FUNCTIONALITY OF LOW DIGESTIBILITY EMULSIONS IN COCOA CREAMS. STRUCTURAL CHANGES DURING *IN VITRO* DIGESTION AND SENSORY PERCEPTION

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ABSTRACT

The objective of this work was to evaluate the application of low digestibility oil/water emulsions as fat source in a cocoa cream. Emulsions were composed by water, sunflower oil and cellulose ethers or xanthan gum. Back extrusion assays were measured before and after *in vitro* digestion and free fatty acids release were measured to evaluate the fat digestibility. Finally consumer acceptability was carried out to determine the degree of liking of each system. The results revealed that all the emulsions confer a suitable consistency to the creams and the structure provided by the hydrocolloids was resistant to digestion, reducing the fat digestibility. However, after gastric digestion only cream with xanthan gum showed a significant increase in consistency what it could be related with an increase in satiety. Regarding the sensory characteristics, the cream elaborated with xanthan gum was rated close to the control cream that received the highest scores.

Keywords: filling cream, hydrocolloids, free fatty acids, texture, acceptability.

1. Introduction

In the last decades, the increasing of a large number of diseases directly linked to fast food has driven the industry to focus on the design and formulation of food products with reduced fat and/or calories content. The contribution of fat in the flavour, texture, appearance and mouthfeel of foodstuffs has been confirmed by several studies (Drewnowski, 1992, 1987; Sandrou & Arvanitoyannis, 2000). Hence, removing or reducing fat adversely affects some of the characteristics reducing the quality of the final product. The main challenge is the manufacture of products with high lipid content, such as pastry and confectionery products. This type of foodstuffs contains a high percentage of saturated fats and/or *trans* fats, which gives them unique textural properties. Lipid content in cocoa creams can be more than 60% and provide a significant effect in organoleptic and physicochemical properties. In order to reduce calories content, fat could be reduced/replaced by a system that can replicate the texture, flavour and palatability of the full-fat counterpart. There are several systems that can be used as fat replacers, including protein-based fat mimetics, carbohydrate-based fat mimetics and fat based replacers (Lucca & Tepper, 1994; Sandrou & Arvanitoyannis, 2000). Most of the low-fat products reformulated in recent years contain carbohydrate-based fat mimetics such as inulin (Tárrega & Costell, 2006; Krystyjan, Gumul, Ziobro, & Sikora, 2015), starch (Laguna, Varela, Salvador, Sanz, & Fiszman, 2012), cellulose (Nsor-Atindana, Chen, Goff, Zhong, Sharif, & Li, 2017) and gums (Ranalli, Andrés, & Califano, 2017; Rather, Masoodi, Akhter, Gani, Wani, & Malik, 2015) that are widely used as thickeners, stabilizers and emulsifiers to compensate the loss of desirable textural attributes when fat is reduced or removed (Mudgil & Barak, 2013).

The incorporation of a polysaccharide (e.g cellulose ether) in the continuous phase of oil/water emulsion allows using vegetable oil in reformulated products. The semi-solid consistency was suitable for mimicking the textural and rheological properties of fat, conferring good sensory acceptability (Martínez-Cervera, Salvador & Sanz, 2015; Tarancón, Fiszman, Salvador, & Tárrega, 2013). An important attribute of emulsion-based food products is the behaviour within the mouth after ingestion that will determine the perceived mouthfeel (McClements, 2015). People like the taste of fat-containing foods. More viscous stimuli are generally perceived as rich in fat content. So, this feeling can be created through the use of stabilizers or thickeners that enhance the perceived creaminess of the reformulated product (Drewnoski, 1990). Nevertheless, depending on fat content and the type of thickener used, the aroma release and taste perceived may change (Wendin et al., 1997). Some studies showed that the use of thickeners results in an increase in texture and a decrease in aroma release and taste, but depending on the type and concentration of hydrocolloid (Arancibia, Castro, Jublot, Costell & Bayarri, 2015; Hollowood, Linforth, &Taylor, 2002). Moreover, some studies have shown that the presence of cellulose ethers or xantan gum in the continuous phase of oil/water emulsion makes more difficult for the digestive fluids to come into contact with the emulsified fat, reducing lipolysis (Espert at al., 2017; Espert, Salvador, & Sanz, 2018).

The objective of this work was to study the effect of low-digestible vegetable oil/water emulsion as a fat source on the structural and sensory properties of cocoa cream, considering a starch base cocoa cream with the same fat content as a control. The emulsions were prepared using cellulose ethers with different chemical substitution (methyl and hydroxypropyl methylcelluloses and xanthan gum as structuring agents. Lipid digestibility was also determined after *in vitro* digestion to evaluate the relationship between structural changes and fat digestibility in this new matrix.

2. Materials and Methods

2.1. Emulsion preparation

Different o/w emulsions were prepared using different hydrocolloids as stabilizers: xanthan gum (XG) (Cargill, France) and three types of cellulose ethers (METHOCEL™ A4M, METHOCEL™ MX and METHOCEL™ K4M) (The Dow Chemical Company, Midland,Michigan, USA). These celluloses present different chemical substitution: A4M and MX are methylcelluloses (30.0% methoxyl and >30.0% methoxyl respectively) and K4M is a hydroxylpropyl methylcellulose (22.5% methoxyl, 7.7% hydroxypropyl). A4M and K4M have approximately the same molecular weight (MW) and a viscosity of 4000 mPa s (measured at 20 °C following ASTM D1347 and ASTM D2363 reference methods (The Dow Chemical Company)). MX has a higher MW and a viscosity of 50,000 mPa s at 20 °C (measured following the same methods).

Emulsions were prepared using the following proportions: 47% (w/w) sunflower oil (Koipe Sol, Deoleo S.A., Spain) 2% (w/w) hydrocolloid and 51% (w/w) water. The total final mass was 200 g.

2.1.1 Cellulose ether based emulsion

At first, cellulose ether was dispersed in sunflower oil using a Heidolph stirrer (Heidolph Instruments GmbH & Co. KG) at 280 min^{-1} for five minutes. Then the mixture was hydrated by gradually adding of water at 1°C while continuous stirring. A water temperature of 1 °C was selected according to the specific hydration requirement of MX cellulose (the highest methoxyl content) and then it was also used for the other cellulose types. Finally the emulsion was homogenized using an Ultra-turrax T18 homogenizer (IKA, Germany) at 6500 rpm for 15 seconds and at 17500 rpm for 60 seconds.

2.1.2 Xanthan gum based emulsion

The XG was dispersed in the water at room temperature (22ºC) water using a Heidolph stirrer at 300-500 rpm for 10 minutes. Then, sunflower oil was gradually added increasing the speed up to 1800 rpm. Stirring continued using a homegenizer (Ultra-turrax) at 6500 rpm for 60 seconds, subsequently at 13500 rpm for 60 seconds and at last 17500 rpm for 60 seconds.

2.2. Creams preparation

Emulsion based cocoa creams were composed of water (30.25%), sugar (Disem, Spain) (10%), skimmed milk powder (Central Lechera Asturiana, Spain) (5%), cocoa powder (Chocolates Valor S.A., Alicante, Spain) (2.5%), starch (CTex 06205, Cargill BV, Netherlands) (2.25%) and emulsion (50%). A food processor (TM31 Thermomix, Vorwrek, Wuppertal, Germany) was used to mix the ingredients. At first, starch, sugar, milk powder, cocoa powder and mineral water were mixed at 90°C for 6 minutes at speed 2 in order to enable starch gelatinization. After that, mixture was allowed to cool at room temperature. Then, the hydrocolloid-based emulsion was added by mixing in the processor for 6 minutes at speed 2 without temperature selection to obtain the filling cream.

Control cream was formulated with the same ingredients, but instead of emulsion sunflower oil (23.5%) was added and the amount of water and starch were increased to 55% and 4% respectively. Control cream was prepared in the same way, but after cooling of the first step, the sunflower oil was added gradually to the mixture at room temperature.

All the creams contained the same proportion of fat (23.5%). They were stored at 5°C for 24 hours before the measurements.

2.3. In vitro digestion

2.3.1. Composition of digestive fluids

Simulated Saliva Fluid (SSF) was prepared according to the method described by Mishellany-Dutour et al. (2011), with some modifications. SSF was composed of 5.2g of $NaHCO_3$, 1.37g $K_2HO_4P \cdot 3\ H_2O$, 0.88g NaCl, 0.48g KCl and 0.44g $CaCl_2 \cdot 2H_2O$, dissolved in 1L of bi-distilled water. 0.70g of α-amylase from porcine pancreas (A3176-1MU, Sigma-Aldrich) and 2.16g of mucin from porcine stomach (M2378, Sigma-Aldrich were added to this solution.

Simulated Gastric Fluid (SGF) was prepared according to a previous study (Sanz, Handschin, Nuessli & Conde Petit, 2007) with some modifications. 3.10g NaCl, 0.11g $CaCl_2$, 1.10g KCl and 5.68ml Na_2CO_3 (1M) were dissolved in 1L of bi-distilled water. The solution was adjusted to pH 2. 0.15g of pepsin from porcine gastric mucosa (P7000, Sigma-Aldrich) was dissolved in 1L of SGF.

Simulated Intestinal Fluid (SIF) was composed of an electrolyte solution and bile and pancreatin solutions. The electrolyte solution was prepared by dissolving 1.25g NaCl, 0.15g KCl and 0.055g $CaCl_2$ in 1L of distilled water. Phosphate buffer solution was prepared (103.5mg $NaH_2PO_4 \cdot 2H_2O$ and 44.5mg $Na_2HPO_4 \cdot 2H_2O$ in 100ml of distilled water) (pH 7) to prepare bile (B8631, Sigma-Aldrich) and pancreatin (P1750 (lipase activity 8 USP units/mg), Sigma-Aldrich) freshly suspensions (Sanz et al., 2007).

2.3.2. In vitro digestion model

To simulate different digestion phases, an *in vitro* digestion model to simulated oral, gastric and small intestine digestion previously described was used (Borreani et al., 2016; Espert et al., 2017).

Oral phase: 50g of sample were mixed for 5 s with 1 ml of Simulated Saliva Fluid (SSF) (0.7 mg/mL α-amylase) in a shaking water bath (Raypa®, Barcelona, Spain) (60 rpm) at 37°C. The ratio saliva/sample was selected considering the data provided by Humphrey & Williamson (2001).

Gastric phase: the "bolus" sample from the oral phase was mixed with 16 mL of Simulated Gastric Fluid (SGF) (0.15 mg/mL) to obtain a final enzyme-sample ratio of 1:250 (v/v). The pH of the mixture was adjusted to 2.0 using 6M HCl (Scharlab S.L., Spain) and incubated for 1 h under continuous agitation (60 rpm) at 37ºC.

Small intestine phase: After gastric step, 10.6 mL of bile extract (46.87mg/mL) solution and 4 mL of electrolyte solution was added to the sample, and the pH was adjusted to 7.0 using NaOH (0.1N). Then, 5.34 mL of pancreatin solution was added to the mix (1:14 (v/v) ratio pancreatin/oil). The resulting mixture was incubated for two hours under continuous agitation at 37ºC.

In order to compare the effects caused by the volume of dilution with the effects caused by the presence of enzymes and pH changes, oral and gastric water dilution incubation was also carried out in which only distilled water was added. The incubation process (time, temperature and shaking conditions) and the dilution factor were the same as that in the samples with enzymes.

2.4. Fat digestibility

To determinate the digestibility of fat, the amount of Free Fatty Acids (FFA) released at the end of *in vitro* digestion were calculated. A pH-stat automatic titration (Mettler Toledo, Spain) was used to monitor automatically the pH at intestinal pre-set value (pH 7.0) by titration of NaOH 0.1N solution (Panreac Química S.L.U., Spain). The volume of NaOH added to neutralize the samples was recorded. A standard curve was prepared using oleic acid (0, 50, 100, 150, 200 and 250 mM) and was used to calculate free fatty acid concentration of the samples ("g oleic acid/g fat").

2.5. Textural properties

TA-XT plus Texture Analyzer equipped with the Texture Exponent software (Stable Microsystems, Godalming, UK) was used to determinate the extrusion properties of the samples. A back-extrusion test was carried out, using a bucket of 50 mm diameter and 75 mm height and a compression probe of 49 mm diameter. The distance force was 15mm, the compression rate 1 mm s-1, and the trigger force 10g. From the force time profiles obtained the area under the curve (AUC; N*s) as a measure of consistency were recorded. Measurements were performed in triplicate.

2.6. Sensory analysis

The sensory analysis was carried out in a sensory room equipped with individual booths designed in accordance with ISO 8589:2007 (ISO, 2007), under artificial daylight and controlled temperature (22ºC).

2.6.1. Free Choice Profile

A total of 20 untrained consumers (60% women, 40% men), with ages ranging from 25 to 50 years old, took part in a Free Choice Profile (FCP) analysis. In the first session, the terms used by each consumer describing the differences among creams were generated by Repertory Grid Method (RGM). The samples were presented in triads and each consumer described the similarities and differences among samples within each triad in their own terms. This method was repeated until all samples were tested. Consumers evaluated the appearance, taste, aroma and texture of the different creams. Each consumer evaluated his own list of terms by rating the intensity for each sample using a 10 cm unstructured line scale with the anchors “Not perceived” and “Intense”. The samples were labelled with random three-digit codes and served at room temperature. Water was provided to clean the palate between samples.

2.6.2. Liking test and CATA questionnaire

A sensory analysis of cocoa creams was carried out by 82 untrained consumers (69% women, 31% men) recruited among the students and employees of the Institute for Agrochemistry and Food Technology (IATA-CSIC). They were asked to taste the five samples of creams (control, A4M, F4M, MX and XG) and rate their overall acceptability and liking of their appearance, colour, taste and texture on a 9-point hedonic scale from 1 = “dislike extremely” to 9 = “like extremely”. After that, the consumers were asked to answer a Check-All-That Apply (CATA) questionnaire. The terms included were previously generated in a session with 20 consumers by using the Repertory Grid method (Table 3). They were first given the most different samples and then they were asked to choose and write down the most appropriate attributes with which to describe the characteristics of the samples. At the end of the session, a consensus on the list of sensory attributes was reached (Stone & Sidel, 2004). The CATA questionnaire included seventeen sensory terms. Each consumer was asked to check the terms that he/she considered appropriate for describing the cream sample. The five cocoa creams samples were served at 20ºC identified with random three-digit codes and were presented monadically following a Williams design. Data acquisition and analysis was performed by Compusense Cloud version 8.8.6642.32014 (Ontario, Canada).

2.7. Statistical analysis

One-way analysis of variance (ANOVA) was applied to study the effects of digestion on the different instrumental and sensorial parameters studied. The least significant differences were calculated by the Tukey test and the significance at $p < 0.05$ was determined.

A Generalized Procrustes Analysis (GPA) was applied to the Free Choice Profile data.

The non-parametric Cochran's test analysis of variance was performed for each descriptor to evaluate whether the CATA question was able to detect differences in the consumer perception of the cocoa creams. A descriptor was no longer considered when Cochran's test found that the differences between samples were not significant. The variability in the frequencies of mention of significant attributes was analysed by using a Correspondence Analysis (CA) and a Multiple Factor Analysis (MFA) was performed on the frequency of mention of the CATA question to assess the relationship between CATA question responses and acceptability scores. Every calculation was carried out using XLSTAT statistical software (2010.5.02 (Addinsoft, Barcelona, Spain)).

3. Results and Discussion

3.1. Textural properties

The texture of emulsion-based products is one of the most important factors that influence their overall sensory acceptance. The texture profiles of creams before and after *in vitro* digestion, as measured by extrusion tests, are shown in Figure 1. Fresh samples have the highest consistency in all creams, as its structure has not been altered by the effect of any digestion step. The use of polysaccharides contributes viscosity to the system, depending on the chemical composition of them. The area under the curve (AUC) values of fresh cocoa creams showed significant differences ($p<0.0001$) depending on the composition of the hydrocolloid used. The creams with A4M showed the highest value of AUC (42.78a), followed by MX methylcellulose and xanthan gum (24.73b and 23.15b respectively) Hydroxypropyl methylcellulose showed lower values (19.54bc), although the control cream presented the lowest AUC value (15.20c). After oral phase, a significant decrease in consistency was observed as compared to the consistency of fresh samples (Table 1). This fact is due to the presence of α-amylase enzyme in SSF that promotes the enzymatic degradation of the starch, causing a loss of consistency (de Wijk, Prinz, & Janssen, 2006; Sanz, Handschin, Nuessli, & Conde-Petit, 2007). This decrease in consistency is more evident in control cream, suggesting that the hydrocolloids provide consistency at the system and, in addition, it is known that the presence of hydrocolloid has a suppressive effect on starch digestibility (Sasaki & Kohyama, 2012). Samples without saliva enzymes (SSF) (saliva dilution samples) exhibit a higher consistency than the corresponding ones with SSF. They showed consistency values close to the fresh samples, showing no significant differences.

After *in vitro* stomach incubation, the extrusion profile of control cream (Figure 1A), MX cream (Figure 1E), A4M cream (Figure 1D), and K4M cream (Figure 1C) did not show significant differences with respect to water stomach dilution (without pepsin and initial pH). These results indicate that the change in consistency in this phase should be attributed to the dilution effect more than to the stomach conditions. However, contrary to cellulose ethers based creams, gastric digestion of XG cream showed a significant increase in AUC compared to its corresponding water dilution (Figure 1B, Table 1). This increase could be related to the behavior of the xanthan gum matrix in the acid pH of the stomach, where its viscous consistency is maintained. Moreover, the XG network weakens and there is more contact between the fat globules, which produces fat coalescence and therefore an increase in the consistency of the system. This behaviour has been also found in xanthan gum emulsions (Espert, Salvador, & Sanz, 2018). Several studies confirm that viscous fibres have been associated with a decrease stomach emptying and slower transit time through the small intestine, and have also been shown to influence blood glucose and cholesterol levels (Dickeman & Fahey, 2006; Mälkki, 2001). Insoluble fibres, such as cellulose, are mostly associated with large bowel function, although both types of fibre enhance postprandial sensations of satiety and to decrease hunger feelings (Juvonen et al., 2009; Howarth, Saltzman, & Roberts, 2001; Gustafsson, Asp, Hagander & Nyman, 1995). It is important to note that control cream showed the least resistance to back-extrusion test after *in vitro* digestion. This could be related to the fact that in control cream liquid fat is not emulsified and there is no hydrocolloid network, which provides consistency and cohesiveness to the system. Therefore, the use of xanthan gum emulsion as a fat replacer in cocoa cream make this product interesting in the design of satiating foodstuffs due to its increase in consistency in stomach phase.

3.2. Fat digestibility

Free Fatty Acids (FFA) are the product of fat digestion, so they are an indicator of the amount of fat which has been digested. The extent of lipid digestion varies depending on the enzymatic activity and a great number of physicochemical factors (Golding, Wooster, Day, Xu, Lundin, Keogh, & Clifton, 2011; Li, Hu & McClements, 2011; McClements, Decker & Park, 2007). Significant differences in free fatty acid generation were found between cocoa control cream (without hydrocolloid emulsion) and cocoa creams based on hydrocolloids emulsion (Table 2). It can be shown that creams based on hydrocolloid emulsions required less NaOH volume to neutralize any FFA produced by digestion, which indicates that these samples had a lower fat digestibility, so less oleic acid concentration was generated. Besides, and increase in the size of the fat globules and in droplet coalescence was observed in all hydrocolloid creams (data not shown). The sample with the lowest fat digestibility was MX cocoa cream, although no significant differences were found among the creams prepared with the different hydrocolloid emulsions. Schneeman, & Gallaher (2001) found that the hydrolysis of triglycerides in the small intestine is related to the available surface area, and an increase in droplet size is associated with a reduced surface area and a reduction in the rate of lipid hydrolysis. On the other hand, it is already known that the presence of fibres potentially influence on lipid digestion, making the access of bile salts and digestive enzymes to the oil phase difficult. Similar results using the same shear speed (60 rpm) were obtained by Hur, Lim, Park, & Joo (2009) and Mugdil & Barak (2013). They found that the molecular and physicochemical differences of the different polysaccharides can be expected to cause significant alterations in their effectiveness at reducing lipid digestion by interfering with the various physiological processes. Pasquier et al. (1996) showed that some viscous fibres reduce the lipid emulsification, lowering of the extent of fat lipolysis. Espinal-Ruiz et al. (2014) found a noticeable decrease in lipid digestion with the presence of methyl cellulose.

In conclusion, the results found evidence that hydrocolloid barrier could prevent the accessibility of the enzyme to the lipid phase, reducing the extent of lipid digestion. So it has been demonstrated that the application of this type of emulsions are feasible to obtain a cream with low digestibility.

3.3. Sensory analysis

3.3.1. Free Choice Analysis

Free Choice Profile (FCP) analysis was performed to determine the attributes that describe the cocoa creams. This analysis provides information about the spontaneous sensations that occur when the product is consumed (Varela and Ares, 2012; González-Tomás & Costell, 2006). The consumers generated different terms, subdivided into appearance, taste, aroma and texture attributes. The results from the FCP analysis are shown in Figure 2, that shows the two dimensions of Generalized Procrustes Analysis (GPA) graph. In this figure the most mentioned attributes and their frequency mention are summarized. The total amount of variance explained by the two dimensions was 73.46%. Dimension 1 accounted for 52.66% of the variance and was mainly related to appearance and texture terms. On the left side of the plot, lumpy appearance, lumpy texture and thick texture were placed which characterized creams elaborated with cellulose emulsions (MX, A4M and K4M). However, on the right side of the plot, terms as creamy appearance, homogeneus and bright appearance and creamy and soft texture were related to control cream and cream elaborated with XG emulsion. Dimension 2 accounted for 20.80% of the variance and was mainly related to taste and texture terms. The A4M and K4M creams were placed in the negative part of the Y axis, and are related to gummy and oily texture, while control cream, XG cream and MX emulsion appeared in the positive part of the Y axis, and were related to sweet and cocoa taste.

Therefore, in this study attributes related with appearance and texture perceived in mouth are obtained. In conclusion, of all creams studied, xanthan gum cream was the one that was related to sensory attributes similar to the control cream.

3.3.2. Liking test and CATA questions

The results of the liking of the different creams are shown in Figure 3. Cocoa control cream was the sample that presented the highest liking scores, although cream elaborated with xanthan gum emulsion obtained similar scores in appearance, colour and texture. Fat is a well-known enhancer of creaminess sensations, due to its lubricating and coating properties, and also is associated to enhanced flavour perception (Wijk, van Gemert, Terpstra, & Wilkinson, 2003). Although all creams have the same fat content, the fact that the control cream presents the oil in free form (not emulsified) could probably affect the different mouth perception, regarding texture and taste and therefore can affect the cream liking.

In order to determine the specific attributes that are related to the liking scores, a CATA test was made. The CATA questionnaire is a technique that is increasingly being applied in food research. It consists of multiple-choice lists of words or phrases from which consumers select those they consider appropriate for describing the sample they have tasted. (Smyth et al., 2006).

In this study, 17 sensory attributes were selected. A non-parametric Cochran's test was used to study the significant differences in the frequencies of the 17 attributes used to describe the creams (Table 3). As can be seen, frequencies of 13 of the 17 attributes studied presented significant differences that indicates that these terms could be used to describe perceived differences in the creams studied. No significant differences in strange aroma, cocoa aroma, strange taste and bitter taste were found. After that, a Correspondence Analysis (CA) was performed with the frequency of mention of the 13 attributes that exhibited significant differences. The first and second dimensions of the CA represent 66.55% and 22.82% of the total variability, respectively (Figure 4A); it could also be observed how the cellulose creams was placed in the negative part of the first dimension. Terms as “lumpy” and “sandy” were associated with the MX cream, and terms as “pasty”, “thick”, “tasteless” and “gummy” were associated with A4M and K4M creams. However, the positive part of the first dimension was related to control and xanthan gum creams described with attributes as “sweet taste”, "hazelnut taste", "bright", "creamy", "soft texture" and "spreadable". Lastly, a Multifactorial Analysis (MFA) (Figure 4B) was made to know which sensory attributes were associated with acceptability, and the layout of the samples is similar to samples shown in Figure 4A. The first and second dimensions explain 61.82% and 25.06% of the total variability, respectively. The control cream was the one that the consumers liked the most and was perceived as the highest in cocoa taste, sweet taste and hazelnut taste, although the xanthan gum cream sample also approaches acceptability values that are linked to the attributes of creamy, soft texture, bright and spreadable. The samples with the lowest liking ratings, however, were the cellulose creams due to the fact that they are lumpy, gummy, sandy, pasty, thick and tasteless. These results are similar to the results obtained with Free Choice Profile so when a large number of consumers is not available, it is possible to obtain a description of the samples from the sensory point of view using the Free Choice Profile technique. However, in liking test a large number of consumers still are need.

Therefore, considering the results obtained in the sensory analysis, it could be concluded that the cocoa cream made with the xanthan gum emulsion presented sensory attributes close to the control cream.

4. Conclusions

The results highlighted a relationship between the type of hydrocolloid used and the structural characteristics of cocoa creams. A4M cream was the cream related with thick and lumpy. *In vitro* digestion of the creams formulated with the emulsions caused a decrease in the consistency, except xanthan gum cream, which showed an increase in consistency after gastric digestion that could be related to a satiety perception. All the studied creams decreased the extent on lipid digestion after *in vitro* incubation, compared to control cream. However, in sensory analysis, only the cream elaborated with xanthan gum was related to positive attributes for texture, flavour an overall liking, close to the control cream, that was the most acceptable cream. It could be concluded that the reformulation of a cocoa cream with hydrocolloid-based emulsion is a good option to obtain food with improved lipid profile and low bioaccesibility. Note that the properties provided by xanthan gum cream make this product interesting in food design of satiating foodstuffs; it has the same sensory properties as a traditional cocoa cream with the advantage that increases the consistency in stomach phase and furthermore, provides a reduction in the lipid digestibility.

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Table 1. Area under the curve (AUC) calculated from the extrusion curves of the different cocoa creams.

COCOA CREAMS	**DIGESTION PHASE**	**AREA (N x mm)**
Control	Fresh	15.20a (2.22)
	Saliva	4.10b (0.96)
	Saliva Dilution	11.35a (1.95)
	Stomach	1.90c (0.46)
	Stomach Dilution	1.23c (0.09)
Xanthan gum	Fresh	23.15a (2.40)
	Saliva	15.34b (0.48)
	Saliva Dilution	21.78a (3.67)
	Stomach	10.60c (0.76)
	Stomach Dilution	6.00d (0.53)
K4M	Fresh	19.54a (1.83)
	Saliva	8.74b (2.03)
	Saliva Dilution	19.02a (1.21)
	Stomach	2.87c (0.31)
	Stomach Dilution	2.86c (0.77)
A4M	Fresh	42.78a (2.54)
	Saliva	18.37b (2.28)
	Saliva Dilution	38.66a (0.85)
	Stomach	4.30c (0.54)
	Stomach Dilution	6.39c (1.74)
MX	Fresh	24.73a (3.76)
	Saliva	14.59b (3.65)
	Saliva Dilution	23.28a (1.81)
	Stomach	5.87c (1.29)
	Stomach Dilution	4.55c (0.47)

[a,b,c]Means with different letter in columns for each digestion phase and each cellulose type indicate significant differences among the sample ($p<0.05$) according to Tukey test. Values in parentheses are standard deviations.

Table 2. Quantity of NaOH required to neutralize FFA released and oleic acid values after *in vitro* digestion.

COCOA CREAMS	ml NaOH	g oleic acid/g fat
Control	5.720a (0.696)	0.132a (0.016)
Xanthan gum	3.905b (0.600)	0.082b (0.004)
K4M	3.778b (0.728)	0.079b (0.010)
A4M	3.500b (0.196)	0.078b (0.003)
MX	3.144b (0.881)	0.063b (0.017)

[a,b]Means with different letter indicate significant differences among the sample ($p<0.05$) according to Tukey test. Values in parentheses are standard deviations.

Table 3. Frequency of selection of CATA terms and p value of Cochran's Q test for differences among cacao filling creams.

Attributes	**p (Cochran test)**	**Frequency of mention**				
		Control	**Xanthan gum**	**K4M**	**A4M**	**MX**
Lumpy	< 0.0001	31	21	31	34	51
Creamy	< 0.0001	36	28	25	15	11
Strange aroma	0.604*	5	7	5	3	3
Cocoa aroma	0.064*	27	26	26	28	16
Thick	< 0.0001	10	29	37	40	27
Bright	< 0.0001	41	30	32	9	9
Tasteless	0.000	3	22	13	14	19
Gummy	0.007	4	18	14	18	10
Cocoa taste	0.013	44	27	34	36	32
Hazelnut taste	0.000	67	49	50	48	46
Spreadable	< 0.0001	39	36	30	17	13
Sweet taste	0.023	37	25	29	24	24
Soft texture	< 0.0001	38	33	22	17	11
Pasty taste	0.001	14	23	33	36	26
Sandy taste	< 0.0001	37	26	45	32	50
Strange taste	0.055*	6	18	11	11	10
Bitter taste	0.924*	5	4	5	4	6

*Attributes that do not present significant differences with Cochran's test.

FIGURE LEGENDS

Figure 1. Back extrusion curves of cocoa creams (A: control; B: xanthan gum; C: K4M, D: A4M; E MX).

Figure 2. Two dimensions GPA plot of the differences among creams. The main descriptors correlated with the first two dimensions of the average space are listed on the boxes and the number of times that the descriptor was mentioned.

Figure 3. Acceptability scores of cocoa creams studied.

Figure 4. Representation of the sensory terms and cream samples: (A) Correspondence Analysis performed on data from the CATA question and (B) Multifactor Analysis using acceptability scores and CATA data for consumers of cocoa creams.

Figure 1A
Click here to download high resolution image

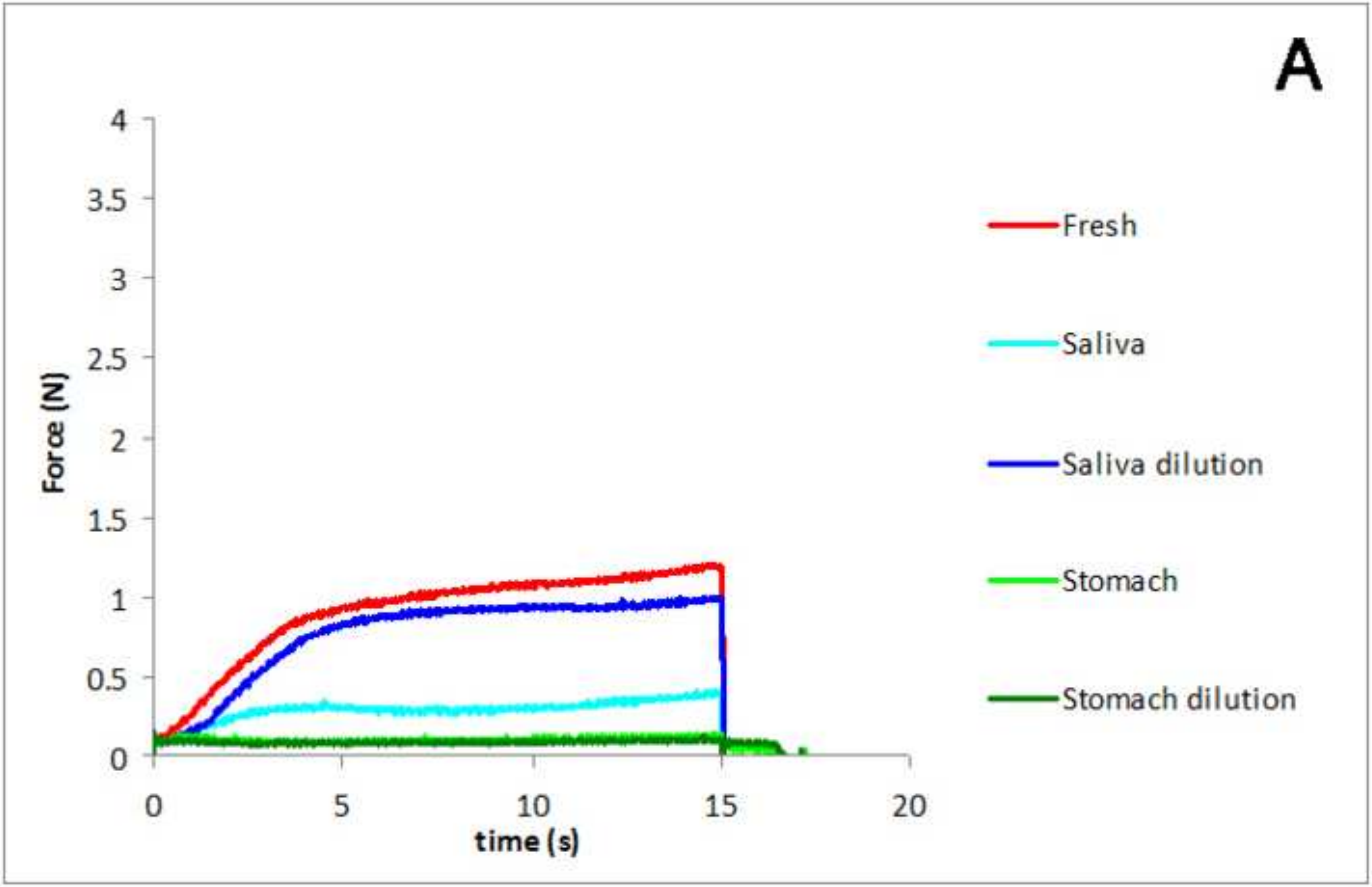

Figure 1B
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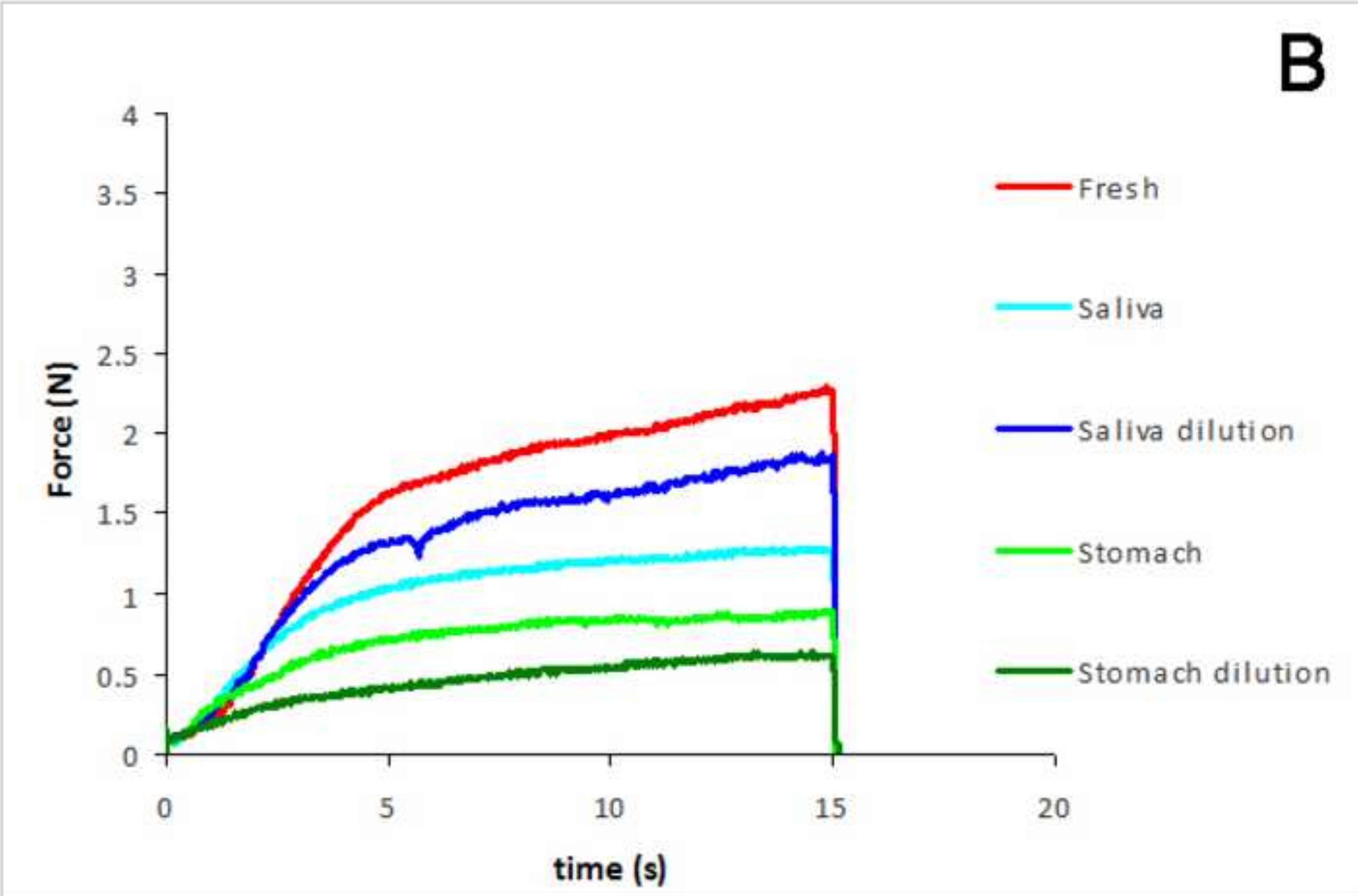

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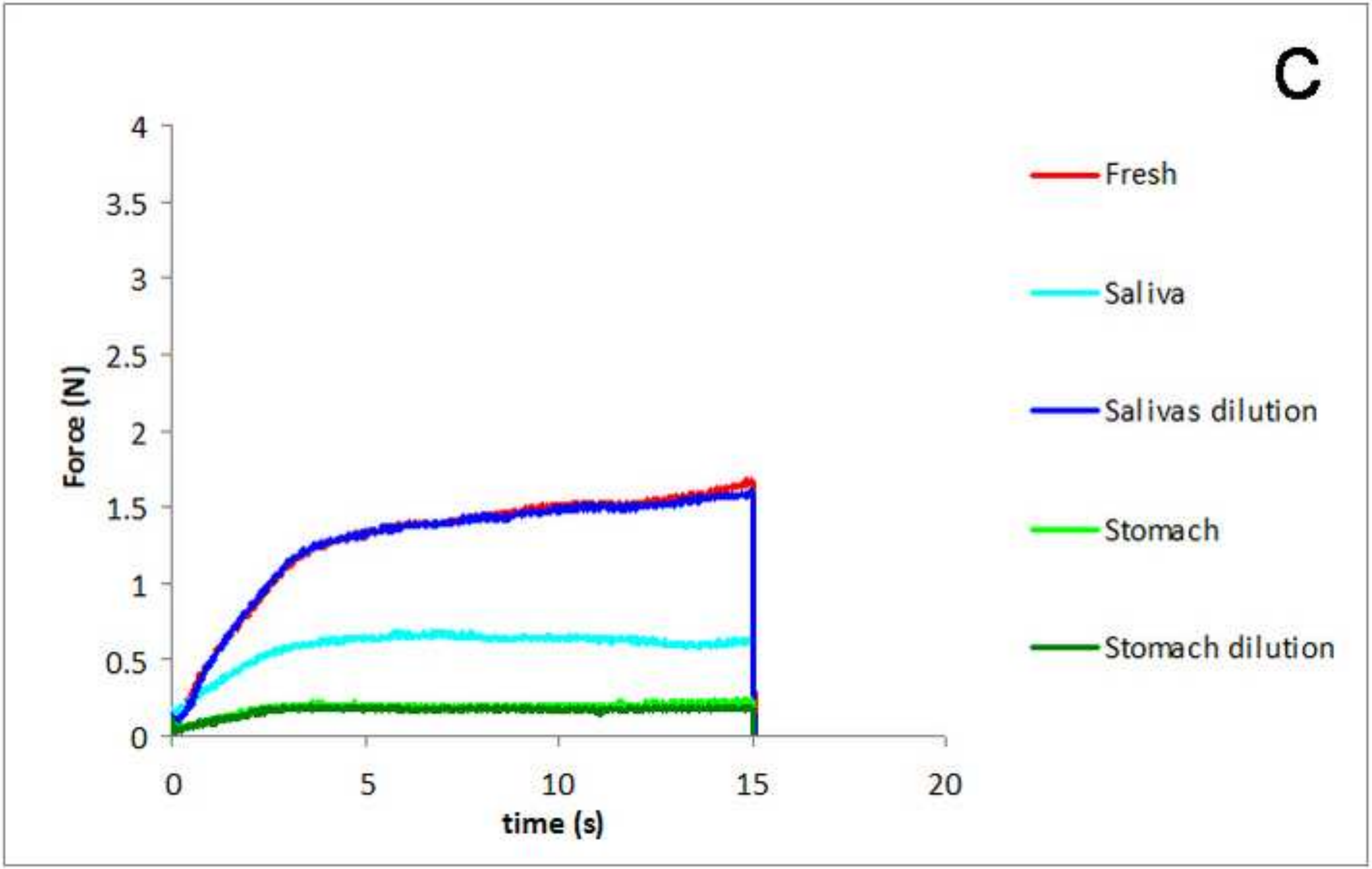

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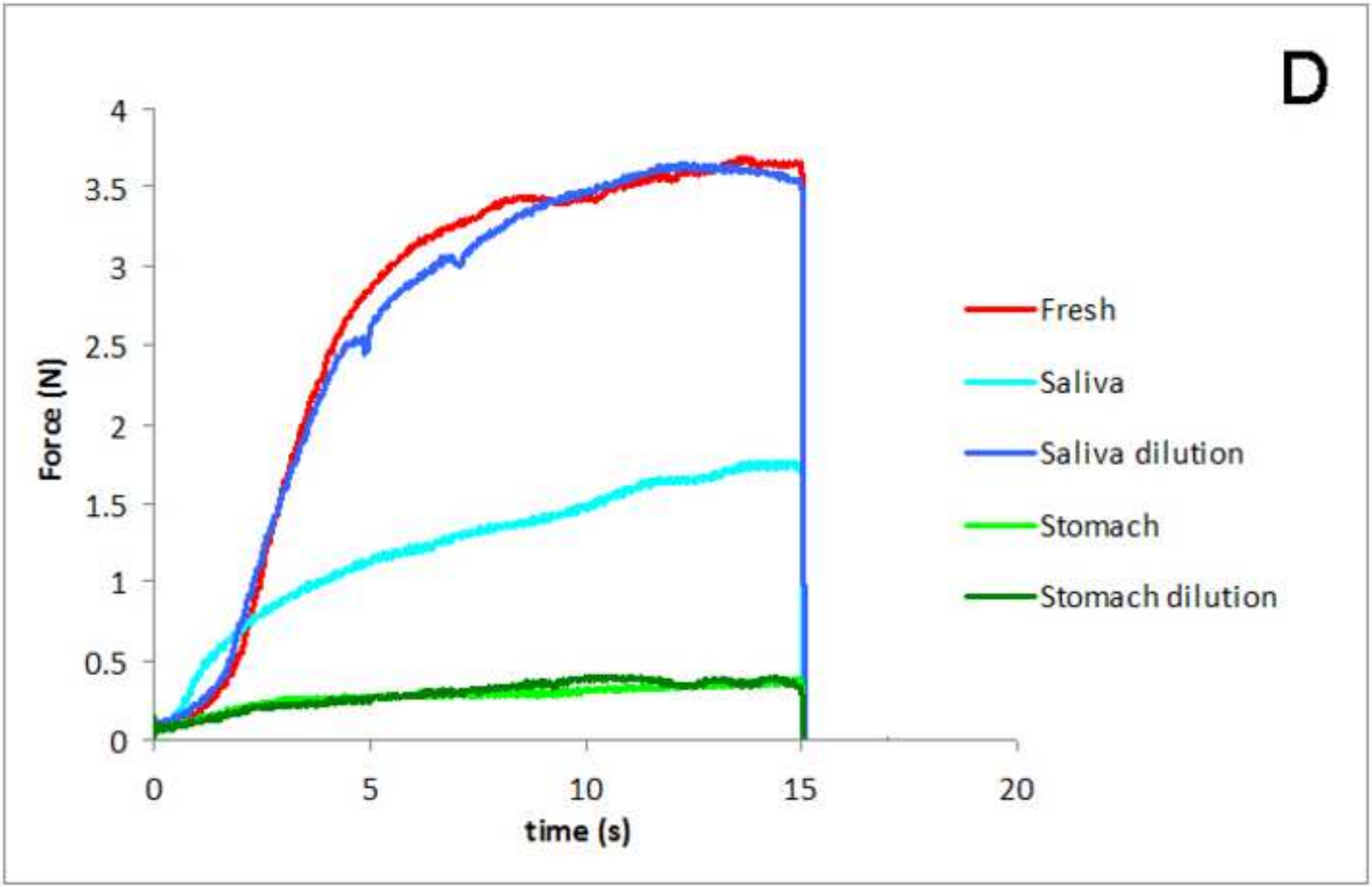

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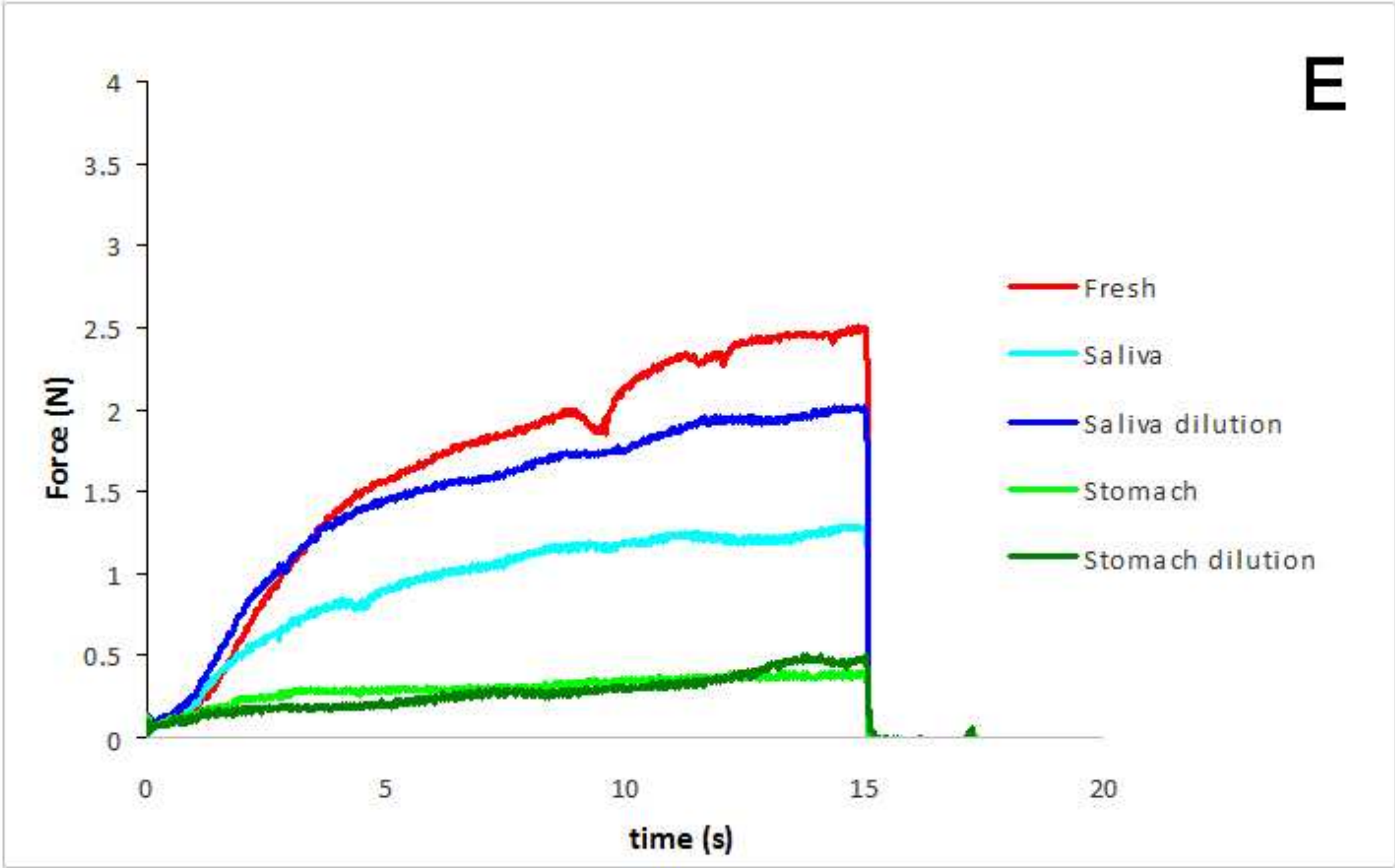

Figure 2

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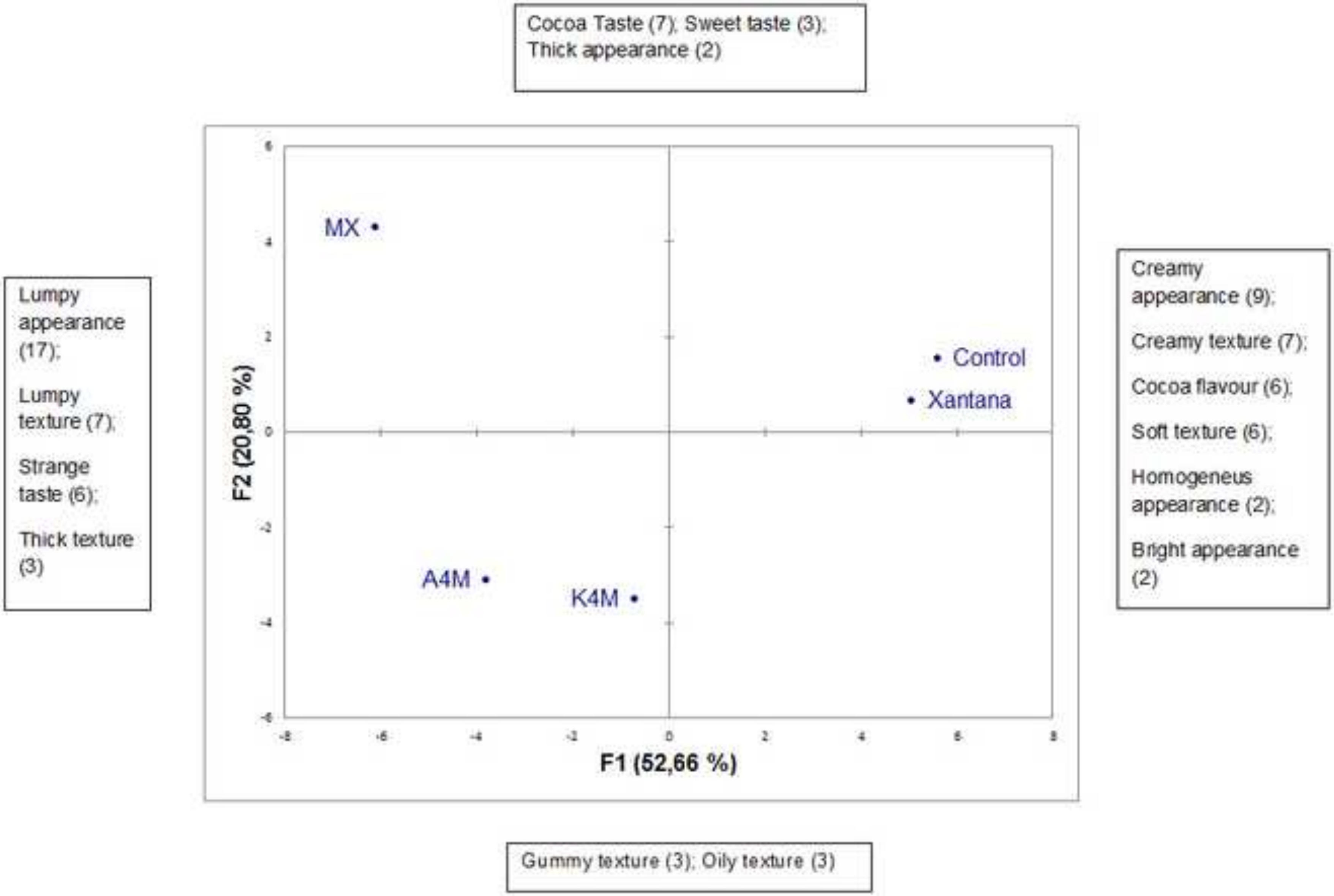

Figure 3
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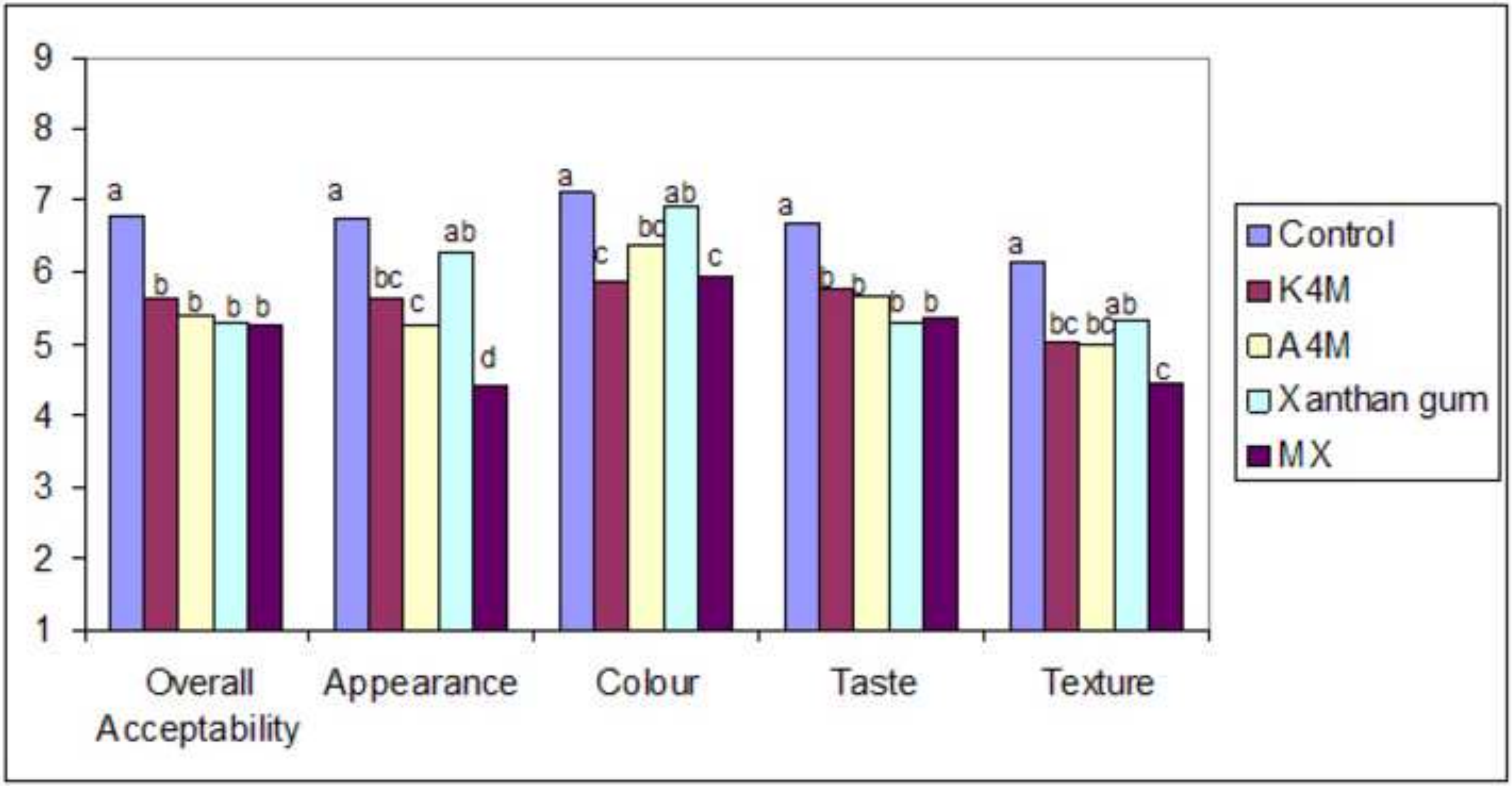

Figure 4A
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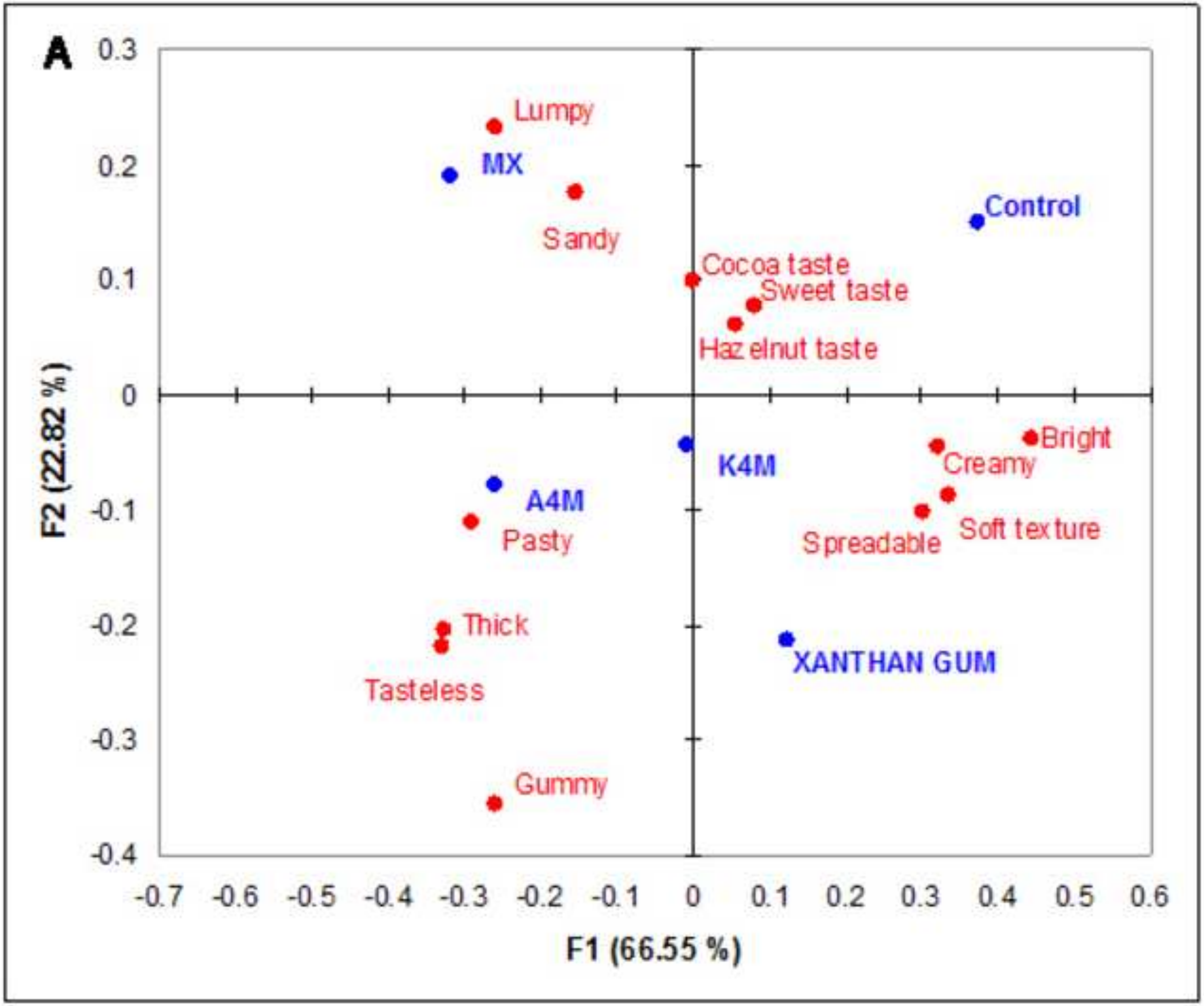

Figure 4B
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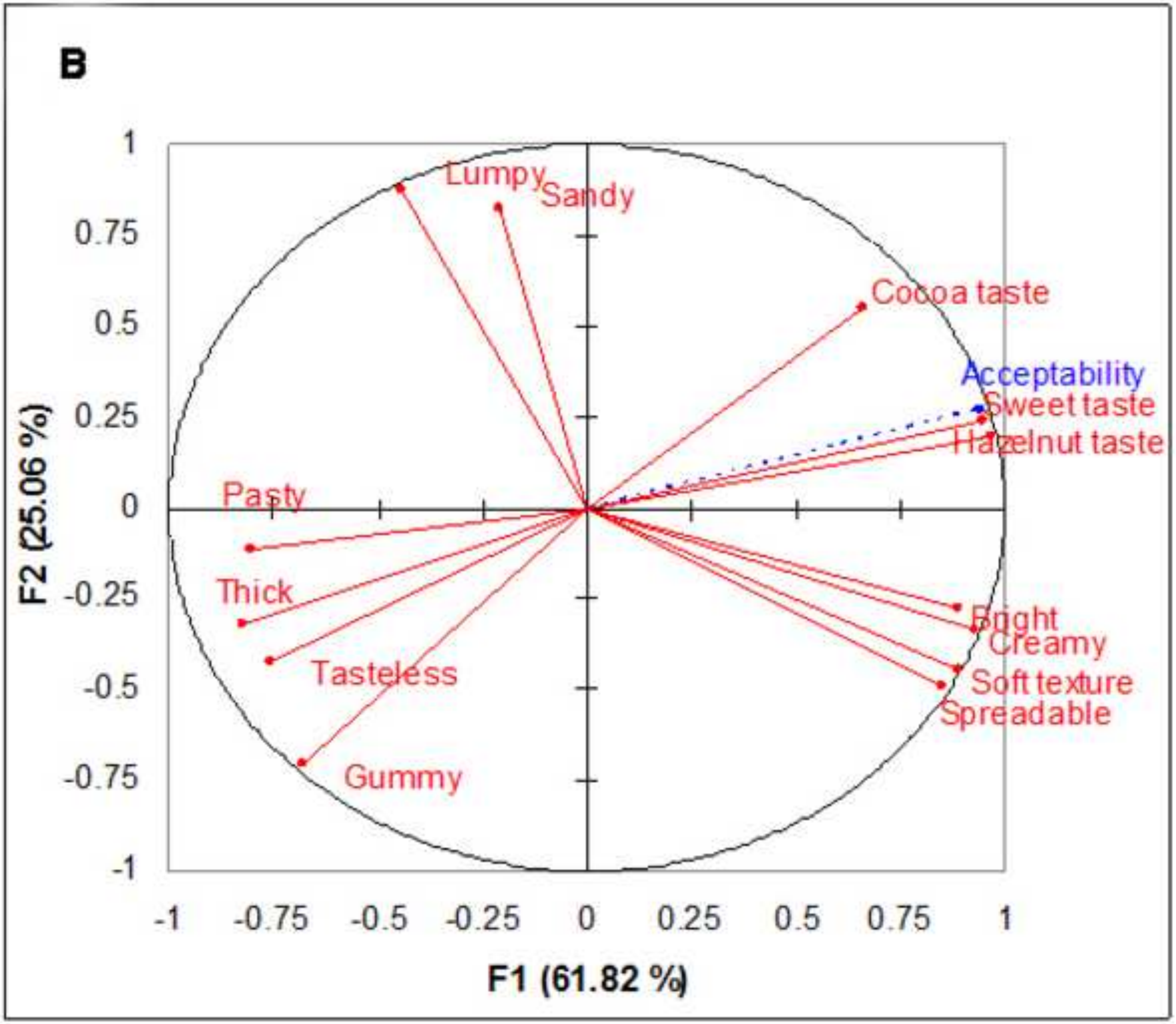

*Graphical Abstract

***Conflict of Interest**

CONFLICT OF INTEREST

Declarations of interest: none

ETHICS STATEMENTS FILE

In sensory analysis, consumers were informed of the procedure and they gave their consent by signing an internal declaration from the sensory laboratory.