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Development and quality evaluation of bottle gourd, *Lagenaria siceraria* (Mol.) Standl. based blend juice

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Bottle gourd [*Lagenaria siceraria* (Mol.) Standl.] based blend juice was formulated using Aonla (*Emblica officinalis* Gaertn.), Lemon [*Citrus limon* (L.) Burm.f.] and Ginger (*Zingiber officinale* Rosc.) juices. Response surface methodology and central composite design with 30 experimental combinations was used for optimization of the blend based on numerical technique. Optimized blend juice was hot filled at 85 ^{o}C in glass bottles, crown corked and were thermally processed in hot water at 85 ^{o}C for 5 min to meet required food safety and standards regulations, 2011. The sensory, physicochemical and microbiological quality of thermally processed blend juice was evaluated during storage for 180 days under ambient conditions (30±2 °C). No significant difference (P>0.05) was found in pH, total soluble solids, total acidity and most sensory attributes of the product during initial 30 days of storage. However, significant difference (P<0.05) was observed in ascorbic acid content, sugars and microbiological counts during further storage. The total loss of ascorbic acid was about 47.68 % and product was microbiologically safe and acceptable during 180 days storage.

Keywords: Blanching, Blend juice, Bottle gourd, Formulation, Storage quality, Thermal processing.

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Introduction

Among various vegetables grown worldwide, bottle gourd [*Langenaria siceraria* (Mol.) Standl.] has a high place in diet. It belongs to *Cucurbitaceae* family and known as Calabash, Doodhi, Lauki, White flowered gourd, Trumpet gourd, Calebassier, Courage bouteille (French), Cojombro, Guiro amargo (Spanish), Upo, Talayag, Gucuzzi, Zucca melon (Philippines), Mokwa and Oo Lo Kwa (China)[1]. Bottle gourd fruit is a good source of vitamins, minerals, cucurbitacins, polyphenols, campesterol and sitosterol along with 1.6 % choline on dry weight basis[2-4]. A 100 g edible portion of raw bottle gourd contains 95.54 g water, 3.39 g carbohydrates, 0.62 g protein, 0.02 g fats, 0.5 g total dietary fiber, 150.0 mg potassium, 26.0 mg calcium, 13.0 mg phosphorus, 10.1 mg ascorbic acid, 2.0 mg sodium and 0.320 mg niacin[5].

Bottle gourd fruit has higher edible index and lower waste index, which proves its importance for processing. Bottle gourd is underutilized fruit in spite of being one of the cheapest source of nutrients and potential source of natural antioxidants. The fruit is also well known for its antihyperlipidemic, anti-stress, adaptogenic, aphrodisiac, analgesic, anti-inflammatory, antidote to certain poisons, alternative purgative, cardio protective, cardio tonic, cooling, diuretic, hepatoprotective and immunomodulatory properties[6,7]. The bottle gourd juice has been used to treat acidity, indigestion, ulcers, nervous diseases and can serve as an effective thirst quencher. A glass of bottle gourd juice taken daily is also considered to prevent premature greying of hair[8].

In bottle gourds, the enzymes responsible for browning are peroxidase (POD) and polyphenol oxidase (PPO) and hence the juice in its pure form is brown in colour, off-flavour and has unpleasant taste. POD, if inactivated during processing, all other enzyme systems are usually inactivated[9-11]. To improve the taste, aroma, palatability, storability and nutritive value of bottle gourd juice, it would be convenient to blend it with other highly nutritive fruit juices namely aonla (*Emblica officinalis* Gaertn.), lemon [*Citrus limon* (L.) Burm.f.] and spice extracts like ginger (*Zingiber officinale* Rosc.). This will serve economic alternative for utilization of this underutilized fruit, particularly for development of nutritive juices.

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Bottle gourd juice in its pure form is very difficult to preserve because of lower acidity (0.17 %). Preservation of low acid juice for long storage at ambient condition, a high thermal processing temperature ≥121 °C is required[12]. Acidification may convert the low acid juice of bottle gourd to an acidic juice by blending with aonla and lemon fruits and allow the use of milder thermal process conditions. Both fruit juices have higher acidity and are rich in vitamins C and minerals. In fact aonla juice has astringent taste and both the anola and the lemon juice turn bitter immediately after extraction due to conversion of a chemical compound and hence processing of fruit juices in its pure form is limited[13,14]. Sensory quality and consumer acceptability of the bottle gourd based blend juice could be further increased by the addition of ginger juice.

Extraction of bottle gourd juice is mostly done manually in the household or at cottage level, which is less hygienic with high chances of degradation within few hours. Current research work on bottle gourd juice in terms of quality production is very limited. Hence, there is an urgent need to develop processing technology and its appropriate blends with other fruit juices for preparing natural health drink with adequate shelf -life. During the last few years the demand for vegetable blended juice has increased due to change in dietary habits, taste preferences and the way of life of present-day consumers. Bottle gourd blended juices have higher nutritional values compared to single fruit juices or synthetic beverages and are not commercially available in the market but have the potential to develop into a 'natural health drink'. Therefore, the objective of this study was to develop a unique processing technology by standardizing and optimizing various parameters at every stage for production of stable, acceptable and quality blend juice based on bottle gourd.

Materials and Methods

Raw materials

Tender bottle gourd (cv. 'ABG-1'), fresh aonla (cv. 'Anand-2') and matured lemon [*Citrus limon* (L.) Burm. f.] fruits were procured from the Horticultural Farm, Anand Agricultural University. Ginger rhizomes were procured from the local vegetable market, Anand (India). The fruits and ginger rhizomes were sorted cleaned and washed using running tap water.

Blanching and juicing

To inactivate POD enzyme in the bottle gourd, fruits were sliced to thickness of 5 mm using a stainless steel slicer machine (Sumeet) and blanched at 100 °C for 3.67 min in a water bath (Electro equipment, New Delhi, India). POD inactivation test was carried out as described by Ranganna[15] with 3 replications and 69.40 % juice yield was obtained from blanched slices using a centrifugal juicer (Rama udyog, Jaipur, India). Juice from anola was also extracted after blanching of whole anola fruits. Fruits to water ratio was maintained to 1:7 w/v during blanching process. Lemon was cut into two halves and juice was extracted by squeezing. Ginger rhizomes were cut and juice was extracted using domestic mixer-cum-grinder. All the juice samples were strained using muslin cloth.

Blending

Response surface methodology (RSM) and central composite design (CCD) was used to formulate 30 experimental combinations for juice blending. The quadratic models were generated for the variables vis-a-vis responses and samples were statistically analyzed. To obtain a desired quality blend juice, multiple responses Design-Expert software, based on numerical technique was used (Design expert® 8.0.7.1, Statease Inc, Minneapolis, USA).

Processing and storage

RSM optimized blend juice was hot filled at 85 °C in the pre-sterilized 200 mL glass bottles, followed by crown corking and was thermally processed (Nova Instruments Pvt Ltd, Ahmedabad) at 85 °C for 5 min at atmospheric pressure to meet required food safety and standards regulations (FSSRs), 2011[Ref. 16]. Thermally processed cooled bottles were stored under ambient conditions (30±2 °C) for 180 days in laboratory incubator (Khera Instruments Pvt Ltd, New Delhi) for quality evaluations.

Chemical analysis

pH and total soluble solids (TSS) (°Brix) values were measured using digital pH meter (Systronics India Limited, India) and refractometer-PAL-1(ERMA, Japan) having 0-53 °Brix range, respectively. Total acidity (%) and ascorbic acid content (mg/100 mL) were determined as described by Ranganna[15]. Sugars (%) were determined using Lane and Eynon method[15]. Total solids (%) were determined according to method described in AOAC[17].

Microbiological analysis

Microbiological quality of juice samples in term of Total Plate Count (TPC), Yeasts and Molds Count

and Coli form Counts were determined using standard procedures as described by Ranganna[15] in sterile environment.

Sensory analysis

Organoleptic quality was evaluated using a 9 point hedonic rating test as suggested by Ranganna[15], where 9 is 'like extremely' and 1 is 'dislike extremely.' A panel of 11 semi trained testers among Faculty of the Institute had carried out the acceptance tests at an ambient condition (30±2 °C), evaluating appearance & colour, flavour, taste, consistency and overall acceptability (OAA) of the blend juice.

Chemicals

Hydrogen peroxide (30 %), sodium bicarbonate (99.5 %), phenolphthalein (1 %), Fehling's solutions (A+B), methylene blue indicator, neutral lead acetate (45 %), potassium oxalate (22 %), HCl and NaOH were procured from Merck, Mumbai, India; Guaiacol (99 %) and ascorbic acid standard (99 %) from SD Fine Chemicals Mumbai, India; metaphosphoric acid (60 %) and sodium salt (98 %) from Loba Chemie, Mumbai, India and plate count agar, PDA and VRBA were procured from Hi-Media Laboratories Pvt Ltd, Mumbai. De-ionized distilled water was used for all assays.

Statistical analysis

The data (mean±standard deviation) obtained were analyzed at 95 % confidence level using the statistical software SAS version 9.3 (SAS Institute Inc, Statistical Analysis System, Cary, NC).

Results and Discussion

Physico-chemical characteristics

Effect on pH and acidity

Mean values showed a gradual increase in acidity (0.84 to 0.94 %) and low reduction in pH (3.42 from initial 3.52) of blend juice during the storage (Table 1). This change might be due to conversion of sugars to acid inherently present in blend juice. Significant difference (P<0.05) in pH and acidity was found during 180 days of storage although it did not affect much the quality of blend juice as reflected in the sensory evaluation. The finding is in agreement with the results reported by Majumdar *et al*[18]. No significant difference was observed at 95 % confidence level during initial 30 days of storage, which showed a good stability of blend juice (Table 2).

Effect on TSS and total solids

The initial mean value of TSS of the optimized blend juice was 5.17 °Brix and was increased to 5.27 °Brix at the end of 180 days of storage (Table 1). Total solids were significantly increased during storage in the blend juice and significant increase during advancement of storage might be due to decrease in pH of the blend juice. No significant difference (P>0.05) was observed in TSS during initial 120 days of storage (Table 1 and 2). However, gradual increase in TSS during storage was noticed, which may be due to hydrolysis of the complex carbohydrates present in the blend juice.

Table 1 — Physico-chemical characteristics of blend juice during ambient storage

Parameters	Storage period (days)						
	0	30	60	90	120	150	180
pH	3.52	3.51	3.49	3.47	3.45	3.44	3.42
TSS (°Brix)	5.17	5.20	5.23	5.23	5.23	5.27	5.27
Total acidity (%)	0.84	0.85	0.87	0.89	0.91	0.92	0.94
Total solids (%)	5.40	5.43	5.53	5.63	5.63	5.70	5.77
Total sugars (%)	3.18	3.17	3.16	3.15	3.15	3.13	3.12
Reducing sugars (%)	2.59	2.62	2.65	2.68	2.73	2.78	2.86
Non- reducing sugars (%)	0.60	0.55	0.51	0.47	0.42	0.35	0.26
Ascorbic acid (mg/100 mL)	38.51	34.45	31.12	28.38	25.33	22.54	20.15

Table 2 — Physico-chemical characteristics ANOVA for blend juice during ambient storage

Source	DF	Type III SS	Mean Square	LSD	F Value	Pr > F
pH	6	0.02229524	0.00371587	0.0108	97.54	<.0001
TSS (°Brix)	6	0.02285714	0.00380952	0.0936	1.33	0.3063
Total acidity (%)	6	0.02729524	0.00454921	0.0127	86.85	<.0001
Total solids (%)	6	0.33238095	0.05539683	0.1430	8.31	0.0006
Total sugars (%)	6	0.00779048	0.00129841	0.0121	27.27	<.0001
Reducing sugars (%)	6	0.16822857	0.02803810	0.0138	452.92	<.0001
Non-reducing sugars (%)	6	0.24599048	0.04099841	0.0206	296.89	<.0001
Ascorbic acid (mg/100 mL)	6	772.8661238	128.8110206	1.2152	267.51	<.0001

The finding is in agreement with the result reported by Li J *et al*[19] and Majumdar *et al*[20].

Effect on sugars

The non-reducing and total sugars decreased during the storage while the reducing sugars increased (Table 1). The total sugars values showed significant difference (P<0.05) because increase in reducing sugars was immediately followed by decrease in non-reducing sugars (Table 2). This change might be due to inversion of non-reducing sugars into reducing sugars in the presence of acidic environment of the blend juice.

Effect on ascorbic acid

The ascorbic acid content of optimized blend juice was 38.51 mg/100 mL and significantly (P<0.05) decreased up to 20.15 mg/100 mL during storage (Table 1 and 2). Increase in ascorbic acid losses was 47.68 % at the end of storage. Result revealed that prolonging storage time at 30 °C temperature increased the ascorbic acid losses more. The ascorbic acid was more sensitive at higher temperature and might have been easily oxidized by both enzymatic and non-enzymatic reactions.

Microbiological characteristics

Effect on TPC

Mean values of TPC of the optimized blend juice was 33 cfu/mL and decreased to 3 cfu/mL from initial followed by increase in yeast and mould counts at the end of 180 days of storage (Table 3). The t test (LSD) analysis revealed that significant difference (P<0.05) was found for the TPC during storage (Table 4). However, no significant difference was found at 95 % confidence level for the total plate counts during 120 to 180 days storage. Decrease in TPC along with advancement of storage period might be due to the inhibitory effect of acidic blend juice towards the total plate counts. **S**imilar results were obtained by Derossi *et al*[21]. TPC was not found more than 50 cfu/mL in blend juice as per Food Safety and Standards Regulations[16], hence the blend juice was microbiologically safe during 180 days storage.

Effect on yeasts & moulds and coli form count

Mean value of yeasts and moulds count of the optimized blend juice was 2 cfu/mL and increased to 6 cfu/mL during storage (Table 3). Significant difference (P<0.05) found for the yeasts and moulds count during 180 days storage (Table 4) was due to the favorable conditions of acidic environment of the blend juice toward the yeasts and moulds. The findings are in agreement with the result reported by William[22]. The coli form count (cfu/mL) in the blend juice was nil initially and was found nil during 180 days storage.

Sensory characteristics

Highest sensory score was 6.43 for appearance and colour while least score was 6.32 for consistency of the blend juice at the end of 180 days of storage (Table 5). Sensory scores of flavour and taste was good during the storage. The blend juice was found to be superior in terms of OAA having score of 6.95 initially, which decreased only by 7.48 % at the end of 180 days of storage. The t tests did not showed significant difference (P>0.05) for the appearance and color, taste, consistency and OAA of the blend juice during initial 30 days of storage (Table 6). However, significant difference (P<0.05) was found for all attributes of sensory scores during 180 days of storage. The OAA gradually decreased along with advancement of storage due to gradual change in physic-chemical characteristics of blend juice. The finding is in conformity with the result reported by Majumdar *et al*[18].

Table 3 — Microbiological characteristics of blend juice during ambient storage

	Storage period (days)						
Parameters	0	30	60	90	120	150	180
Total plate counts (cfu/mL)	33	20	11	7	5	4	3
Yeasts & moulds count (cfu/mL)	2	3	4	5	6	6	6
Coli form counts (cfu/mL)	0	0	0	0	0	0	0

Table 5 — Sensory characteristics of blend juice during ambient storage

	Storage period (days)						
Parameters	0	30	60	90	120	150	180
Appearance and colour	6.94	6.86	6.80	6.69	6.62	6.52	6.43
Flavour	6.96	6.78	6.62	6.52	6.46	6.42	6.35
Taste	6.89	6.85	6.81	6.73	6.63	6.50	6.38
Consistency	6.85	6.83	6.74	6.60	6.53	6.43	6.32
Overall acceptability	6.95	6.91	6.78	6.63	6.51	6.50	6.43

Table 4 — Microbiological characteristics ANOVA for blend juice during ambient storage

Source	DF	Type III SS	Mean Square	LSD	F Value	Pr > F
Total plate counts (cfu/mL)	6	2195.2381	365.87302	1.8721	320.14	<.0001
Yeasts & moulds count (cfu/mL)	6	52.666667	8.7777778	1.1464	20.48	<.0001

Table 6 — Sensory characteristics ANOVA for blend juice during ambient storage

Source	DF	Type III SS	Mean Square	LSD	F Value	Pr > F
Appearance and colour	6	0.67216190	0.11202698	0.1354	18.73	<.0001
Flavour	6	0.82742857	0.13790476	0.0731	79.13	<.0001
Taste	6	0.63706667	0.10617778	0.0564	102.28	<.0001
Consistency	6	0.73432381	0.12238730	0.0554	122.39	<.0001
Overall acceptability	6	0.78416190	0.13069365	0.0693	83.42	<.0001

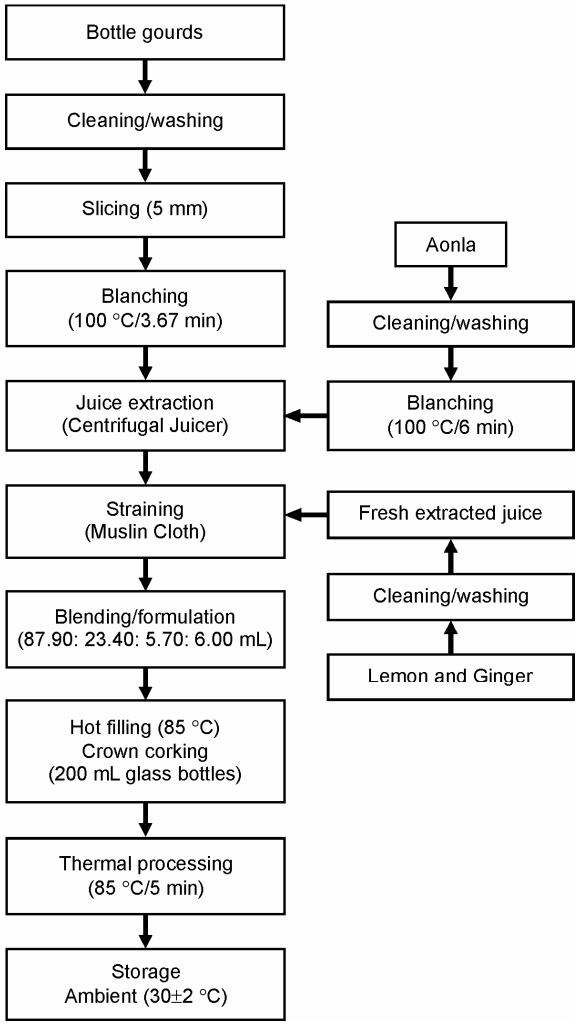

Fig. 1 — Suggested process flow chart for the production of bottle gourd based blend juice

Process flow chart

As a result of various experiments conducted for development of quality blend juice; it was found that an acceptable quality blend juice based on bottle gourd could be prepared using the suggested process as depicted in Fig. 1. Proportion of 87.90 mL blanched bottle gourd juice, 23.40 mL blanched aonla juice, 5.70 mL fresh lemon juice and 6.00 mL fresh ginger juice should be blended. Blended juice should be hot filled at 85 °C in 200 mL glass bottles followed by crown corking. Filled and sealed bottles should be thermally processed immerging in hot water at 85 °C for 5 min followed by cooling at room temperature. Store the processed bottles at an ambient condition (30±2 °C) for the safe consumption up to 180 days.

Conclusion

A quality bottle gourd based blend juice could be prepared through blending aonla, lemon and ginger juices with an application of minimal thermal processes and without adding any chemicals and preservatives in it. The blend juice so prepared was stable and microbiologically safe in line with FSSRs, 2011 up to 180 days of storage at an ambient condition (30±2 °C). The process will enable large scale commercial production of quality and stable blend juice based on bottle gourd as a natural health drink as it satisfies the sensory, physico-chemical and required microbiological criteria.

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