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Original Article

Sensory Acceptability and Shelf life of Prebiotic Pineapple Juice with Gum Arabic (*Acacia Senegal*) Addition

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ABSTRACT: Gum Arabic (GA) is a soluble dietary fibre exudate attained from *Acacia Senegal* has potential as a prebiotic agent in foods. This study was conducted to develop prebiotic pineapple juice using four different percentage of GA (*Acacia Senegal*) (0%, 4%, 8% and 12% of GA) in the juice. Sensory evaluation was conducted using an acceptance test and the most accepted formulation was chosen based on colour, odour, viscosity, taste and overall acceptability attributes. From this study, the sample with 4% of GA incorporation in the pineapple juice was selected and further analysed for its physicochemical and microbiological properties during chill storage (4 ± 1°C) for 9 weeks. Results showed that this sample retained the physicochemical quality for 9 weeks of storage except for redness, yellowness, turbidity and pH. At the end of the storage period, the mean of total plate count (TPC) and mould and yeast count (MYC) were recorded as 0.25 log CFU/ml and 3.83 log CFU/ml, respectively. However, the microbial loads were maintained at an acceptable level until two months (8 weeks) of chill storage. As a conclusion, the development of prebiotic pineapple juice by adding 4% of GA was feasible due to its high sensory acceptance and ability to retain its overall quality during chill storage.

Keywords: *Gum Arabic, Acacia Senegal, prebiotic, pineapple juice*

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INTRODUCTION

Prebiotic is the substances that are indigestible, stimulates the growth of beneficial intestinal microorganisms in the colon and thus cause a physiological effect which is beneficial to health[1,2]. Prebiotic food is a type of functional food that has been incorporated with prebiotic substances such as fructooligosaccharides (FOS),galactooligosaccharides, arabinose, galactose and inulin which are commonly used in present market[1].The fortification of functional beverages with prebiotic can achieve the modulation of gut microbiota by rising population of specific bacteria through consumption of prebiotic that selectively stimulates the growth of a limited number of bacteria residing in the colon. Gum Arabic (GA) has attracted attention due to its prebiotic properties[3]. Gum Arabic(GA) is a soluble dietary fibre, has potential as a prebiotic agent that can promote the growth of beneficial bacteria and inhibit the proliferation of harmful bacteria in the colon[3]. It is an edible biopolymer, released as exudates from Acacia trees such as Acacia Senegal or Acacia Seyal which grow widely in the African region of Sahe in Sudan[4].It was widely used as an emulsifier, stabilizer in foods and beverage and for surface coating due to its functionality of protective colloid. The properties of non-viscous and highly water solubility render GA as a substitute for beverages[5]. The suggestion of an optimal daily dose of GA is 10g, which can increase the numbers of Bifidobacteria, Lactobacilli and Bacteroidesin colon[5]. Therefore, this amount of ingestion should be considered in the formulation of prebiotic products such as prebiotic fruit juice if using gum Arabic as the prebiotic source.

Other than that, several studies have been carried out to develop fruit juices fortified with prebiotics like FOS and inulin in apple and orange juice[6,7]. It was found that the juices incorporated with FOS had shown stability on physicochemical properties (colour, pH and titratable acidity) and no microbial activity during two months of storage at temperature 4°C[6]. However, the majority of the studies mainly used FOS and inulin in the fruit juice. Limited studies were carried out using GA especially on tropical fruit such as pineapple which contains various nutrients like vitamin C, vitamin B1, vitamin B6, dietary fibre, and potassium[8].

Therefore, this study was carried out to evaluate the sensory acceptability and the shelf life of prebiotic pineapple juice with Gum Arabic addition during storage at chill temperature (4ºC) for 9 weeks.

MATERIALS AND METHODS

Raw materials and preparation of prebiotic pineapple juice

Pineapple (*Ananascomosus*) of the variety of Josaphine was purchased from a retail market in Kuala Terengganu, Malaysia. Pineapple with maturity stage of a full-yellow stage was chosen for juice extraction. The crown and leaves were removed and the fruit (with skin) was pre-washed with sterilized water before peeling process was carried out. The fruit cylinders were cut into strips and being pulped in fruit juice (National MJ-68M, Malaysia) and filtered with a muslin cloth to remove the fruit pomace. Juice preparation was carried out by diluting the juice with 50% of water. The diluted juice was added with 0 (control), 4, 8 and 12% GA before the heating process. Then, juice was pasteurized (95ºC, 30 seconds) and was added with citric acid to adjust the pH to 3.5±0.2[9]for microbial stability. In the meantime, cleaned amber glass bottles were boiled for 10 minutes and dried in the oven at 110ºC for 15 minutes. The juice was poured in a still hot bottle by hot filling technique and then cooled to 30ºC by inserting the filled bottle into cold water before being stored in the chiller at 4ºC.

Sensory evaluation test

Sensory evaluation was conducted for prebiotic pineapple juices containing different concentrations (0%, 4%, 8% and 12%) of GA to evaluate the acceptance level of samples by 35 untrained panellists from Universiti Malaysia Terengganu (UMT) based on sensory attributes including colour, odour, viscosity, taste and overall acceptability. A 7-point hedonic scale (1 represents dislike extremely while 7 represents like extremely) was used in this evaluation. The most accepted prebiotic pineapple juice was selected for shelf life study.

Shelf life study of prebiotic pineapple juice

The most acceptedprebiotic pineapple juice during the sensory evaluation test and the control juice(0% gum Arabic) were further analysed for shelf life study. The samples were stored at chill (4 ± 1°C) for 9 weeks and were analysed for their physicochemical and microbial properties at every 7 days interval.

Physicochemical properties

A colorimeter (Konica Minolta, Japan) was used to measure the yellowness (b*) of the prebiotic pineapple juices. For the turbidity, the prebiotic pineapple juicewas centrifuged(Gyrozen 1580R, Korea) at 3000 rpm for 10 minutes and the supernatant was collected. The collected supernatant was filled in the plastic cuvette and the turbidity was measured using spectrophotometer (Genesys 20, UK) at 650nm[10] wavelength.The total phenolic content was analyzed (at week 0 and 9 only) using the modified procedure of Folin-Ciocalteu metal reduction assay[11,12]with gallic acid as a standard. Prebiotic pineapple juice was centrifuged at 3000 rpm for 10 minutes and the supernatant was diluted by five times. Approximately 0.1ml of diluted juice sample was taken and added with about 0.5ml of Folin-Ciocalteu (2N) reagent and 0.6ml of distilled water and left for 3 minutes. Approximately 1.5ml of 7% sodium carbonate was then being added to the sample for neutralization. After 7 minutes, the sample was diluted to a final volume of 10ml with deionised water and the sample was vortexed at 2000 rpm for 1 minute. After 30 minutes of reaction in a water bath at 40ºC, the sample and standard were pipetted into a cuvette and the absorbance were read at 726nm (wavelength) using a spectrophotometer. The total phenolic content was determined from a standard concentration curve and reported in mg/L equivalents of Gallic acid.

Microbiological properties

Total plate count (TPC) was used to evaluate the microbiological quality of the prebiotic pineapple juice. TPC determination was carried out at 7 days interval for 9 weeks. The TPC procedure was conducted based on Bacteriological Analytical Method (BAM) used by Maturin and Peeler with modification[13]. About 10 ml from each sample was added into 90ml of 0.1% sterile peptone water (sample is diluted to 10^{-1}) and vortexed. Serial dilutions of the homogenate (10^{-2} and 10^{-3}) were made. Then, about 0.1ml of each dilution was pipetted onto Plate Count Agar (PCA) and spread by using L-spreader aseptically. Plates were covered after inoculum soaking in the agar. The plates were incubated at 37±1ºC for 24 hours. After incubation, the colonies on the plates in the range 30-300 were counted in CFU/ml.

Mould and yeast count (MYC) was carried out with the procedure described by Tournas et al. with slight modification[14]. MYC determination was carried out at 7 days' interval for 9 weeks. The procedures were similar to the determination of total plate count (TPC) but the agar used was replaced withDichloran rose bengal chloramphenicol (DRBC) agar. The plates were then incubated for 5 days at 25ºC. After incubation, the colonies in the ranges of 10-150 were counted in CFU/ml.

Statistical analysis

Statistical application, Minitab 17 software (Minitab Inc., USA) was used to analyse the data obtained from the microbiological, physical and chemical analyses as well as sensory evaluation at 95% confident level (α=0.05). One-way analysis of variance (ANOVA) was conducted to determine whether there was a significant difference between the prebiotic pineapple juices. Fisher's Least Significant Difference (LSD) was further carried out to determine the significant difference among the independent variables if significant difference occurred between the means of data ($p<0.05$). Two-way ANOVA was conducted to determine the interaction between the formulation and storage time.

RESULTS AND DISCUSSION

Sensory evaluation

Figure 1 shows no significant difference ($p>0.05$) in the colour acceptability of all prebiotic pineapple juice samples (A, B, C and D). Juice with 12% GA (sample D) showed the lowest odour and viscosity acceptability level with no differences ($p>0.05$) with juice that contains 8% GA. Significant low ($p<0.05$) acceptability on viscosity was obtained by sample D indicated that too high viscosity (47.5 mPas) juice (unpublished data) did not accept by the panellist. The acceptance level of taste was also reduced with increased in GA concentration. For instance, sample D which had the highest total soluble solids (18.1ºBrix) and titratable acidity (0.373%)among the samples (unpublished data) showed a negative impact on the taste and overall acceptability of the finished product.

Sample B (4% GA) with acceptability mean score of 5.2 ± 1.3 (overall acceptance) was chosen as the most accepted sample in this study. This sample was further investigated for shelf life study in term of microbiological and physicochemical properties of the prebiotic pineapple juice.

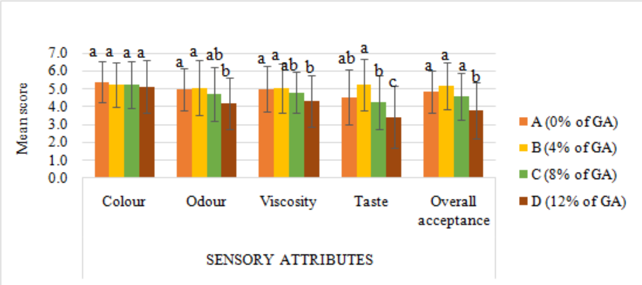

Figure 1: Sensory acceptability mean scores of prebiotic pineapple juices with different concentration of gum Arabic; 0% GA (■), 4% GA (■), 8% GA (■), 12% GA (■). Bars represent the mean score and the error bars represent standard deviation (n=35). Bars with similar letters on the histogram are not significantly different(within the same attribute)($p>0.05$).

Shelf life study of prebiotic pineapple juice at 4ºC for 9 weeks

The study of physicochemical properties and microbiological quality of pineapple juices (0 and 4%) during storage were evaluated based on two main effects (factor) which were formulation and storage duration. The interaction effect between those factors was first determined before analyzing the variable differences (for each response).

Yellowness (b*)

There was a significant interaction effect ($p<0.05$) between the formulation and storage time in b* values of pineapple juice. As shown in Figure 2, the b* values of both formulations decreased with storage time but control sample showed the higher values. Since yellow is the major colour of pineapple juice, thus the amount of yellow-associated pigment is an excellent indicator to measure the quality[17]. Morgan stated that the yellow colour of pineapple juice is proportional to the total carotenoid concentration[18]. Wibomo et al. also found that the decrease of yellowness in pasteurized orange juice was due to degradation of major carotenoid (α-carotenoid and β-carotenoid)[19].

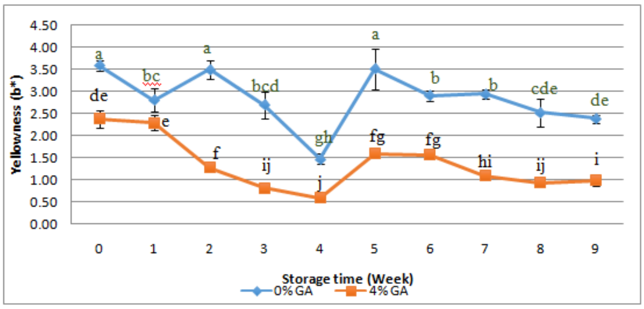

Figure 2: Yellowness (b*) value of pineapple juices with 4% GA (■) and without GA (◆) throughout 9 weeks of chill storage (4ºC). Points are the experimental data that expressed in mean and the error bars represent standard deviation (n=3). Points with the different letters on the chart are significantly different between samples and storage time ($p<0.05$).

Turbidity

There was an interaction effect ($p<0.05$) between the concentration of GA and storage time on the turbidity of pineapple juices. Based on Figure 3, the turbidity of pineapple juice (with 4% of GA) gradually decreased during storage while the pineapple juice without GA (control) kept stable throughout the storage time. The stability of turbidity in control during storage was in agreement with the findings of Tandon et al.who reported that the turbidity of hot-filled pasteurized apple cider remained constant during storage[20]. Present results indicated that the cloud stability of pineapple juice with GA had decreased slightly with storage time. Babbar et al. depicted the cloud loss in juice during storage could be attributed to the coalescence of tiny insoluble droplets of oxidized phenolic compounds into dispersed larger droplets[21]. Besides, the high turbidity of pineapple juice with 4% of GA caused by the electrostatic repulsive force between negatively charged colloid (GA) and protein[22].

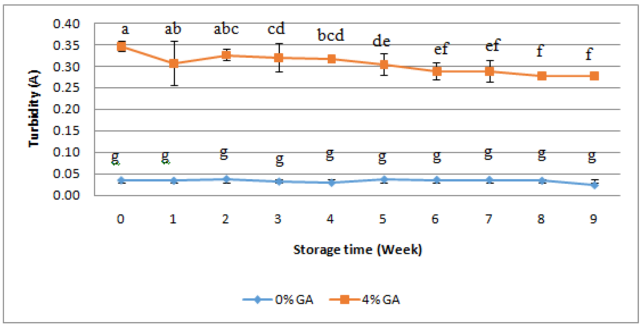

Figure 3: Turbidity of pineapple juices with 4% GA ( ) and without GA ( ) throughout 9 weeks of chill storage (4°C). Points are the experimental data that expressed in mean and the error bars represent standard deviation (n=3). Points with the different letters on the chart are significantly different between samples and storage time ($p<0.05$).

Total phenolic content

There was no interaction effect ($p>0.05$) between the concentration of GA and storage time on the total phenolic content of pineapple juices. From Table 1, the amount of phenolic content in the pineapple juice with GA (20.62 mg GAE/100 ml) was significantly higher as compared to the pineapple juice without GA (15.23 mg GAE/100ml). Therefore, it can be seen that the inclination of the amount of phenolic content in pineapple juice with GA was attributed to the phenolic contents of GA. As supported by Amoussa et al. and Lowry et al., it was also reported that *Acacia species*contain phenolic[23,24].In addition, GA was also reported to show its ability to minimize the oxidation process of phenolic compounds by protecting them from environmental conditions like oxygen and temperature[25].After 9 weeks of chill storage, the amount of phenolic content in both pineapple juices were observed to be similar to the initial amount ($p>0.05$).It may due to the absence of polyphenol oxidase (PPO) which can cause the phenolic to oxidize[26], causing it to decrease throughout the storage.Therefore, the absence of PPO might help in preserving this compound during storage. As supported by Krapfenbauer et al., polyphenol oxidase (PPO) in pineapple juices can be completely inactivated after treatment of high-temperature short time (HTST) at 80°C[27]. This might be the reason on why the amount remains during chill storage.

Table 1: Total phenolic content of pineapple juice with GA (4%) and without GA at week 0 and 9th week of chill storage

Formulation of pineapple juice	Storage time		The concentration of gallic acid (mg GAE/100 ml)
	Week 0	Week 9	
0% GA (Control)	17.70 ± 1.63	12.75 ± 0.07	15.23[B]
4% GA	21.67 ± 1.14	19.05 ± 0.71	20.62[A]
Concentration of gallic acid (mg GAE/L)	20.08[a]	15.90[a]	

Data represent a mean and standard deviation from two independent replications for each sample.
Value with the same superscript letter ([a])in the same row are not significantly different ($p>0.05$).
Value with different superscript letters([A-B])in the same column are significantly different ($p<0.05$).

Microbiological quality

Total plate count (TPC)

There was no interaction effect ($p>0.05$) between the concentration of GA and storage time on the total plate count (TPC) of prebiotic pineapple juice. Control sample exhibited a higher mean value of TPC counts (1.34 log CFU/ml) as compared to pineapple juice with 4% GA (0.25 log CFU/ml). The TPC of pineapple juice with 4% GA remained stable until week 8 and increase significantly at week 9.The addition of GA in pineapple juice had significantly decreased the microbial load in the samples. Previous findings described that the antimicrobial activity of GA has been attributed to the presence of secondary metabolites and terpenes that are included in the group of phytoprotectants and also others such as the salts, cyanogenic glycosides and enzymes in polysaccharides in GA[28]. The data in Table 2 shows that the juices with and without GA were still acceptable for microbiological safety since the microbial loads at the end of 9 weeks of storage were below the limit recommended by Food and Environment Hygiene Department which is 7 CFU/ml in pasteurized food requiring refrigeration including fruit juices[29].

Table 2: Total plate count (TPC) of pineapple juices with GA (4%) and without GA throughout 9 weeks of chill storage

Formulation of pineapple juice	Storage Time (Week)										TPC (log CFU/ml)
	0	1	2	3	4	5	6	7	8	9	
0% GA (Control)	0.00 ± 0.00	1.15 ± 1.63	1.00 ± 1.41	1.15 ± 1.63	1.00 ± 1.41	1.35 ± 1.91	1.00 ± 1.41	1.00 ± 1.41	2.69 ± 0.13	3.05 ± 0.21	1.34^{A}
4% GA	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	2.45 ± 0.21	0.25^{B}
TPC (log CFU/ml)	0.00^{b}	0.58^{b}	0.50^{b}	0.58^{b}	0.50^{b}	0.68^{b}	0.50^{b}	0.50^{b}	1.35^{ab}	2.75^{a}	

Data represent a mean and standard deviation from two independent replications for each sample.
Value with the same superscript letter (^{b})in the same row are not significantly different ($p>0.05$).
Value with different superscript letters ($^{A-B}$)in the same column are significantly different ($p<0.05$).

Mould and Yeast count (MYC)

Based on Figure 4, the interaction of the concentration of GA and storage time had a significant effect on the MYC of pineapple juice. MYC of pineapple juice with 0% of GA appeared at 5thweek, however pineapple juice with 4% of GA had MYC at one week earlier than the former. Starting from 7th week, the MYC of pineapple juice with 0% of GA was significantly higher than that in pineapple juice with 4% of GA. The MYC for pineapple juice without GA was 5.43 logCFU/ml while the pineapple juice with 4% of GA was 3.82 log CFU/ml by the end of the 9th week of storage. This finding indicated that the growth of yeast and mould in pineapple juice without GA (0%) was faster than that with juice with GA addition. This growth retardation may be attributed to antimicrobial properties of GA[28]. A similar result was shown in findings of Nwaokoro and Akanbi, who found that the tomato-carrot juice blend with hydrocolloid (carboxylmethylcellulose, xanthan gum and guar gum) and stored at refrigeration temperature had obtained lower yeast and mould count as compared to the juice without the hydrocolloids[30].According to Gulf Standards (2000), the maximum yeast and mould count permitted for juices sold in the Gulf region are 3 log CFU/ml[31]. The result revealed that pineapple juice with 4% of GA can still be acceptable before the 8th week of storage under recommendation from Gulf Standards for fruit juices.

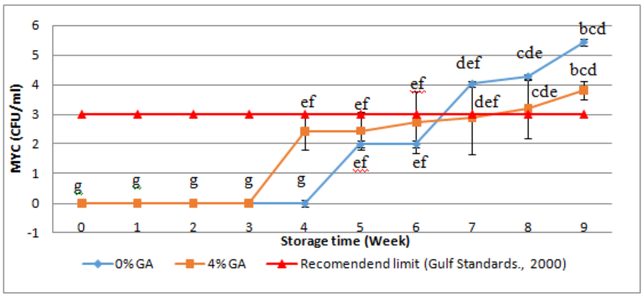

Figure 4: Mould and yeast counts (MYC) of pineapple juices with 4% GA (orange line) and without GA (blue line) throughout 9 weeks of chill storage (4°C). Points are the experimental data that expressed in mean and the error bars represent standard deviation (n=3). Points with the different letters on the chart are significantly different between samples and storage time ($p<0.05$).

CONCLUSION

All samples with GA had similar sensory acceptability level on attributes of colour, odour, viscosity, taste and overall acceptability except for sample with 12% of GA addition. The prebiotic pineapple juice with 4% of GA was chosen as the most accepted product.For shelf life study, yellowness of the juices decreased with storage time, with control sample showed higher values compared to juice with GA. Sample with GA showed a steady decreasing trend in turbidity. The total phenolic content of juice with 4% GA was higher than control and remained stable during chill storage.In conclusion, prebiotic pineapple juice with 4% GA remained safe for consumption for up to two months of chill storage.

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