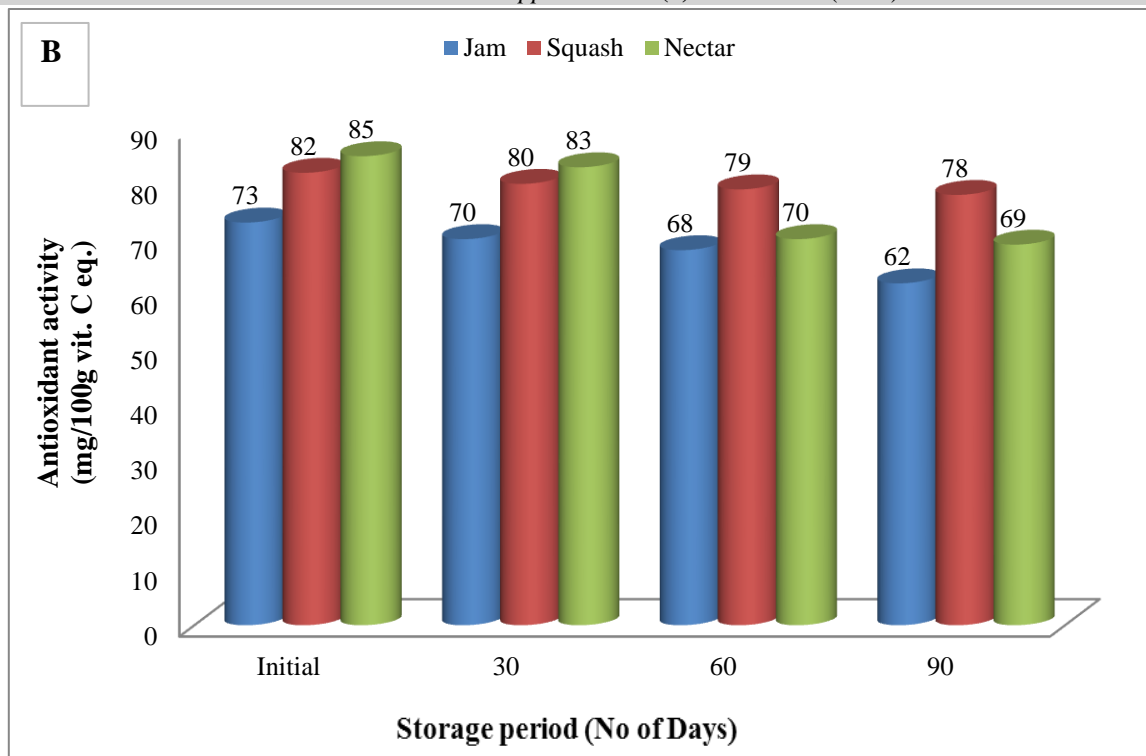


Note: A- Room temperature

B- Refrigerated temperature

Fig 3: Change in total phenolic content during storage on products





Note: A- Room temperature

B- Refrigerated temperature

Fig 4: Change in antioxidant activity during storage on products

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