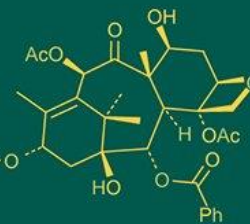
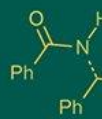
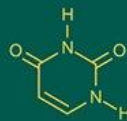
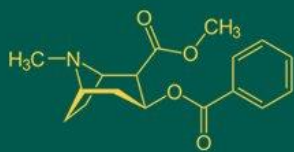


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Studies on physio chemical properties and storage life of mulberry - aloe-vera blended squash

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Abstract

The present study was undertaken to investigate the physio-chemical properties and storage stability of mulberry-*aloe vera* blended squash. Fully ripe mulberry fruits and fresh *aloe vera* leaves were processed into juices, which were blended in six proportions (90:10, 80:20, 70:30, 60:40, 50:50, and 40:60) following a factorial completely randomized design (CRD) with three replications. The formulations were standardized at 35% juice content, 45°Brix TSS, spice extract, and 600 ppm sodium benzoate. The squash samples were stored at room temperature, and observations were recorded at 0, 30, 60, and 90 days of storage. Results revealed that mulberry juice contained significantly higher levels of TSS (15.98°B), ascorbic acid (19.43 mg/100 g), reducing sugars (8.54%), total sugars (11.17%), total phenolics (185.35 mg GAE/100 g), flavonoids (118.45 mg QE/100 g), antioxidant activity (98.93%), and anthocyanins (104.90 mg/100 g), compared to aloe vera juice, which exhibited lower nutritional values. Increasing the proportion of aloe vera in the blends led to a reduction in TSS, sugars, ascorbic acid, phenolics, flavonoids, antioxidant activity, and anthocyanins. Storage studies indicated a gradual decline in ascorbic acid, phenolics, flavonoids, antioxidants, and anthocyanins, while reducing and total sugars increased, and titratable acidity decreased significantly over 90 days. Among the treatments, T₁ (90% mulberry: 10% aloe vera) consistently retained maximum nutritional quality and sensory acceptability throughout storage, while aloe vera-rich blends exhibited lower stability. The study concludes that mulberry-*aloe vera* squash, particularly mulberry-dominant blends, can serve as a novel functional beverage with good nutritional retention, storage stability, and consumer appeal, offering scope for commercialization and value addition in the fruit processing sector.

Keywords: Mulberry, *aloe vera*, blended squash, physico-chemical properties, storage stability, functional beverage, antioxidant activity, sensory quality

Introduction

Food processing has historically played a vital role in enhancing preservation, minimizing post-harvest losses, and ensuring food security by making nutritious and safe food available to consumers (Floros *et al.*, 2010; Weaver *et al.*, 2014). Despite India's significant fruit production, less than 2% is processed, compared to over 60% in developed nations, which highlights the need for value-added products (Thallapally, 2011).

Mulberry (*Morus* spp.) is a nutrient-rich fruit valued for its high anthocyanin, phenolic, and flavonoid content, contributing to strong antioxidant properties. It can be processed into various products such as jams, juices, wines, and food colorants (Özgen *et al.*, 2009; Jan *et al.*, 2021). Nutritionally, mulberries are rich in ascorbic acid, minerals, proteins, and bioactive compounds like quercetin and rutin, making them a promising functional food (Zou *et al.*, 2014; Stintzing *et al.*, 2002).

Aloe vera (*Aloe barbadensis*) is another important crop with a long history of medicinal and nutritional use. Its gel contains bioactive compounds such as polysaccharides, flavonoids, vitamins, and phenolics, which provide antioxidant, antimicrobial, anti-inflammatory, and immunomodulatory benefits (Rodriguez *et al.*, 2010; Usman & Alam, 2024). Due to these properties, aloe vera has wide applications in food, cosmetic, and pharmaceutical industries (Boghani *et al.*, 2012) [7].

Blending fruit juices is an effective strategy to enhance flavor, nutritional value, and consumer acceptability. Combining mulberry with aloe vera may yield a novel functional beverage with improved health-promoting attributes (Bhardwaj & Mukherjee, 2011; De

Carvalho *et al.*, 2007). However, little research has been conducted on the preparation and storage stability of such blended squashes. In this context, the present study was undertaken with the objectives:

To standardize the recipe for mulberry-aloe vera blended squash.

To conduct sensory evaluation of the product.

To assess its physicochemical properties, storage stability, and microbial quality.

Materials and Methods

The current research titled "Preparation of Mulberry - Aloe-vera Blended Squash" was carried out in the Post graduate laboratory, department of horticulture, VNMKV Parbhani during the 2024-25 period. The fully matured and healthy mulberry specimens were obtained from the Sericulture Research Unit at VNMKV, Parbhani, Maharashtra. The selected mulberry fruits were fully ripe, exhibiting a color range from purple to black. The unripe, distorted, diseased, damaged, and off-type specimens were discarded. The matured, healthy aloe vera leaves were procured from Department of Horticulture, VNMKV, Parbhani. Different spices like cardamom, black pepper, cumin, common salt, ginger and sugar were acquired from local market of Parbhani.

Extraction of mulberry juice

The ripe mulberry fruits, devoid of dirt, dust, and other foreign materials, were subjected to washing with tap water in the laboratory. Subsequently, juice extraction was performed manually, utilizing 1 kg of fruits, which were pressed to obtain the juice. The extracted juice was stored in PET (polyethylene terephthalate) jars and maintained at a temperature of -18 °C in deep freezers.

Processing of Aloe vera juice

The aloe vera leaves underwent a washing process under running water, followed by trimming at the tip, lower portion, and sides to eliminate thorns. The leaves were then set aside for a duration of 2-3 hours to facilitate the removal of toxic materials. The leaves were subsequently severed using a stainless-steel knife, followed by the extraction of the pulp. The pulp was subsequently processed in the mixer and filtered. The extracted juice was stored in PET (polyethylene terephthalate) jars at a temperature of -18°C in deep freezers.

Preparation of mulberry aloe vera blended squash

The mulberry and aloe vera juices were combined in various ratios according to the treatment specifications. The blended juices were subsequently combined with the sugar syrup, along with the spice extract and ginger juice. The spice extract was prepared by boiling a ground mixture of various quantities of spices, specifically cumin (2.5 g), cardamom (1 g), black pepper (2.5 g), and common salt (5 g), in 200 ml of water. Subsequently, the mixture was strained and combined with 20 ml of ginger juice following the established procedure (Thakur *et al.*, 2016). Sodium benzoate was subsequently added as a preservative at a concentration of 600 ppm for each treatment. The appetizer was contained in sterilized glass bottles.

Experimental Design

The study on *mulberry-aloe vera blended squash* was conducted using a Factorial Completely Randomized Design (CRD) with six treatments and three replications. The experiment was designed to assess the effect of different blending ratios of mulberry and aloe vera juices on the physicochemical, microbial, and sensory quality of squash during storage. The squash samples were stored in sterilized glass bottles at room temperature, and observations were recorded at 0, 30, 60, and 90 days of storage.

Table 1: Treatment details

Sr. No.	Mulberry juice (%)	Aloe vera juice (%)
T ₁	90	0
T ₂	80	10
T ₃	70	20
T ₄	60	30
T ₅	50	40
T ₆	40	50

Parameters to be taken

In the present study, the physicochemical and sensory parameters of mulberry-aloe vera blended squash were observed. Chemical attributes included total soluble solids (TSS) measured using a hand refractometer (Ranganna, 2014), pH and titratable acidity determined following AOAC (2012) methods, and ascorbic acid content estimated by the 2,6-dichlorophenol indophenol titration method (AOAC, 2012). Reducing and total sugars were quantified using Fehling's solution titration (AOAC, 2012). The physico-chemical quality was further assessed through total phenolic content by the Folin-Ciocalteu method (Ahmed & Abozed, 2015), total flavonoids using aluminium chloride colorimetric assay (Marinova *et al.*, 2005), antioxidant activity by the DPPH radical scavenging method (Luo *et al.*, 2009), and anthocyanin content following Ranganna (2014). Sensory attributes such as colour, flavour, taste, and overall acceptability were evaluated by a semi-trained panel using a 9-point hedonic scale (Amerine *et al.*, 1965). The experimental data were analyzed statistically using factorial completely randomized design (CRD) as suggested by Gomez and Gomez (1984).

Results and Discussion

Physico-chemical characteristics of mulberry and aloe vera

The physico-chemical properties of mulberry and aloe vera juices, as presented in Table 2, revealed significant variations between the two. Mulberry juice recorded a TSS of 15.98 °B, with corresponding values of pH (3.61), titratable acidity (0.65%), ascorbic acid (19.43 mg/100 g), reducing sugars (8.54%), total sugars (11.17%), and high levels of bioactive compounds including total phenolics (185.35 mg GAE/100 g), flavonoids (118.45 mg QE/100 g), antioxidant activity (98.93%), and anthocyanins (104.90 mg/100 g). In contrast, aloe vera juice showed a much lower TSS (2.08 °B), with pH of 4.74, titratable acidity of 0.12%, ascorbic acid content of 2.07 mg/100 g, reducing sugars of 0.32%, and total sugars of 0.68%. The bioactive profile of aloe vera was also comparatively lower, with total phenolics (12.21 mg GAE/100 g), flavonoids (9.79 mg QE/100 g), and antioxidant activity (86.79%).

Table 2: Physio chemical charactersitics of mulberry and aloe vera

Constituents	Mulberry	Aloe vera
TSS (°Brix)	15.98	2.08
pH	3.61	4.74
Titrateable acidity (%)	0.65	0.12
Ascorbic acid (%)	19.43	2.07
Reducing sugar (%)	8.54	0.32
Total sugars (%)	11.17	0.68
Total phenolic content (mg GAE/100g)	185.35	12.21
Total flavonoid content (mgQE/100g)	118.45	9.79
Antioxidant activity (%)	98.93	86.79
Anthocyanin (mg/100g)	104.90	ND

Effect of blending and storage on chemical attributes of squash

The TSS of mulberry-aloe vera blended squash was standardized to 45°B at the beginning of storage across all treatments. A slight reduction in TSS was observed with increasing proportions of aloe vera juice, although the treatment effect remained statistically non-significant during storage as presented in Table 3, with mean values ranging between 45.75% and 46.15%. In contrast, the storage period significantly influenced TSS, which recorded the lowest mean (45.73%) at 30 days and the highest (46.80%) at 90 days, statistically comparable with 60 days (46.27%). The treatment × storage interaction was non-significant. Similar findings regarding TSS stability during storage of fruit-based beverages have been reported in blended squash and

nectar products (Ranganna, 2014; Bhardwaj & Mukherjee, 2011; Jan *et al.*, 2021). The ascorbic acid content of mulberry-aloe vera blended squash decreased significantly with higher proportions of aloe vera in the formulation, with maximum values in T₁ (90:0) (12.54 mg/100 g) and minimum in T₆ (40:50) (7.58 mg/100 g). Storage also caused a steady decline, from 11.29 mg/100 g at the initial stage to 8.40 mg/100 g after 90 days. The reduction is attributed to oxidative degradation and conversion to dehydro-ascorbic acid or furfural during storage. Similar declining trends in vitamin C have been reported in blended beverages like aonla-aloe vera juice (Sharma *et al.*, 2023), sea buckthorn squash (Selvamuthukumaran & Khanum, 2013), and jackfruit-aloe vera RTS (Hossain *et al.*, 2017).

Table 3: Effect of blending and storage period on TSS and ascorbic acid of Mulberry-Aloe vera blended squash

Treatment	TSS (%)				Mean	Ascorbic acid(mg/100g)				Mean
	Storage period (Days)					Storage period(Days)				
	0	30	60	90		0	30	60	90	
T ₁ (90:0:: Mulberry: aloe vera)	45.00	46.01	46.56	47.05	46.15	14.12	13.14	11.84	11.05	12.54
T ₂ (80:10:: Mulberry: aloe vera)	45.00	45.91	46.42	46.95	46.07	13.02	12.72	10.07	9.75	11.39
T ₃ (70:20:: Mulberry: aloe vera)	45.00	45.74	46.33	46.85	45.98	11.59	11.38	9.73	8.97	10.42
T ₄ (60:30:: Mulberry: aloe vera)	45.00	45.66	46.25	46.77	45.92	10.72	11.16	8.80	7.70	9.60
T ₅ (50:40:: Mulberry: aloe vera)	45.00	45.57	46.07	46.67	45.83	9.87	9.72	7.83	6.93	8.59
T ₆ (40:50:: Mulberry: aloe vera)	45.00	45.49	45.97	46.53	45.75	8.44	9.01	6.92	5.97	7.58
Mean	45.00	45.73	46.27	46.80		11.29	11.19	9.20	8.40	

Reducing sugars were significantly influenced by treatments and storage. Squash with higher mulberry content (T₁: 90:0) showed the highest mean reducing sugars (11.20%), while T₆ (40:50) recorded the lowest (6.66%) as presented in Table 4. Across storage, values increased from 8.18% initially to 10.29% at 90 days, likely due to hydrolysis of non-reducing sugars into reducing sugars. These findings are consistent with studies on guava-aloe vera nectar (Singh *et al.*, 2019), bael-aloe vera RTS (Tiwari & Deen, 2015), and mulberry appetizers (Hamid & Thakur, 2017) [20-21].

Total sugar content also declined with greater aloe vera incorporation, ranging from 15.57% in T₁ (90:0) to 10.53% in T₆ (40:50). However, storage showed a significant upward trend, with values increasing from 12.38% at day 0 to 14.28% at 90 days. The increase may be due to inversion of non-reducing sugars into reducing sugars during storage. Similar increases in total sugars during storage have been documented in mulberry-aloe vera nectar (Rahman, 2021), bael-aloe vera RTS (Tiwari & Deen, 2015), and rhododendron squash (Thakur *et al.*, 2020).

Table 4: Effect of blending and storage period on reducing sugars and total sugars of Mulberry -Aloe vera blended squash

Treatment	Reducing sugars (%)				Mean	Total sugars (%)				Mean
	Storage period (Days)					Storage period (days)				
	0	30	60	90		0	30	60	90	
T ₁ (90:0:: Mulberry: aloe vera)	10.05	10.09	11.97	12.69	11.20	14.97	14.83	15.96	16.53	15.57
T ₂ (80:10:: Mulberry: aloe vera)	9.48	10.01	11.89	12.24	10.90	14.59	14.70	16.02	16.37	15.42
T ₃ (70:20:: Mulberry: aloe vera)	8.77	8.89	10.79	10.35	9.70	12.70	12.76	14.00	14.54	13.50
T ₄ (60:30:: Mulberry: aloe vera)	7.94	8.32	9.84	9.82	8.98	11.52	11.79	13.01	14.42	12.69
T ₅ (50:40:: Mulberry: aloe vera)	7.09	7.21	7.94	8.66	7.73	10.71	10.83	11.83	12.30	11.42
T ₆ (40:50:: Mulberry: aloe vera)	5.72	6.10	6.88	7.94	6.66	9.81	9.82	10.95	11.54	10.53
Mean	8.18	8.44	9.88	10.29		12.38	12.46	13.63	14.28	

Titrateable acidity was significantly affected by blending and storage. The highest mean acidity was found in T₁ (1.38%) and the lowest in T₆ (1.21%), showing a decline with increasing aloe vera content. Storage also reduced acidity, from 1.35% at the initial stage to 1.24% at 90 days, likely due to interaction of organic acids with sugars and amino acids. These results align with earlier findings in sea buckthorn squash (Selvamuthukumar & Khanum, 2013), guava-aloe vera nectar (Rani & Babu, 2015), and wild prickly pear beverages (Chauhan *et al.*, 2019) [9].

Effect of blending and storage on physico-chemical attributes of squash

The total phenolic content of mulberry-aloe vera blended squash declined significantly with increasing aloe vera proportion, ranging from 171.62 mg GAE/100 g in T₁ (90:0) to 86.81 mg GAE/100 g in T₆ (40:50) which is presented in Table 5. Storage also caused a gradual reduction, with mean

values decreasing from 129.69 mg GAE/100 g at the initial stage to 124.73 mg GAE/100 g at 90 days. The decline is attributed to the oxidation of phenolic compounds, polymerization, and interactions with proteins during storage. Comparable reductions in phenolic compounds have been reported in box myrtle squash (Thakur *et al.*, 2016), wild aonla appetizer (Thakur *et al.*, 2018), and orange-aloe vera RTS (Kausar *et al.*, 2019). Flavonoid content was significantly higher in mulberry-rich formulations, with T₁ (90:0) recording 109.32 mg QE/100 g compared to 55.97 mg QE/100 g in T₆ (40:50). A steady decline was observed over the storage period, from 87.35 mg QE/100 g initially to 81.65 mg QE/100 g at 90 days, likely due to oxidative and enzymatic degradation of flavonoids. Similar decreases in flavonoid content during storage were observed by Neelam & Deen (2024) in fruit-aloe vera blends and by Thakur *et al.* (2020) in rhododendron squash.

Table 5: Effect of blending and storage period on total phenolic content and flavonoid of Mulberry -Aloevera blended squash

Treatment	Total phenolic content (mgGAE/100g)				Mean	Total Flavonoid content (mgQE/100g)				Mean
	Storage period (Days)					Storage period (days)				
	0	30	60	90		0	30	60	90	
T ₁ (90:0:: Mulberry: aloe vera)	174.21	172.11	170.21	169.96	171.62	112.02	110.12	108.32	106.82	109.32
T ₂ (80:10:: Mulberry: aloe vera)	159.71	157.41	155.23	154.96	156.83	104.02	101.72	99.82	98.62	101.04
T ₃ (70:20:: Mulberry: aloe vera)	136.91	134.17	133.61	131.64	134.08	95.02	92.52	90.72	89.22	91.87
T ₄ (60:30:: Mulberry: aloe vera)	117.69	115.01	113.17	111.07	114.24	83.02	80.42	78.52	77.12	79.77
T ₅ (50:40:: Mulberry: aloe vera)	100.41	98.10	96.73	95.89	97.78	71.02	68.32	66.72	65.02	67.77
T ₆ (40:50:: Mulberry: aloe vera)	89.21	87.71	85.47	84.83	86.81	59.02	56.92	54.82	53.12	55.97
Mean	129.69	127.42	125.74	124.73		87.35	85.00	83.15	81.65	

Antioxidant activity decreased significantly with higher aloe vera incorporation, ranging from 40.74% in T₁ (90:0) to 37.29% in T₆ (40:50). Storage also showed a declining trend, with values dropping from 40.52% at day 0 to 36.86% at 90 days as presented in Table 6. The reduction is attributed to the degradation of anthocyanins and ascorbic acid during storage. Comparable findings were reported in guava-aloe vera nectar (Singh *et al.*, 2019), wild prickly pear beverage (Chauhan *et al.*, 2019) [9], and jamun appetizer (Bhatt, 2019) [6]. Anthocyanin levels were significantly

higher in mulberry-dominant blends as presented in Table 6, with T₁ (90:0) showing 91.43 mg/100 g compared to 50.53 mg/100 g in T₆ (40:50). Storage led to a progressive decline, from 74.14 mg/100 g initially to 66.18 mg/100 g at 90 days, mainly due to the high susceptibility of anthocyanins to oxidative degradation. These findings are in line with previous studies on mulberry appetizer (Hamid & Thakur, 2017) [20-21], wild pomegranate appetizer (Thakur *et al.*, 2017), and mulberry-aloe vera nectar (Rahman, 2021).

Table 6: Effect of blending and storage period on antioxidant and anthocyanin activity of Mulberry -Aloevera blended squash

Treatment	Antioxidant activity (%)				Mean	Anthocyanin				Mean
	Storage period (Days)					Storage period (days)				
	0	30	60	90		0	30	60	90	
T ₁ (90:0:: Mulberry: aloe vera)	43.00	42.16	39.51	38.30	40.74	94.43	91.52	90.69	89.07	91.43
T ₂ (80:10:: Mulberry: aloe vera)	42.44	41.95	39.49	37.99	40.47	94.41	92.70	91.60	80.95	89.92
T ₃ (70:20:: Mulberry: aloe vera)	40.36	40.65	38.19	36.70	38.98	78.70	76.88	74.23	67.27	74.27
T ₄ (60:30:: Mulberry: aloe vera)	39.25	40.24	37.84	36.69	38.50	64.17	64.43	62.82	57.69	62.28
T ₅ (50:40:: Mulberry: aloe vera)	39.13	39.08	37.63	36.36	38.05	61.08	59.87	53.98	53.93	57.21
T ₆ (40:50:: Mulberry: aloe vera)	38.95	38.38	36.69	35.13	37.29	52.08	51.87	50.03	48.14	50.53
Mean	40.52	40.41	38.23	36.86		74.14	72.88	70.56	66.18	

Conclusion

The present investigation demonstrated that blending mulberry and aloe vera juices significantly influenced the physicochemical and nutritional attributes of squash during storage. Mulberry juice, being rich in sugars, phenolics, flavonoids, anthocyanins, and ascorbic acid, contributed to higher antioxidant potential, while aloe vera imparted functional properties and balanced acidity. Increasing aloe vera proportion resulted in lower TSS, sugars, ascorbic acid, phenolics, flavonoids, and anthocyanins, whereas mulberry-

dominant blends maintained superior nutritional and sensory quality. Storage studies revealed a gradual decline in ascorbic acid, phenolic compounds, flavonoids, antioxidant activity, and anthocyanins, while reducing and total sugars increased due to inversion of non-reducing sugars. Titrateable acidity also declined with storage, likely due to utilization of organic acids in biochemical reactions. Among treatments, T₁ (90% mulberry: 10% aloe vera) consistently exhibited the best retention of quality attributes and overall acceptability

up to 90 days of storage, suggesting that mulberry-rich formulations are more stable and consumer-preferred.

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Conflict of Interest

The authors declare no conflicts of interest. They bear sole responsibility for the content and composition of the paper.

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