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PHYSICOCHEMICAL, MICROBIOLOGICAL AND ORGANOLEPTIC ASSESSMENT OF NECTARS PREPARED FROM GUAVA AND BUTTERNUT WINTER SQUASH BLENDS

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ABSTRACT: *Five blends of different ratios from guava and butternut winter squash were prepared to produce nectars. Prepared samples named guava, guava-butternut winter squash (9:1 w/w), guava-butternut winter squash (8:2 w/w), and guava-butternut (6:4 w/w). Physicochemical constituents, microbial counts and sensory characteristics were determined for prepared nectars during storage for 6 months. The acidity of nectar samples increased gradually during storage periods. Meanwhile, the loss of ascorbic acid contents increased with increasing storage time. The decrease of total sugars and total carotenoids of nectar samples were slightly decreased. Microbial analysis of prepared nectars revealed that, bacterial counts, yeast and molds counts were gradually decreased with increasing storage period. On the other hand, sensory attributes after preparation of nectar samples and after 6 months of storage indicated that, nectar prepared from guava- butternut winter squash (8:2 w/w) had the highest organoleptic scores followed by guava- butternut winter squash (9:1 w/w), guava-butternut winter squash (7:3 w/w) and guava-butternut winter squash (6:4 w/w) respectively.*

Key words: *Guava, Butternut winter squash, Nectars, Microbial count, Sensory evaluation.*

INTRODUCTION

Juice blending is a new concept in the manufacture of fruit juices. It initiates a new aroma which could be favorable to the interesting consumer. Beside, other nutritional and economical trends could be attained. Butternut winter squash newly introduced in Egypt which arrive in the growing season and has a long shelf life, its flavor is sweet, moist and pleasantly nutty. Winter squash is also a source of potassium, niacin, iron, calcium, phosphorus and beta carotene (Attia and Rizk 2009) as a general rule, the deeper the orange color, the higher the beta carotene content. Beta-carotene is converted to vitamin A in the body. Vitamin A and beta carotene can protect the cell membranes and others cellular structure from the damage caused by oxygen free radicals. The precursor of vitamin A, beta carotene; is an antioxidant like selenium, which can inhibit excess of oxidation of fats (lipid peroxidation) in the cell. (Morton, 1990).

The metabolic implications of carotenoids in the human diet have become an important issue in today's nutritional interest. In addition to their role as provitamin A, carotenoids may play an important role in body tissues as free radical quenchers and antioxidants (Bendich and Olsen 1992). Beside, its high content of dietary fiber plays an important role in reducing the incidence of colon cancer (Aguero *et al.*, 2008). Stated that members of cucurbitaceae family make significant contribution to our intake of vitamins and minerals. There has been interest in carotenoids for possible anticarcinogenic activity (Zhang *et al.*, 1992).

Accordingly butternut winter squash (*Cucurbita moschata Duch*) consumption is recommended. Moreover, they are easy to cultivate and are available all year round. Beside they are cheap and of better taste. As such the nutritional deficiency in vitamin A and C, which causes many diseases, may be reduced (Gonzalez *et al.*, 2001).

The objective of this research are carried out to produce nectars from different blends of guava and butternut winter squash which were higher in nutrients such as vitamins and minerals. Therefore, studying the physicochemical and microbiological aspect as well as sensory evaluation of different blends immediately after the production and during storage for 6 months was carried out.

MATERIALS AND METHODS

Materials

Guava fruits (*Psidun guava, L.)* were brought from private farm at Helwan. Fruits were picked at the ripe stage in September 2008.

Butternut winter squash (*Cucurbita moschata*) were brought from Kaha Agriculture Research Station Horticulture Research Institute in March 2008. Fig (1).

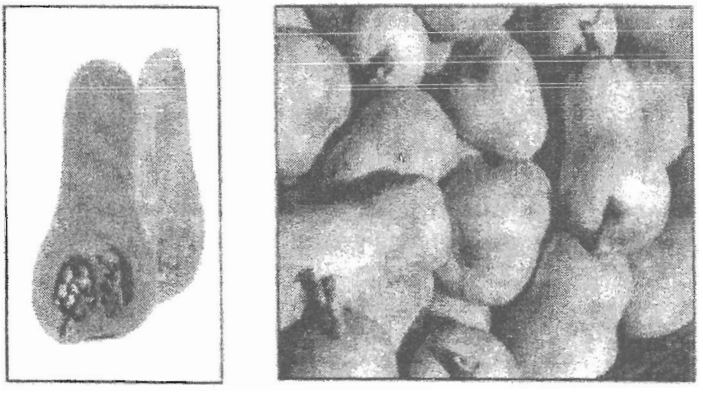

Fig. (1): Butternut winter squash

Manufacture of nectar samples

Preparation of juice

Guava fruits were washed, cut into quarters and pulped with the warring blender and screened. Butternut winter squash was washed, cut into froths and peeled. The center were scooped out with a spoon to remove seeds and fiber. The squash cut into one cubes which were blanched at 90°C for 4 min. and cooled at 20°C after that they were pulped with warring blender and screened.

Homogenized pulp of guava and squash were used to prepare different blends as follows in Table (1)

Table (1). The blends of guava and butternut winter squash extracted juices.

Materials	Blend 1	Blend 2	Blend 3	Blend 4	Blend 5
Guava	100 %	90.0 %	80.0 %	70.0 %	60.0 %
Butternut winter squash	0.0 %	10.0 %	20.0 %	30.0 %	40.0 %

Preparation of nectar

Each 100 ml of prepared nectar contain 0.2% citric acid, 0.20% carboxymethylcellulose, 0.10% sodium sorbate and adjusted of total soluble solids to 18% by using the sucrose. Nectar were filled in white glass bottles, closed tightly, pasteurized at 85°C for 20 min. then cooled an stored at room temperature(25 ± 2°C) for 6 months.

Methods

Moisture content, total soluble solids, total acidity as citric acid, reducing sugar, total sugars, ash, ascorbic acid, protein, fat, crude fibers and minerals (Ca, Iron, and P) were determined according to methods described in (A.O.A.C 1995).

Determination of total carotenoids

Total carotenoids were determined by the method of (Ready and Sistrunk 1980) summarized as follows: five gram of tested sample was extracted with 40 ml hexane, 60 ml ethanol. After blending, the hexane layer as separated by adding 2% cold NaCl solution. Sample was diluted (1: 10) and the O.D was measured at 440 nm and compared to β-carotene standard curve.

Microbiological assay

Fifty grams samples were placed into sterilized flasks and 450 ml of sterile phosphate buffer solution was added. The mixture was mechanically shaken for 30 min. at steady speed. Serial dilutions were prepared in sterile saline solution for the following tests.

Total viable count

Duplicate plates were inoculated with 1 ml for each dilution and thoroughly mixed with 10 to 15 portion of nutrient agar (Difco Manual 1985); the plates were incubated for 48 hrs at 37°C .

The plates of the suitable dilution were recorded after the incubation period. The counts/gm food materials were calculated.

Yeast and mold count

Duplicate plates were inoculated with 1 ml for each dilution and thoroughly mixed with 10 – 15 ml portion of potato dextrose agar pH 5-6. the solidified plates were incubated for 48 hrs at 30°C. The plate counts of the suitable dilution were recorded after the incubation period. The counts/gm food materials were calculated.

Sensory evaluation

Sensory evaluation was carried out by ten panelists of nectar samples at zero time of storage and after storage period using the method described by (Reitmeier and Nonnecke 1991).

Statistical methods

Data were statistically analyzed to facilitate comparing the least significant differences (LSD) between means of different values according to (Snedecor and Cochran (1973).

Results and Discussion

Chemical constituents of guava fruits and butternut winter squash

Chemical constituents of guava fruit and butternut winter squash are presented in Table (2). It could be noticed that, fresh guava contain 83.80% moisture content, 0.71% ash, 0.35% fat, 0.85% protein, 2.76% reducing sugar 8.56% total sugars and 3.12% crude fiber, while fresh butternut winter squash contained 87.52% moisture content, 0.60% ash, 0.18% fat, 0.90% protein, 7.35% reducing sugar, 9.50% total sugars and 0.90% crude fiber. On the other hand, total soluble solids and acidity were 9.40 and 0.54% in guava and 10.50 and 0.09% in butternut winter squash respectively. While, the ascorbic acid, total carotenoid, irons, phosphorus and calcium in guava were 208, 0.20, 0.70, 24 and 21 mg/100gm respectively. However, the corresponding values in butternut winter squash were 21.0, 3.40, 0.80, 30.0 and 44.0 mg/100gm. The butternut winter squash contained the highest content of carotenoid, calcium and phosphorus compared with guava fruits.

Table (2). Physicochemical properties of fresh guava fruits and butternut winter squash.

Constituent		Fresh cultivar	
		Guava	Butternut winter squash
Moisture	%	83.80	87.52
Total solids	%	16.20	12.48
Total soluble solids	%	9.40	10.50
Total acidity	%	0.54	0.09
Protein	%	0.85	0.90
Fat	%	0.35	0.18
Crude fiber	%	3.12	0.90
Ash	%	0.71	0.60
Reducing sugar	%	2.76	7.35
Non reducing sugar	%	5.80	2.15
Total sugar	%	8.56	9.50
Ascorbic acid	mg/100gm	208.0	21.0
Total carotenoid	mg/100gm	0.20	3.40
Iron	mg/100gm	0.70	0.80
Phosphorus	mg/100gm	24.0	30.0
Calcium	mg/100gm	21.0	44.0

Changes in chemical composition of prepared nectar samples during storage at room temperature

Changes in chemical constituent of nectar samples stored at (25°C±2°C) up to 6 months are presented in Table (3). Results show that, total soluble solids were 18% in all fresh nectar samples and slightly noticeable changes were observed during storage being 17.5after 6months of storage.

The acidity of nectar samples increased gradually during the storage period and the incremental changes of acidity were 15.63, 13.33, 16.00, 23.81% and 16.67% for guava, guava-butternut squash (9 : 1 w/w), guava-butternut squash (8 : 2 w/w), guava-butternut squash (7 : 3 w/w) and guava-butternut squash (6 : 4 w/w) respectively.

These results are in agreement with (Leonard and Rose, 1960) who found that, the slight increment in acidity may be due to trans formation of pectin into pectic acid during prolonged storage.

Table (3). Changes in chemical constituents of nectar samples during storage for 6 months at room temperature (25 ± 2°C)

Constituent	Storage period in months				Change %	Storage period in months				Change %
	0	2	4	6		0	2	4	6	
	Guava					Guava-butternut winter squash (9: 1 w/w)				
Total soluble solids%	18.0	18.0	18.0	17.5	- 2.78	18.0	18.0	17.82	17.75	- 1.39
Total acidity%	0.32	0.35	0.36	0.37	+ 15.63	0.30	0.30	0.32	0.34	+ 13.33
Total sugars%	17.82	17.46	17.29	17.00	- 4.60	17.72	17.53	17.21	16.93	- 4.46
Ascorbic acid mg/100gm	160.0	140.0	130.0	100.0	- 37.50	140.0	125.0	100.0	86.0	- 38.57
Total carotenoid mg/100gm	0.12	0.12	0.11	0.11	- 8.33	0.26	0.25	0.25	0.25	- 3.85
	Guava-butternut winter squash (8:2w/w)					Guava-butternut winter squash (7: 3 w/w)				
Total soluble solids%	18.0	17.90	17.85	17.80	- 1.11	18.0	18.0	17.75	17.50	- 2.78
Total acidity%	0.25	0.26	0.26	0.29	16.00	0.21	0.23	0.25	0.26	+ 23.81
Total sugars%	17.68	17.46	17.20	16.84	- 4.75	17.60	17.20	17.00	16.62	- 5.57
Ascorbic acid mg/100gm	115.0	40.0	72.0	50.0	- 56.52	98.0	72.0	55.0	39.0	- 60.20
Total carotenoid mg/100gm	0.42	0.41	0.41	0.40	- 4.76	0.63	0.62	0.62	0.61	- 3.17
	Guava-butternut winter squash (6: 4 w/w)									
Total soluble solids%	18.0	18.0	17.85	17.75	- 1.39					
Total acidity%	0.18	0.20	0.22	0.21	+ 16.67					
Total sugars%	17.70	17.39	17.12	16.90	- 4.52					
Ascorbic acid mg/100gm	86.0	65.0	47.0	32.0	- 62.79					
Total carotenoid mg/100gm	0.96	0.94	0.93	0.92	- 4.17					

Ascorbic acid content of nectar samples are presented in Table (3). Data indicated that, the loss of ascorbic acid content increased with increasing storage time. The loss in ascorbic acid were 37.5, 38.57, 56.52, 60.20% and 62.79% for nectar guava, guava-butternut squash (9 : 1), guava-butternut squash (8 : 2), guava-butternut squash (7 : 3) and guava-butternut squash (6 : 4) respectively, after 6 months of storage. Similar results were reported by (Kurata *et al.,* 1973) and (El-Gharably *et al.,* 2004). The loss of ascorbic acid could be ascribed to the oxidation of the vitamin during storage.

Results in Table (2) show the changes in total sugars of studied nectars stored at room temperature (25 ± 2°C) up to 6 months. It could be noticed that a slight reduction in total sugars were occurred among all tested samples stored at room temperature. The total sugar levels of the nectars stored up to 6 months were lower than those at initial storage. These results may be due to the chemical enzymatic reaction during prolonged storage to decompose total sugars to more simple compounds mentioned by (Canellas *et al.,* 1993).

The reduction levels in total sugars at the end of storage period could be arranged in an ascending order as follows 4.46%, 4.52%; 4.75% and 5.57% for (9 : 1 w/w), (6 : 4 w/w), (8 : 2 w/w) and (7 : 3 w/w) for guava-butternut winter squash respectively.

Carotenoids content of nectar samples are presented in Table (3). Data indicate that, a slight reduction in total carotenoids among all tested samples stored at room temperature. The total carotenoids levels of the nectars stored up to 6 months were lower than those at initial storage. The loss of carotenoid may be due to their conjugated double bond systems which make them labile to oxidative decomposition.

The reduction levels in total carotenoids at the end of the storage period could be arranged in an ascending pattern as follows 3.17%, 3.85%, 4.17% and 4.67% for (7 : 3 w/w), (9 : 1 w/w), (6 : 4 w/w) and (8 : 2 w/w) for guava butternut winter squash respectively.

Microbial exanimation

Total bacterial and yeast and mold counts of nectar samples were determined before storage and subsequently every two months along with six months of storage at ambient temperature. The obtained data are presented in Table (4). It could be observed that both bacterial as well as yeast and mold counts decreased with increasing the storage period since bacterial counts were 3.76, 4.25, 4.92, 5.20 and 5.80 x 10^3 at zero time of storage where as after six months of storage they were 0.42, 0.54, 0.58, 0.62 and 0.64 x 10^3 for guava, guava butternut winter squash (9 : 1 w/w), (8 : 2 w/w) , (7 : 3 w/w) and (6 : 4 w/w) respectively.

Table (4). Changes in total bacterial counts, yeast and molds for nectar samples during storage for 6 months at room temperature (25 ± 2°C)

Nectar samples	Total bacterial counts (X 10^3 /gm)				Change %	Yeast and molds counts (X 10^2 / gm)				Change %
	Storage period (months)					Storage period (months)				
	0	2	4	6		0	2	4	6	
Guava	3.76	1.90	0.69	0.42	- 88.83	5.60	2.23	1.20	0.45	- 91.96
Guava-butternut winter squash (9: 1 w/w	4.25	2.20	0.92	0.54	- 87.29	5.72	2.96	1.63	0.69	- 87.94
Guava-butternut winter squash (8 : 2 w/w)	4.92	2.84	0.98	0.58	- 88.21	6.35	3.86	1.92	0.72	- 88.66
Guava-butternut winter squash (7: 3 w/w)	5.20	3.00	1.05	0.62	- 88.08	6.98	4.32	2.00	0.83	- 88.11
Guava-butternut winter squash (6: 4 w/w)	5.82	3.20	1.24	0.64	- 89.00	7.32	4.80	2.21	0.92	- 87.43

Accordingly as for yeast and mold counts the initial counts before storage were 5.60, 5.72, 6.35, 6.98 and 7.32 x 10^2 where as after six month of storage they were 0.45, 0.69, 0.72, 0.83 and 0.92 x 10^2 for guava, guava butternut winter squash (9: w/w), (8: 2 w/w), (7: 3 w/w) and (6:4 w/w) respectively. The corresponding decreasing values for bacterial counts were 88.83, 87.29, 88.21, 88.08 and 89.00 where as the corresponding decreasing values for yeast and molds counts were91.96, 87.94, 88.66, 88.11 and 87.43. it could be included that both bacterial and yeast and mold counts were gradually decreased during ambient storage.

Sensory evaluation

The organoleptic evaluation of food products is of primary importance, since it reflects the consumer reference for a respective food product. Accordingly, it is necessary to run a test for the sensory aspects of the food before marketing to avoid any possible hazards that could be happened during marketability. The color, taste, odor and overall acceptability of consumer to a respective food material, will give a combined criteria for the organoleptic parameters.

Color, taste, odor and overall acceptability scores of prepared nectar samples made with guava fruit, guava-butternut squash (9 : 1 w/w), (8 : 2 w/w), (7 : 3 w/w) and (6 : 4 w/w) are presented in Table (5). It could be observed that, blended nectar prepared from guava- butternut squash (8: 2 w/w) had given the highest scores for color, odor and overall acceptability before and after storage at room temperature (25 ± 2°C) than other tested samples. Therefore, these samples came in the first order in comparison with other prepared nectars up to 6 months of storage at room temperature(25 ± 2°C); however, the nectar samples prepared from guava- butternut squash (9: 1 w/w) came in the second order. The prepared nectar from guava- butternut squash (6: 4 w/w) received un acceptable before and after for six months of storage at room temperature (25 ± 2°C).

Table (5). Sensory evaluation of nectar samples at zero time of storage and after storage period of six months at room temperature (25±2°C).

Nectar samples	Sensory attribute after preparation				Sensory attribute after 6 month of storage			
	Color	Taste	Odor	Overall acceptability	Color	Taste	Odor	Overall acceptability
Guava	8.40 ^{b}	9.10 ^{a}	9.0 ^{ab}	9.00 ^{ab}	8.20 ^{bc}	9.00 ^{ab}	9.00 ^{ab}	9.00 ^{ab}
Guava-butternut winter squash (9: 1 w/w	8.60 ^{b}	9.20 ^{a}	9.20 ^{A}	9.20 ^{a}	8.40 ^{a}	9.10 ^{a}	9.10 ^{a}	9.30 ^{a}
Guava-butternut winter squash (8 : 2 w/w)	9.30 ^{a}	9.60 ^{a}	9.60 ^{A}	9.50 ^{a}	9.20 ^{a}	9.50 ^{a}	9.40 ^{a}	9.40 ^{a}
Guava-butternut winter squash (7: 3 w/w)	8.20 ^{bc}	8.20 ^{bc}	7.50 ^{c}	8.00 ^{c}	8.00 ^{c}	7.30 ^{d}	7.62 ^{d}	7.30 ^{d}
Guava-butternut winter squash (6: 4 w/w)	6.40 ^{e}	6.50 ^{e}	5.20 ^{f}	5.60 ^{f}	6.00 ^{e}	5.20 ^{f}	5.00 ^{f}	4.80 ^{f}

* Value with different letters in the same column are significantly different at $p < 0.05$.

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تقييم الخواص الطبيعية والكيماوية والميكروبيولوجية للمشروبات المحضرة من مخاليط الجوافة والكوسة الشتوى

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الملخص العربي

تم إعداد خمسة خلطات من المشروبات بنسب مختلفة من الجوافة والكوسة الشتوي وهذة الخلطات هى الجوافة ، الجوافة – الكوسة الشتوي (بنسبة ٩ : ١) ، الجوافة – الكوسة الشتوي (بنسبة ٨ : ٢) ، الجوافة – الكوسة الشتوي (بنسبة ٧ : ٣) ، الجوافة – الكوسة الشتوي (بنسبة ٦ : ٤). ودرست الخواص الطبيعية والكميائية والميكروبيولوجية والاختبارات الحسية لهذة المشروبات بعد الإعداد وكذلك بعد التخزين لمدة ستة اشهر.

ولقد دلت النتائج المتحصل عليها حدوث زيادة تدريجية في الحموضة أثناء تخزين هذة المشروبات وحدوث فقد في محتوى العينات من حمض الاسكوربيك هذا الفقد يزيد مع زيادة فترة التخزين وكان النقص في السكريات الكلية والكاروتينويدات طفيفا أثناء التخزين.

أوضحت التحليلات الميكروبيولوجية لعينات المشروبات التي تم إعدادها حدوث نقص تدريجي في العدد الكلى للبكتريا والخمائر والفطريات بزيادة فترة التخزين

ومن ناحية أخرى أتضح من الاختبارات الحسية أن أفضل المشروبات التي تم إعدادها وتقييمها حسيا قبل وبعد فترة التخزين كانت المحضرة من خلطات الجوافة – الكوسة الشتوي (بنسبة ٨ : ٢) يليها خلطة الجوافة – الكوسة الشتوي (بنسبة ٩ : ١) ثم خلطة بنسبة (٧: ٣) ثم خلطة نسبة ٦ : ٤) على التوالي.