Replacement of gelatin with liquid *Opuntia ficus-indica* mucilage in marshmallows. Part 1: Physical parameters

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ABSTRACT

The objective of this study was to replace gelatin in marshmallows with different concentrations of fluid mucilage, combined with different concentrations of powdered hydrocolloids. Nine different formulations were prepared: control (100% gelatin); 75% mucilage + 25% xanthan (MX); 75% mucilage + 25% agar-agar (MA); 75% mucilage + 25% guar (MG); 80% mucilage + 20% xanthan (8M2X); 80% mucilage + 20% agar-agar (8M2A); 80% mucilage + 20% guar (8M2G); 75% mucilage + 12.5% xanthan + 12.5% agar-agar (MXA); 75% mucilage + 12.5% xanthan + 12.5% guar (MXG); and 75% mucilage + 12.5% agar-agar + 12.5% guar (MAG). Consistency, texture, tenderness of gel and shear measurements were determined, along with color (L* values, as well as C* and Hº values) and a_w. There were significant ($p<0.05$) differences between the different samples for all measurements. The best formulation for gelatin replacement was found to be the 75% mucilage + 12.5% xanthan + 12.5% agar combination (MXA), as it only differed significantly ($p<0.05$) from the control (100% gelatin) sample in regard to shear, as measured by the Warner Bratzler Shear. It was significantly ($p<0.05$) less tender and resembled the shear of commercially available marshmallows in South Africa. All samples had a light, greyish yellow color.

Keywords**:** mucilage; xanthan; agar-agar; gelatin; marshmallows; *Opuntia ficus-indica*

INTRODUCTION

Marshmallows are one of the earliest confections known to man (Olver, 2000) and were originally made from the root sap of the marshmallow (*Althaea officinalis)* plant. The first marshmallows were made by boiling pieces of the marshmallow root pulp with sugar until it thickened, whereafter the mixture was strained and left to cool. In 2000 B.C., Egyptians combined the marshmallow root with honey, and made a candy which was reserved for gods and royalty (Groves, 1995; Olver, 2000).

Modern marshmallow confections were first made in France around 1850 when marshmallows were cast and molded individually. When mass production became possible, by 1900, marshmallows were made by implementing the starch mogul system. By 1955, Alex Doumak of Doumak, Inc. patented a new manufacturing method, called the extrusion process, which changed the history of marshmallow production and is still used today (Groves, 1995).

Marshmallows are made by using sweeteners and emulsifying agents. Proportionally, there is more corn syrup than sugar, because it increases the ability of the sugar to dissolve and retards crystallization. Corn starch, modified food starch, water, hydrocolloids, gelatin, and/or whipped egg whites are used in various formulations, resulting in a specific texture. Hydrocolloids act as emulsifiers in marshmallows and provide the aeration that makes marshmallows puffy; however, they also act as gelling agents. Most marshmallows also contain natural and/or artificial flavoring (Groves, 1991; Olver, 2000).

Opuntia spp. is a genus of < 200 species of readily recognizable cacti, growing from small ground-hugging plants to massive shrubs. The fiber content has the capacity to absorb large amounts of water, forming viscous or gelatinous colloids and absorbing organic molecules (Saenz *et al.*, 2004; Anderson, 2008).

The Cactaceae family, of which *O. ficus-indica* is a part, is characterized by the production of slime, which mainly consists of mucilage. When cutting through a cladode, mucilage is the soluble fiber found in the thick slimy fluid, where they are stored in cells, commonly referred to as "nopal dribbles". Thus, these complex hydrophilic polysaccharide carbohydrates molecules, with a highly branched structure and containing varying proportions of L-arabinose, D- galactose, L-rhamnose, and D-xylose, as well as galacturonic acid, may not only bind water as they increase in volume, but also sugars and fats, which are caught within the matrix of mucilage and water (Goycoolea and Cardenas., 2003; Saenz *et al.*, 2004; Ramirez-Tobias *et al*., 2012).

Mucilage is, therefore, commonly referred to as a gum or hydrocolloid. Hydrocolloids are water-soluble dietary fiber that can be used as healthy additives for profitable food products. None of the mucilage found in cladodes is hydrolyzed or absorbed by the human digestive system, but they can make up alimentary fiber (Sepulveda *et al.*, 2007; Osuna-Martinez *et al*., 2014).

Mucilage has a strong emulsifying capability in that it reduces surface interfacial tensions, stabilizes oil-in-water emulsions and does not contribute to the viscosity of the system. Its stabilizing properties are similar to those of xanthan and guar gums. It can also form part of gelled dessert and powdered drinks, to be reconstituted with water or milk (Bensadon *et al*., 2010).

Another possible function for mucilage could be as a fat mimic, as it has the required tendency to increase the viscosity of the water phase. With increased concentration, it will exhibit gelling capabilities. Mucilage can, therefore, be used to emulsify systems, but as it is not a true emulsifier, processing costs would be reduced, while providing the final product with a natural and healthy ingredient as a replacement for fat (Bensadon *et al.,* 2010; Milani and Maleki, 2012).

Today, new facts at the molecular level of chemistry, as well as on functional properties, allow improved knowledge on the selection of a suitable hydrocolloid for a specific product. Cactus mucilage definitely has the advantages of being natural, healthy and cheap. However, despite having potential uses, it is not yet an industrial hydrocolloid (Saenz *et al.*, 2004).

The aim of this study was to replace gelatin in marshmallows with different concentrations of liquid mucilage, in combination with different concentrations of powdered hydrocolloids. Various physical parameters, such as consistency, percentage sag, shear, color and water activity (a_w), were determined.

MATERIALS AND METHODS

Extraction of liquid mucilage

Mucilage, from the *Opuntia* cultivar Algerian, was extracted according to the patented method of Du Toit and De Wit (2011). The cladodes (one-year-old with an average mass of ± 700 g, delivering a wet mucilage yield of ± 34% per cladode) were scrubbed and disinfected with chlorinated water to lengthen the shelf life period of the mucilage, and to eliminate impurities and thorns present. The green outside peel was removed with a vegetable peeler. The cladodes were then sliced into squares and cooked in a microwave oven (900 W) for 3 min at 100% power, until soft. The cooked, soft cladode pieces were pulverized in a food processor (300W) to break the skin, which aids in the extraction of the mucilage. The pulp was centrifuged in a Beckman centrifuge, model J2-21, (Beckman Coulter Inc. 2505 Kraemer Blvd. Brea, CA 92817 USA) for 15 min at 8000 rpm, to separate the liquid mucilage from the solids.

Preparation of marshmallows

The formulation for the marshmallows that were used as the control sample is given in Table 1. The gelatin was soaked in cold water. The sugar, salt, syrup and the remaining water were heated and stirred to dissolve the sugar. After the mixture reached a temperature of 118 °C, the gelatin was added. The stiffly beaten egg whites were added to the hot mixture, whilst continuously mixing it until thick and creamy. The mixture was then poured into a greased pan, cooled overnight and cut into 25-mm squares. The squares were rolled in a mixture of corn flour and icing sugar, and stored in an airtight container (Foods and Cookery, 1991).

Table 1. Formulation of control marshmallow (Foods and Cookery, 1991).

Ingredient	Percentage (%)
Gelatin	2.30
Water cold, to hydrate gelatin	11.52
Sugar	46.09
Water	28.81
Egg whites	9.22
Syrup	1.93
Salt	0.13

Replacement of gelatin with liquid mucilage and powdered hydrocolloids

Nine different formulations were prepared according to Table 2. The three hydrocolloids that were chosen to be used in combination with the fluid mucilage, to replace gelatin in the formulation, were gum guar, agar, and xanthan. Guar was chosen because of the synergistic effect that it displayed when used in combination with xanthan (Pangborn, *et al*. 1978). According to Bensadón *et al*. (2010), the stabilizing properties of mucilage are similar to those of xanthan and guar gums. Agar was also chosen, because it is commercially used in South Africa in chocolate candies to stabilize the marshmallow layer, such as *Cadburys Chocolate log* (Made in South Africa by Mondelez South Africa (PTY) LTD. 18 Harrowdene Office Park, Kelvin Drive, Woodmead Sandton, 2129, SA.).

The control sample contained 100% gelatin and one of the experimental samples 100% liquid mucilage. The lowest concentration at which the mucilage was used in the formulations was 75%. The lowest concentration at which the other hydrocolloids were added, was 12.5 %. The sample containing 100% liquid mucilage did not gel and could not be used for any of the physical texture analysis tests.

Physical texture analysis of marshmallow samples

Consistency

A glass plate, placed on paper with concentric circles drawn on it, at intervals of 0.5 cm was used. An open-ended tube, 2 cm x 2 cm in diameter, filled with 5 mL of the liquid sample, was placed in the center of the circles. After the tube was lifted, the liquid sample was allowed to flow for 2 min. The distance, traveled by the liquid, was then measured at each 90º section of the circle and the average calculated; the line spread value (LSV) was the mean of the four values obtained (Kim, 2007) and expressed in cm.

Texture

The penetrometer was used to determine the comparative tenderness and penetration properties of semi-solid substances (Mohos, 2010). The cone penetration test (ASTM D217) was used and the depth was measured in mm. The larger the reading, the more tender the product. The more tender the sample, the deeper the penetrometer will sink into the sample and, thus, the higher the penetration number will be. The flat attachment tests resistance against pressure; thus, the higher the value the more compression is achieved, indicating a more tender softer product. The results were expressed in mm.

Tenderness of gel

The percentage sag test was used to determine the gel tenderness and was expressed as percentage of the height, before unmolding. For this test, the depth of the marshmallow sample was measured in its container, by using a probe. After the sample was unmolded onto a flat surface (McWilliams, 1989), the depth was measured again. Percentage sag was then calculated as the change in height of the sample measured in the container or mold, compared to that of the freestanding gel placed on a flat surface. The greater the percentage sag value, the more tender the gel (McWilliams, 1989).

Table 2. Substitution hydrocolloids, combinations of hydrocolloids, substitution percentages and weights used in the preparation of marshmallows samples.

% substitution	Actual weight replacement (g)	Gelatin substitution	Formulation Code
Control	143.27g	gelatin	G
100%	143.27g	mucilage*	M
75% + 25%	107.45g+35.82g	mucilage* and xanthan	MX
75% + 25%	107.45g+35.82g	mucilage* and agar	MA
75% + 25%	107.45g+35.82g	mucilage* and gum guar	MG
80% + 20%	114.62g+28.65g	mucilage* and xanthan	8M2X
80% + 20%	114.62g+28.65g	mucilage* and agar	8M2A
80% + 20%	114.62g+28.65g	mucilage* and gum guar	8M2G
75%+12.5%+12.5%	107.45g+17.9g+17.9g	mucilage* and xanthan and agar	MXA
75%+12.5%+12.5%	107.45g+17.9g+17.9g	mucilage* and xanthan and gum guar	MXG
75%+12.5%+12.5%	107.45g+17.9g+17.9g	mucilage* and agar and gum guar	MAG

* Liquid mucilage, while other hydrocolloids were powdered.

Shear

Shear analysis was performed with the Instron Universal Testing Machine (UTM Model 3344). The samples from each of the treatments were cooled down to room temperature (19-25°C) for at least 24 hours, before shear force measurements were taken and for each sample six repetitions were done. Samples were prepared, with a core diameter of more or less 12.7 mm and a length of 10 mm, as advised by the prescribed method. The load cell size was 1 kilo Newton. A Warner Bratzler shear device, mounted on the Instron Universal Testing Machine, was used and the reported value in kg represented the average of the peak force measurements of each sample. Sample compression data was generated by the Instron Bluehill software (Instron, 2004).

Color analysis

Color measurements were performed on the marshmallow samples, 24 hours after preparation, using a Minolta CR 400 Chroma Meter, with an 8 mm measuring area at a 0º viewing angle. Calibration was done by using the white calibration plate and measurements were done in sextuplicate. The CIELAB color space (CIE, 1986) was used for measurements, with L* representing lightness. Chroma (C*), as well as Hue Angle (Hº), was calculated by using a* and b* values in the respective formulas: $C^* = \sqrt{(a*)^2 + (b*)^2}$ and $H^o = \tan^{-1}\left[\frac{b*}{a*}\right]$ (Ripoll *et al*. 2011; Tapp *et al*., 2011), where C* represents saturation and Hº the tone, which indicates color variation in the plane formed by the coordinates a* and b* (Chervin *et al*., 1996).

Water activity (a_w)

The water activity of the marshmallow samples in a_w containers (height of 5 mm and diameter of 39 mm) was measured with a Novasina Thermoconstanter TH 200 water activity meter, at room temperature (20 ºC). The means of the replications for the marshmallow samples were recorded (Mathenjwa *et al.,* 2012).

Comparison between best experimental formulation and commercial marshmallows

The best experimental formulation sample was then tested against four commercial marshmallow brands in South Africa: Woolworths white (WW); Woolworths traditional white (WTW); Beacon white (BW); and Manhattan white (MW). Only Woolworths white is made according to the traditional process, while the other three are extruded. The texture of the extruded marshmallows is tougher than marshmallows made by the traditional way. For this comparison, the line spread test was omitted, because the commercial samples could not flow upon melting. All the other textural tests were performed, as described above.

Statistical Analysis

All results were captured in multiple spreadsheets in Microsoft Excel 2007. A one-way analysis of variance (ANOVA) procedure (NCSS, 2007) was used to determine the effect of mucilage/ hydrocolloid formulations on the physical parameters of the marshmallow samples. Differences were considered statistically significant at $p<0.05$ level or lower. Treatment means were then compared by means of the Tukey-Kramer multiple comparison test at $\alpha = 0.05$.

RESULTS AND DISCUSSION

The replacement of gelatin with mucilage, in combination with guar, agar agar and xanthan, had a significant ($p<0.05$) effect on all the evaluated physical parameters of the marshmallows (Table 3).

Consistency

For the line spread test, the MX sample had the lowest value, signifying that the consistency of this formulation was the highest of the all the samples. This sample differed significantly ($p<0.05$) from all the other samples (Table 3). The consistency of the control sample (G = gelatin) did not differ from that of 8M2A and MAG; however, it was significantly ($p<0.05$) higher than MA, MG, 8M2X and 8M2G, and significantly ($p<0.05$) more fluid than MX, MXA, and MXG. Sample 8M2G had the highest value for the line spread test, indicating that it was the most fluid and was significantly ($p<0.05$) more fluid than all the other samples (Table 3).

Since the control sample contained 100% gelatin and resulted in a successful product, a line spread test value of around 19.72 mm could result in a successful product. The combinations 8M2A and MAG did not differ from the consistency of the control sample (Table 3). In both of these samples, agar was used in combination with mucilage, which could be responsible for the satisfying results. Agar is used in confectionaries, because of its solubility and the strength it confers upon the product (Nussinovitch, 1997). It was also found that, as soon as the percentage mucilage increased, so did consistency (Table 3).

Texture

The sample with the highest measurement for the flat attachment was the control sample (G). According to McWilliams (1989), the larger the reading, the tenderer is the product, as the attachment will sink deeper into a tender sample, resulting in a higher penetration value. The samples with the lowest reading were MA, 8M2X, and MAG, indicating that these samples were less tender (Table 3). Sample MX did not differ from MG and 8M2G, but was significantly ($p<0.05$) harder than G, 8M2A, MXA, and MXG. Although MXA was also significantly ($p<0.05$) harder than G, it was the mucilage combination closest to the tenderness achieved by the control sample, containing 100% gelatin. Agar is known as a ‘gelling type’ hydrocolloid, like gelatin (Saha and Bhattacharya, 2010), but it has stronger setting properties (five times greater), so agar form gels at lower concentrations (Condrasky, 2014). Xanthan, on the other hand, is used for thickening (Saha and Bhattacharya, 2010) and adds elasticity (Condrasky, 2014). The more crumbly texture caused by agar (Condrasky, 2014) may have been softened by the xanthan, resulting in a tenderer texture.

For the cone attachment, the lowest value was measured for the 8M2A sample, which did not differ from MA, MAG, and 8M2G; these samples were the tenderest and differed significantly ($p<0.05$) from all the other samples (Table 3). The MG sample was significantly ($p<0.05$) less tender than MXG, MXA and G, and significantly ($p<0.05$) tenderer than all the other samples (Table 3). Finally, MXA was significantly ($p<0.05$) less tender than the control sample G (197.00), which contained 100% gelatin.

Tenderness of gel

The sample that scored the lowest percentage sag was the control sample (G), indicating a strong gel texture, as the greater the percentage sag value, the more tender the gel is (McWilliams, 1989); this sample differed significantly ($p<0.05$) from all the other samples (Table 3). The 8M2G and MAG samples had the highest percentage sag, meaning that they were the tenderest of all the samples. The sample closest to the control sample (G), again, was MXA, which was also the best sample in the penetrometer tests. Although it did not differ from MX and 8M2X, it had the lowest numerical percentage sag.

Shear

For the physical attribute shear, the lowest value was scored by three samples, 8M2A, 8M2G, and MAG. These did not differ from the values recorded by the control, MA and MG. MXA, the toughest of all the samples, differed significantly ($p<0.05$) from all the other samples. This sample, which is a combination of 75 % mucilage + 12.5 % xanthan + 12.5 % agar, was the most promising hydrocolloid formulation thus far. For this attribute, it measured at the opposite end of the scale, compared to the control sample G. As mentioned earlier, xanthan does not form a gel when used alone. In combination with certain hydrocolloids, such as guar and locust bean gum (LBG), gels are formed, because of synergistic interactions. Xanthan shows quite spectacular synergistic interactions with other non-gelling polysaccharides of the galactomannan family (Saha and Bhattacharya, 2010), giving an increase in consistency (Casas and García-Ochoa, 1999) and gel formation (Rodriguez - Hernández and Tecante, 1999). It might be possible that there may also be an interaction between xanthan and mucilage, which requires further investigation.

The MA combination had a low score which did not differ from that of the control sample. This indicated that the shear increase was introduced by the 12.5% xanthan in the MXA combination. This is in agreement with the findings of Al-Assaf *et al*. (2006), who stated that xanthan shows a very high consistency at low shear rate and is thus preferred as a thickener or stabilizer for dispersed solids or emulsions.

Table 3. Physical and color properties of different formulations of hydrocolloids, in the making of marshmallows.

Property	G	MX	MA	MG	8M2X	8M2A	8M2G	MXA	MXG	MAG
Consistency (cm)	1.97 ± 0.02^{d}	1.42 ± 0.06^{a}	2.14 ± 0.10^{e}	2.32 ± 0.07^{f}	2.27 ± 0.12^{f}	2.05 ± 0.01^{de}	2.33 ± 0.07^{f}	1.83 ± 0.11^{c}	1.66 ± 0.15^{b}	1.98 ± 0.05^{d}
Texture (flat)* (mm)	0.23 ± 0.33^{f}	0.77 ± 0.22^{ab}	0.53 ± 0.20^{a}	0.90 ± 0.19^{bc}	0.53 ± 0.03^{a}	1.33 ± 0.15^{d}	0.90 ± 0.09^{bc}	1.90 ± 0.05^{e}	1.10 ± 0.21^{cd}	0.53 ± 0.07^{a}
Texture (cone)* (mm)	19.70 ± 0.83^{f}	14.87 ± 0.80^{b}	0.15 ± 0.08^{a}	16.03 ± 1.00^{c}	14.13 ± 0.17^{b}	0.14 ± 0.02^{a}	0.19 ± 0.06^{a}	18.47 ± 0.56^{e}	17.27 ± 0.28^{d}	0.17 ± 0.00^{a}
Tenderness of gel (%)	0.96 ± 1.11^{a}	9.18 ± 1.10^{bc}	19.84 ± 1.76^{d}	16.38 ± 3.18^{d}	8.85 ± 1.65^{bc}	9.74 ± 1.50^{c}	29.78 ± 3.63^{e}	5.41 ± 0.86^{b}	9.70 ± 3.18^{c}	27.90 ± 6.51^{e}
Shear(kg)	0.07 ± 0.03^{a}	0.45 ± 0.20^{bc}	0.06 ± 0.02^{a}	0.05 ± 0.03^{a}	0.33 ± 0.15^{b}	0.02 ± 0.01^{a}	0.02 ± 0.01^{a}	0.69 ± 0.10^{d}	0.56 ± 0.22^{cd}	0.02 ± 0.01^{a}
a*	-0.12 ± 0.09^{f}	-0.57 ± 0.25^{de}	-1.08 ± 0.18^{c}	-2.39 ± 0.07^{a}	-0.64 ± 0.13^{de}	-1.09 ± 0.24^{c}	-1.86 ± 0.08^{b}	-0.71 ± 0.05^{d}	-0.44 ± 0.11^{e}	-1.13 ± 0.13^{c}
b*	9.29 ± 0.87^{b}	8.16 ± 2.53^{ab}	16.67 ± 0.20^{c}	20.61 ± 0.72^{e}	7.89 ± 2.11^{ab}	16.26 ± 0.31^{c}	18.13 ± 0.42^{cd}	9.64 ± 0.35^{b}	6.34 ± 2.15^{a}	19.20 ± 0.63^{de}
L*	90.03 ± 1.88^{d}	90.63 ± 1.96^{d}	85.67 ± 1.80^{c}	82.52 ± 0.85^{b}	89.25 ± 1.77^{d}	86.40 ± 0.50^{c}	81.27 ± 1.11^{b}	91.33 ± 0.36^{d}	89.47 ± 1.79^{d}	77.59 ± 2.86^{a}
C*	9.29 ± 0.07^{b}	8.18 ± 2.54^{ab}	16.70 ± 0.18^{c}	20.74 ± 0.71^{e}	7.91 ± 2.12^{ab}	16.30 ± 0.30^{c}	18.22 ± 0.41^{cd}	9.67 ± 0.35^{b}	6.35 ± 2.15^{a}	19.23 ± 0.63^{de}
H°	90.27 ± 0.55^{a}	93.82 ± 0.62^{bc}	93.71 ± 0.66^{bc}	96.62 ± 0.42^{e}	94.69 ± 0.34^{d}	93.84 ± 0.89^{bc}	95.87 ± 0.32^{e}	94.24 ± 0.44^{cd}	94.09 ± 0.42^{bcd}	93.37 ± 0.34^{b}
a_w	0.78 ± 0.01^{de}	0.78 ± 0.01^{ef}	0.76 ± 0.01^{bc}	0.77 ± 0.01^{cd}	0.76 ± 0.01^{ab}	0.80 ± 0.01^{g}	0.78 ± 0.01^{de}	0.75 ± 0.01^{a}	0.79 ± 0.01^{fg}	0.78 ± 0.01^{de}

Means with different superscripts in the same row differ significantly ($p < 0.05$).

Texture (flat) =penetrometer flat; Texture (cone) =penetrometer cone; a=redness; b*= yellowness; L*=lightness; C*= Chroma (saturation index); H°=Hue angle; a_w=water activity.

Samples:

G Gelatine
MX 75% Mucilage and 25 % Xanthan
MA 75% Mucilage and 25% Agar
MG 75% Mucilage and 25%Guar Gum
8M2X 80% Mucilage and 20% Xanthan
8M2G 80% Mucilage and 20% Guar Gum
8M2A 80% Mucilage and 20% Agar
MXA 75% Mucilage and 12.5% Xanthan and 12.5% Agar
MXG 75% Mucilage and 12.5% Xanthan and 12.5% Guar Gum
MAG 75% Mucilage and 12.5% Agar and 12.5% Guar Gum

Color analysis

Values for a* and b* measurements will not be discussed and is only included in Table 3 for clarification of the C* and Hº calculations. L* values distinguish between bright and dull colors (Konica, 1998). All the samples tested showed high values for L*, with MXA having the highest value; this sample did not differ from the control sample and appeared to be the best formulation of hydrocolloids to replace gelatin (Table 3). The MAG sample had the lowest score for lightness, indicating that it was significantly ($p<0.05$) darker when compared with the other samples. All the samples containing xanthan had the highest L* values, suggesting that xanthan had a lightening effect on the final product. Except for sample MXG, which contained xanthan, guar had a darkening effect on all the marshmallow samples (Table 3). In a study by Brotnowska and Makiewics (2006) on the influence of xanthan and guar on the color of mayonnaise, sensory profiling indicated that both hydrocolloids intensified the creamy color of low-fat mayonnaise. With an increase in the hydrocolloid, the characteristic yellow color disappeared and became a light creamy color, which increased with increasing concentration of the hydrocolloid. Considering that the color of a food is the first contact point of the consumer (Alvarenga *et al*., 2012) and that the consumer may want a bright white marshmallow, the L* value may play an important role.

Chroma is an indication of the saturation of colors, indicating whether they are vivid/strong to dull, to weak/greyish (McGuire, 1992; Konica, 1998). All the samples had values ranging from 6.35 to 20.74 (Table 3), putting them horizontally closer to the center or greyish/ weak area in the plane formed by the coordinates a* and b* (Chervin *et al*., 1996; Konica, 1998). The sample with the lowest C* value was MXG, which was the dullest or most greyish sample; it differed significantly ($p<0.05$) from G, MA, MG, 8M2A, 8M2G, MXA and MAG, but not from MX and 8M2X. This could be attributed to xanthan that was used in the formulations. With the L* values, the xanthan-containing samples were also the lightest. Sample 8M2X differed significantly ($p<0.05$) from MA, MG, 8M2A, 8M2G and MAG, but not from G, MX, MXA, and MXG. Sample MA differed significantly ($p<0.05$) from G, MX, MG, 8M2X, MXA, MXG and MAG, but not from 8M2A and 8M2G. Sample 8M2G differed significantly ($p<0.05$) from all the samples, but not from 8M2A and MAG, while MG had the highest value for C* and differed significantly from all the samples, except from MAG. Samples containing guar had thus a stronger saturation of color than the other formulations.

Hue is the term used for the classifications of colors, for example red, yellow, blue, etc. For this attribute, all the samples had values ranging between 90º and < 96º (Table 3), and when the hue angle in this region, the color is yellow (Konica, 1998; XRite, 2007; Zhang *et al*., 2007). The highest values were found for both samples containing guar, namely MG (75% mucilage and 25% guar gum) and 8M2G (80% mucilage and 20% guar gum), while the lowest Hº value was for the sample containing gelatin (Table 3). For the samples where xanthan was added to the formulations, the hue angle increased significantly ($p<0.05$) for samples 8M2X (80% mucilage and 20% xanthan), MXA (75% mucilage and 12.5% xanthan and 12.5% agar) and MXG (75% mucilage and 12.5% xanthan and 12.5% agar).

Although the various marshmallow formulations differed significantly ($p<0.05$) amongst each other, it could be concluded that all samples had a light greyish yellow color.

Water activity

All the samples fell within the range of intermediate moisture foods (IMF). These are foods with a_w of 0.65 - 0.85, which is largely responsible for its protection from microbial spoilage (Garbutt, 1997). The sample with the lowest a_w value was MXA; this sample differed significantly ($p<0.05$) from all the other samples (Table 3). The sample with the highest a_w of 0.80, namely 8M2A, differed significantly ($p<0.05$) from all the samples, but still fell within the range of IMF. The MXA combination had a significantly ($p<0.05$) lower a_w than the control (G) sample, rendering it even more acceptable as a substitute for gelatin in the marshmallow formulation. This water dispersing property is common to all hydrocolloids; however, the extent of thickening varies with the type and nature of the hydrocolloid (Saha and Bhattacharya, 2010). Nammakuna *et al*. (2009) found that addition of hydrocolloids to rice crackers increased its moisture content, which correlated to significantly higher a_w in treated samples, as compared to the controls.

It can be concluded that the best textural attributes were associated with sample MXA, consisting of 75% mucilage, 12.5% xanthan, and 12.5% agar. This sample showed good results in all the above mentioned physical evaluations and it was the closest sample to the control. It is then clear that in combination with xanthan and agar, mucilage can be used as a substitute for gelatin in the production of marshmallows.

Comparison between best experimental formulation and commercial marshmallows

Texture

The sample with the highest measurement for the penetrometer with the flat attachment was the MXA. As noted earlier, the larger the reading, the tenderer is the product, as the attachment will sink deeper into a soft sample, resulting in a higher penetration value. The sample with the lowest reading was BW, indicating that this sample was not tender (Table 4). The two Woolworths samples did not differ, although the one was made in the traditional way and the other one was extruded. All the other samples differed significantly ($p<0.05$). The gelling agent for all four commercial samples was gelatin. For this test, the MXA sample resembled the tenderness of the MW sample the most, although they differed significantly ($p<0.05$).

When the cone attachment was used on the penetrometer, the samples were divided into three groups: WW and BW, with the lowest scores, which differed significantly ($p<0.05$) from the next group, MXA and MW, which in turn differed significantly ($p<0.05$) from WTW, which had the highest score (Table 4), indicating that this sample was the tenderest. For these two measurements, the BW samples scored the lowest values in each instance, confirming that it was the least tender sample. It should be noted that again the measurements for MXA and MW did not differ for this attachment.

Tenderness of the gel

Both MXA and MW had the lowest values for percentage sag and both samples differed significantly ($p<0.05$) from BW and WTW, but not from WW; this sample differed significantly ($p<0.05$) from WTW (Table 4). Samples MXA and MW, thus, had tough gel textures. The greater the percentage sag value, the more tender is the gel (McWilliams, 1989); therefore, the WTW sample was the tenderest gel.

Shear

For the physical attribute shear, the lowest value was scored by MXA, indicating least shear. Interestingly, all the commercial samples differed significantly ($p < 0.05$) from this sample, as well as from one another. From these samples, the sample with the least shear was WTW. Sample MW was the toughest sample, with BW second toughest, followed by WW. This confirms the belief that traditionally made marshmallows are not as tough as the extruded ones. For extrusion to be successful, the mixture contains modified corn starch, in addition to gelatin, the temperature of extrusion has to be just right so that the mixture will flow under pressure, but hold its shape after leaving the pipe (Mulvaney, 2015).

Color analysis

Values for a* and b* measurements will not be discussed and is only included in Table 4 for clarification of the C* and Hº calculations. All the samples, again, showed high values for L* (lightness). The sample with the lowest L* value was MXA and it differed significantly ($p<0.05$) from all the other samples, the reason for this being the greenish color of the fluid mucilage, which lowered the L* value. The BW sample differed significantly ($p<0.05$) from MXA, WW, and MW, but not from WTW; this sample differed significantly ($p<0.05$) from MXA and WW. The commercial sample with the highest value for lightness was WW (Table 4). In a study by Periche *et al.* (2015), it was found that the factor that had the greatest influence on lightness, was the percentage of gelatin; the higher the level of gelatin, the higher the lightness. Although there were significant differences ($p<0.05$) between the samples for this color attribute, all samples were light, which is of importance for the consumer looking for a white marshmallow.

The C* values were significantly ($p<0.05$) different amongst the samples, except for samples BW and MW, which did not differ from each other. The coordinates, again, placed the samples in the greyish / weak yellow spectrum (McGuire, 1992; Konica, 1998; Sepulveda, *et al*., 2007). Sample MXA had significantly ($p<0.05$) the highest value for C*, while sample WW had significantly ($p<0.05$) the lowest value. Despite these significant ($p<0.05$) differences, the experimental sample and the commercial samples had values in the same range, and the use of mucilage, along with agar and xanthan, would have a favorable impact on the acceptability of the consumer.

For Hº, all the values were between 90.71º and 99.22º (Table 4), placing the color again in the yellow region (McGuire, 1992; Konica, 1998; Zhang *et al*., 2007). The highest value was found for the WW and WTW samples and the lowest Hº value was calculated for the BW sample. The mucilage sample, MXA, differed significantly ($p<0.05$) from the three previously mentioned samples, but not from the commercial brand MW.

Although the samples differed significantly ($p<0.05$) amongst each other for the color analysis, they all had a light greyish yellow color.

Table 4. Physical and color properties for MXA and four commercial marshmallows brands.

Property	MXA	WW	WTW	BW	MW
Texture (flat)* (mm)	1.90 ±0.06^{d}	0.78 ± 0.15^{b}	1.00 ± 0.14^{b}	0.52 ± 0.13^{a}	1.43 ± 0.16^{c}
Texture (cone)* (mm)	18.47 ± 0.76^{b}	17.38 ± 0.73^{a}	20.10 ± 0.26^{c}	17.13 ± 0.51^{a}	18.92 ± 0.75^{b}
Tenderness of gel (%)	5.41 ± 1.15^{a}	9.18 ± 1.48^{ab}	16.38 ± 4.27^{c}	9.74 ± 2.01^{b}	5.41 ± 1.15^{a}
Shear (kg)	0.69 ± 0.14^{a}	25.36 ± 6.55^{c}	16.47 ± 4.01^{b}	41.00 ± 5.54^{d}	67.42 ± 6.11^{e}
a*	-0.71 ± 0.07^{b}	-0.98 ± 0.10^{a}	-1.13 ± 0.12^{a}	-0.07 ± 0.12^{c}	-0.77 ± 0.09^{b}
b*	9.64 ± 0.47^{d}	5.81 ± 0.58^{a}	6.97 ± 0.85^{b}	8.31 ± 0.94^{c}	8.25 ± 0.15^{c}
L *	91.33 ± 0.48^{a}	96.39 ± 0.21^{d}	94.40 ± 1.02^{bc}	93.61 ± 0.36^{b}	94.90 ± 0.54^{c}
C*	9.67 ± 0.47^{d}	5.89 ± 0.59^{a}	7.06 ± 0.85^{b}	8.31 ± 0.94^{c}	8.29 ± 0.15^{c}
H(°)	94.25 ± 0.59^{b}	99.61 ± 0.72^{c}	99.22 ± 0.65^{c}	90.71 ± 0.89^{a}	95.30 ± 0.61^{b}
a_w	0.75 ± 0.01^{e}	0.52 ± 0.01^{a}	0.65 ± 0.01^{d}	0.63 ± 0.00^{c}	0.60 ± 0.01^{b}

Means with different superscripts in the same row differ significantly ($p < 0.05$). *Texture (flat) =penetrometer flat; *Texture (cone) =penetrometer cone; a*=redness; b*= yellowness; L*=lightness; C*= Chroma (saturation index); H°=Hue angle; a_w=water activity.
Samples:

MXA=75% Mucilage and 12.5% Xanthan and 12.5% Agar
WW=Woolworths white

WTW=Woolworths traditional white

BW=Beacon white

MW=Manhattan white

Water activity

Again, all the samples fell within the range of intermediate moisture foods (IMF) and even lower. The sample with the highest a_w value was MXA, which differed significantly ($p<0.05$) from all the other samples (Table 4). The liquid mucilage has a high a_w of 0.96 (De Wit, 2015), which contributed to the high a_w of the MXA sample. It is interesting to note that all the commercial samples had values that differed significantly ($p<0.05$). Furthermore, the traditional commercial sample, WTW, was the only commercial sample with a_w of 0.65, while the other commercial samples had lower values. Sample BW, an extruded commercial brand, had a_w of 0.63, which is lower than the recommended range of 0.65 and 0.85. This low a_w ensures the microbial stability of the product and lengthens the shelf life. Sample MW had an even lower a_w of 0.60, qualifying it to be a low moisture food. The lowest a_w was achieved by WW, with a value of 0.52, also qualifying as a low moisture food.

CONCLUSIONS

Marshmallows were selected as the potential vehicle for the incorporation of cactus pear mucilage in different formulations, using a variety of hydrocolloids. The MXA sample, containing 75% mucilage, 12.5% xanthan, and 12.5% agar, obtained the best results for all the physical parameters tested and could be a potential formula requiring further research. Marshmallows containing mucilage in combination with agar and xanthan might also be a good alternative for similar products, where only one hydrocolloid, for example, agar is used as a gelling agent, resulting in a tough texture. Furthermore, gelatin-less marshmallows might also open up a market which was restricted by the use of gelatin, namely for Halaal and Kosher consumers.

Since the consumer has the final decision in whether a newly developed product is acceptable, it is necessary to submit it to a sensory panel of regular marshmallow consumers to determine its overall acceptability, taste, aftertaste, as well as the two most important attributes in the case of this aerated gelled product, namely appearance and texture.

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