THE USE OF STEVIA AND MONK FRUIT SWEETENERS FOR SUGAR REPLACEMENT IN GREEN TEA AGAR JELLIES

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ABSTRACT

This study aimed to develop healthy agar jellies with low-sugar content, linking to a decreased risk for heart attack, diabetes, and other chronic diseases. Two natural non-nutritive sweeteners, stevia and monk fruit were studied at 25%, 50%, 75%, and 100% sugar replacement based on the equivalent sucrose sweetness. The physical properties assessed on the total soluble solids, firmness, color, and syneresis. Still, the sensory attributes on color, taste, flavor, texture, and overall acceptability was evaluated by a 9-point hedonic scale. The more sugar replacement lowered the total soluble solids but increased the gel firmness ($p<0.05$) and the lightness (L^*). The syneresis declined in reduced-sugar jellies with stevia and monk fruit against the whole sugar jellies. The stevia and monk fruit replacement affected agar jellies' taste and flavor attributes ($p<0.05$). Jellies with 50% and 75% sugar replacement, either stevia or monk fruit, were preferred over 25% and 100% replacement. The jelly with 50% stevia was the optimal formulation.

Keywords: Agar jelly, healthy dessert, monk fruit, stevia, sugar replacement

INTRODUCTION

The global growth of the sugar-free food and beverage market has risen to a 9.36% CAGR during the forecast period (2021-2026) (Mordor intelligence, 2021). Increasing concerns regarding chronic diseases, such as hypertension, heart disease, diabetes, and obesity, have correlated with new free or lower sugar foods introduced to the food market. The sugar level limit by the American Heart Association (2021) recommended most men and women should consume no more than 36 g and 25 g of added sugar per day. Replacing sugary foods and drinks with non-nutritive sweeteners, a substance that provides a sweet taste and few or no calories but offers no nutritional benefits such as vitamins and minerals can help people gain weight, tooth decay, and better health. Artificial non-nutritive sweeteners widely available for the food industry are saccharine, acesulfame-K, aspartame, and sucralose; these are considered safe under an acceptable daily intake (ADI) level of each sweetener approved by the FDA. However, two plant-based sweeteners, including stevia and monk fruit extracts, have been increasingly applied in food products because they are natural and Generally Recognized as Safe (GRAS) for use (Tey *et al*., 2017).

Stevia, or *Stevia rebaudiana*, is an intensely sweet-tasting, zero-calorie plant extract providing 120-200 times sweeter than sucrose. It has a negligible effect on tooth decay, blood glucose, and insulin levels; it is attractive to people with health concerns (Gao *et al*., 2016). Stevia is commonly heat and pH stable, maintaining its sweetening power during processing and does not caramelize; thus, it has the potential to be widely used. Sometimes it tastes bitter, has a licorice-like or metallic aftertaste, mainly used in high concentrations (Goyal *et al*., 2010). In the meantime, monk fruit (*Siraitia grosvenoriiis*), commonly known worldwide as Luo Han Guo, is a gourd native to South East Asia. It is a zero-calorie sweetener with compounds called mogrosides, having high biological effects and sweet taste (Li *et al*., 2014). In 2010, the extract of monk fruit was approved in the USA as GRAS for non-nutritive sweetening and flavor-enhancing purposes. It is around 150-250 times the sweetness of sucrose, pH and heat stable, and has no tooth decay and side effects but may cause rare digestive issues if consuming too much of it, such as nausea and upset stomach. Some people notice an unpleasant aftertaste at its high concentrations. In recent years, pharmacological studies reveal the monk fruit has several health-protective properties such as liver protection, anti-oxidative, anti-inflammatory, anti-hyperglycemic, and anti-cancer effects (Pandey and Chauhan, 2020). Monk fruit is becoming more popular in the natural health sector, but it is more expensive than other sweeteners, especially it costs more than twice that of stevia.

The sugar reduction frequently has caused changes in product characteristics and sensory acceptability against full-sugar counterparts. Several non-nutritive sweeteners are permitted for use in foods with sugar reduction on a sweetness equivalency basis. However, the sweeteners are only successful if they denote a similar sensory profile as sugar. To date, information comparing the quality of low-calorie products with stevia against monk fruit is little. Therefore, this study assessed the physical properties and sensory profile of agar jellies with sugar replacement by stevia and monk fruit. The optimal formulation with palatable and acceptable was also investigated.

MATERIALS AND METHODS

Materials

Agar powder (Platapiantong Seng Huad Co., Ltd., Thailand), green tea powder (Cha Tramue, Siam F.B. Product Co., ltd., Thailand), and sugar were purchased from a supermarket. Stevia (BetterSteviaTM, Now Foods, IL, USA) and monk fruit extract (NuNaturals, Inc, OR, USA) were used.

Experimental Design

Eight green tea agar jellies with sugar replacement by stevia and monk fruit were prepared. The sugar replacement was calculated as equivalent sweetness basis; 45 mg of stevia is equivalent to 6 g of sugar, and 35 mg of monk fruit is equivalent to 4 g of sugar. Thus, the stevia amounts used for replacing 25%, 50%, 75%, and 100% sugar content (16 g in recipe) was 0.03, 0.06, 0.09, and 0.12 g, respectively. At the same time, the monk fruit used was 0.035, 0.07, 0.105, and 0.14 g, respectively.

Green Tea Agar Jelly Production

A regular agar jelly recipe included 1 g agar powder and 16 g sugar in 100 mL green tea water. Initially, a green tea bag (1 g) was soaked in a 100 mL hot water (80 ± 2 °C) for 20 min. Added agar powder, followed by sugar, and thoroughly stirred for 5 min. Poured the hot mixture (60 ± 2 °C) into cups (2 × 4 cm^2) and refrigerated at 4 °C for 20 min.

Physical Analysis

Total soluble solid was measured by a hand refractometer (Atago®, Fukaya factory, Saitama, Japan). A texture analyzer (LRX Plus, Lloyd Instruments, Hampshire, UK) was performed for the firmness measurement. The instrument was equipped with a cutting test cell and operated at a crosshead speed 200 mm/min. The peak force (N) was recorded and reported as the firmness.

Color analysis was determined using a colorimeter (MiniScan EZ, Hunter Associates Laboratory, Reston, VA). L^* (lightness) (100 = white, 0 = black), a^* (+ = red, − = green) and b^* (+ = yellow, − = blue) were recorded.

A gel sample packed in a polyethylene bag was stored in a refrigerator (10 ± 2 °C) for 2, 4, and 6 days. The syneresis was determined by the difference of sample weight before and after storage to sample weight, as according to Banerjee and Bhattacharya (2011) with some modification.

Table 1. Physical properties of green tea agar jellies with different sweeteners

Treatment	Sugar replacement (%)	Total soluble solid (° brix)	Firmness (N)	L^* (Lightness)	a^* (Redness)	b^* (Yellowness)
Control	-	18.1 ± 0.30^a	5.05 ± 0.17^d	2.16 ± 0.13^d	7.28 ± 0.01^a	3.57 ± 0.14^c
Stevia extract	25%	6.9 ± 0.48^b	5.29 ± 0.09^c	2.85 ± 0.12^b	5.53 ± 0.02^c	4.25 ± 0.05^b
	50%	5.2 ± 0.25^c	5.65 ± 0.15^c	2.44 ± 0.01^c	5.75 ± 0.18^c	4.36 ± 0.14^b
	75%	3.2 ± 0.22^d	5.80 ± 0.66^c	2.36 ± 0.03^c	5.56 ± 0.06^c	4.65 ± 0.05^b
	100%	1.8 ± 0.18^d	6.21 ± 0.13^b	2.51 ± 0.05^c	5.86 ± 0.04^c	4.36 ± 0.03^b
Monk fruit	25%	7.1 ± 0.35^b	6.47 ± 0.03^b	3.43 ± 0.04^a	6.44 ± 0.03^b	5.20 ± 0.06^a
	50%	5.0 ± 0.38^c	6.86 ± 0.25^b	3.09 ± 0.09^b	6.83 ± 0.08^b	5.23 ± 0.17^a
	75%	2.8 ± 0.12^d	7.19 ± 0.10^a	3.08 ± 0.21^b	6.45 ± 0.05^b	5.74 ± 0.05^a
	100%	1.5 ± 0.06^d	7.28 ± 0.34^a	3.22 ± 0.14^a	6.33 ± 0.02^b	5.69 ± 0.06^a

Means in the same column with different superscripts are different ($p<0.05$).

Sensory Evaluation

The sensory test was performed in an individual booth, and served with samples coded with random three-digit numbers. Sixty panelists, who regularly consume jellies, were instructed to assess the color, taste, flavor, texture, and overall acceptability. Sensory evaluation was assessed using a 9-point hedonic scale (1 = extremely dislike, 9 = extremely like). All panelists were invited to clean and rinse their palates between samples with drinking water (Lawless and Heymann, 1998).

Data Analysis

The statistical analysis was conducted on analysis of variance (ANOVA) and Duncan's new multiple range test using the SPSS version 17.0 (Cochran and Cox, 1992). Principal component analysis was performed by the R-program for the external preference mapping (R Foundation for Statistical Computing, Vienna, Austria).

RESULTS AND DISCUSSION

Agar jelly characteristics

The impact of sugar replacement with stevia and monk fruit on the characteristics of agar jellies are presented in Table 1.

a. Soluble solids in agar gel

The control jelly was made from the whole sugar, showing the highest soluble solid content. Sugar provides a soluble solid contributing to a significant relation to °brix. With increased sugar replacement by both stevia or monk fruit, the samples significantly declined in soluble sugar. A small amount of these sweeteners was used instead of sugar because of their high sweeter than sucrose.

Thus, it negligibly raised a soluble solid in jellies. The finding was in line with the study of Bajwa and Mittal (2015) for the reduced-sugar milk drink supplemented with mango pulp and sucralose. It was agreed by Rocha and Bolini (2015), who studied the sweetness perception of passion fruit juices with sucrose and high-intensity sweeteners, including aspartame, stevia extract, sucralose, and neotame.

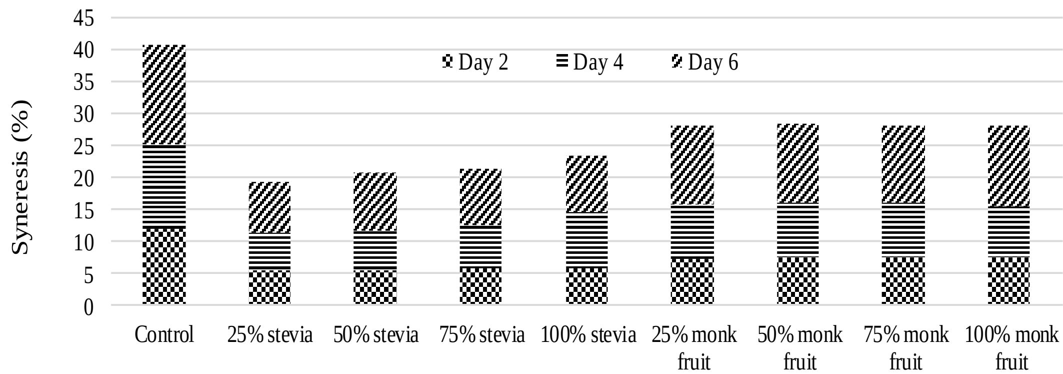


Figure 1. Syneresis of agar jellies with different levels of sugar reduction for 6 days at refrigerated storage.

b. Gel texture

Higher gel firmness in agar jellies was associated with increasing sugar replacement (Table 1). The control sugar jelly was rigid and brittle and had easily broken down, showing the lowest firmness. The more decreasing sugar content has caused the jellies with stevia and monk fruit less brittle and elastic but more cohesive, presenting a higher firmness. This observation denotes that sugar has a considerable effect on the gel. The high sugar content can reorganize the agarose bonds, contributing to the formed agar gel (more elastic but less cohesive) (Dorohovich *et al*., 2018). The sugar itself might tighten the cross-linking of the agar molecules by its high water-binding capacity for the hydration of free water in the gel, greatly structuring the gel network (Saha and Bhattacharya, 2010). Vilela (2015) showed lower gel strength of pectin jellies with one-third sugar reduction with sorbitol and xylitol against the sugar control. Lau *et al*. (2000) suggested that the strength of the gel structure is associated with the type and concentration of hydrocolloids levels and the amount of total solids content in the gel system.

c. Color

According to Table 1, increases in *L** and *b** values were observed in agar jellies with stevia and monk fruit, but *a** values were decreased ($p<0.05$). The control had the lowest *L** or darker than the jellies with both sweeteners. An explanation might involve the jelly chromaticity derived from the mixed color between green tea and brown pigment by the sugar-amine reaction through the Maillard browning formation (Alais and Linden, 1991). In other words, stevia and monk fruit jellies were lighter or more transparent with high intensely yellow. This is related to their lower sugar participating in the brown formation reaction; the less sugar content, the less the intensity of darker color. According to Gao (2016), the lower *a** might be due to lowering the availability of sugar amounts to form brown pigments through the non-enzymatic browning reaction. When considering the *h*° value, the control had the lowest (*h*° = 26.1), relating to a reddishorange shade, while the jellies with stevia (*h*° = ~37.8) and monk fruit (*h*° = ~40.2), which became more yellowish-orange tonality due to higher *h*° (data not shown). The monk fruit extract has light yellow, making jelly with higher *h*° than the stevia, a white powder.

Table 2. Sensory evaluation of green tea agar jellies with different sweeteners

Treatment	Sugar replacement (%)	Color	Taste	Flavor	Texture	Overall acceptability
Control	-	6.38±1.41^{b}	6.96±1.49^{a}	6.67±1.76^{a}	6.54±1.53^{a}	6.96±1.52^{a}
Stevia extract	25%	6.57±1.10^{b}	4.33±1.55^{d}	5.33±1.43^{c}	5.65±1.36^{b}	4.83±1.37^{b}
	50%	6.71±1.30^{ab}	6.50±1.41^{b}	6.29±1.55^{b}	6.58±1.32^{a}	6.68±1.40^{ab}
	75%	6.98±1.29^{a}	6.75±1.51^{b}	6.30±1.52^{b}	6.25±1.98^{ab}	6.95±1.58^{a}
	100%	6.95±1.35^{a}	4.00±1.77^{d}	5.21±1.56^{c}	5.04±1.85^{c}	4.42±1.35^{c}
Monk fruit	25%	6.50±1.57^{b}	5.71±1.43^{c}	5.64±1.38^{c}	6.00±1.50^{b}	5.50±1.14^{b}
	50%	6.72±1.54^{ab}	6.08±1.21^{b}	6.33±1.31^{b}	6.54±0.93^{a}	6.63±1.06^{ab}
	75%	7.08±1.47^{a}	6.63±1.79^{b}	6.45±1.70^{b}	6.42±1.56^{a}	6.80±1.67^{a}
	100%	6.85±1.79^{a}	4.71±1.94^{d}	5.54±1.56^{c}	5.25±1.90^{c}	4.83±1.70^{c}

Means in the same column with different superscripts are different ($p<0.05$)

d. Water weeping

Figure 1 shows the results of water expelling determined in the control and agar jellies with stevia and monk fruit during 6 days in refrigerated storage (10 ± 2 °C). Syneresis, a liquid that is expelled or extracted from a gel, is undesirable because the wetness can cause unacceptable changes in texture, smell, and appearance, sensory preference and promote the growth of microbiological spoilage. The result revealed a higher syneresis in the control jelly than those with sugar replacement at every 2-day interval storage. In general, the agar gel forms a meshwork that contains pores, which have sizes depending on the concentrations of agarose and sugar. The agar gel with sugar may have large pores in the gel structure, and it would contribute to shrinking over and releasing higher water during aging (Somboon *et al*., 2014). The decrease in syneresis was related to lowered total solids in the samples with stevia and monk fruit inclusion. The low amount of sugar content could affect the internal hydrophobicity by incorporating the water-binding capacity of sugar. It might cause more reduced pore sizes in the agar gel matrix, lowering the water expelling from reduced-sugar agar jellies (Divoux *et al*., 2015). It noted that the syneresis was decreased mainly in the agar jelly with stevia compared to that with monk fruit.

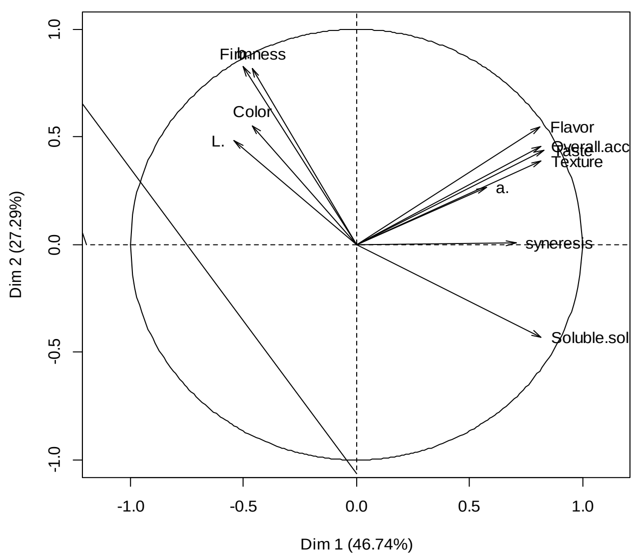
Sensorial preference

a. 9-point hedonic scale

The variation of sensory attributes was observed in samples sweetened with stevia and monk fruit against the control made from the sugar, as presented in Table 2. Jellies with stevia and monk fruit were transparent than the control, particularly at 75% and 100% replacement, thus raising their color scores. The level of sugar replacement was crucial on the preference of taste, flavor, and texture, showing their higher scores ($p<0.05$) in the jellies with 50% and 75% sugar replacement relative to 25% and 100% replacement. Most panelists disliked the aftertaste (bitter/metallic), thereby rejecting the jellies with 100% sugar replacement. They preferred the balanced sweetness from blending stevia or monk fruit with white sugar based on the equivalent sweetness. Since each sugar replacer has a sweeter taste than cane sugar (Tey *et al*., 2017), the panelists might find the jellies with 50% and 75% sugar replacement had taste and flavor in the right level. With stevia and monk fruit inclusion, the 75% sugar replacement samples received the highest overall acceptability but were not statistically significant (p>0.05) compared to 50% replacement. The products were acceptable, corresponding to the "like slightly" response.

The ANOVA result shows whether the differences between the attributes are statistically significant according to sugar replacement. It does not disclose the most appreciated sample. The preference mapping was used to depict the main attributes affecting the liking and determine the best sample.

(a) Variable projection



(b) Sample projection

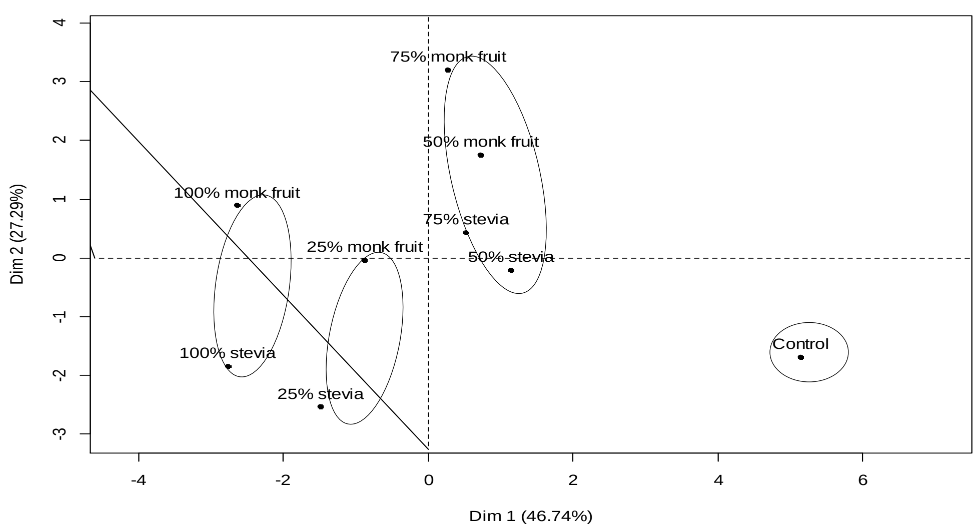


Figure 2 Preference mapping of agar jellies: (a) variable projection and (b) sample projection

b. Preference mapping

Figure 2a shows that Dim 1 can explain 46.74% of the total variability, followed by Dim 2 with 27.29%, and thus the total is 74.03% of the total variability. The high value assured that consumers could satisfactorily discriminate among the jellies. Most sensory attributes, such as taste, flavor, texture, and overall acceptability, were positively correlated in Dim 1. Still, Dim 2 positively correlated with firmness and $L*$ and $b*$ values. The result demonstrated a clear distinction between the four groups. The first is the control, followed by samples with 50% and 75% stevia and monk fruit replacement, samples with 25% stevia and monk fruit replacement, and samples with 100% stevia and monk fruit replacement are the last, respectively (Figure 2b). It indicates a significant distribution of the prominent discrimination subjecting to the level of sugar replacement. The control, positioned in the positive quadrant, was characterized by the preferable sensory attributes (Figure 2b). Moreover, the control was also characterized by the higher soluble solids, which linked to a full-sugar recipe. This observation emphasizes the role of sugar on the agar jelly texture, as the gel is brittle with a reduced firmness value, corresponding to a highly desirable texture. The more sugar replacement, the lower the sensory perception because the sugar influences the taste and flavor of the samples. Most panelists were not accustomed to the taste quality and negative attributes, such as unpleasant aftertaste of the sweeteners, reflecting lowered their sensory evaluation. Consequently, samples with 100% sugar replacement were far from the control, showing less acceptable (Figure 2b). At the same time, the sample with 50% stevia replacement was located close to the control against other samples, implying that it was more preferred.

CONCLUSION

Stevia and monk fruit can be used to partially replace sugar content in green tea agar jellies. The level of sugar replacement was significant on the quality of the jellies. Total soluble solids and syneresis were decreased in reduced-sugar samples, but instrumental firmness increased. The combination of sugar and stevia or monk fruit produced a mixture that has a unique taste and flavor. Jellies with stevia and monk fruit had more transparency and lower scores for taste and flavor than the control. Panelists preferred the samples with 50% and 75% sugar replacement with stevia or monk fruit to those with 25% replacement. In contrast, they denied the samples with 100% replacement. This result suggests basic information on development of reduced-sugar jellies made with stevia or monk fruit. The storage stability of these jellies should be further study.

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