USING DIFFERENT FIBERS TO REPLACE FAT IN SPONGE CAKES. IN VITRO STARCH DIGESTION AND PHYSICO-STRUCTURAL STUDIES

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USING DIFFERENT FIBERS TO REPLACE FAT IN SPONGE CAKES. *IN VITRO* STARCH DIGESTION AND PHYSICO-STRUCTURAL STUDIES

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ABSTRACT

This study assessed the effect of substituting 30% of fat by soluble, insoluble fiber, or a mix of both fibers in sponge cake quality, structure, acceptability and starch digestibility. The apparent viscosity of the different formulations was measured and micro baking was simulated. Texture profile tests were carried out and the crumb structure was examined. In vitro digestion was performed to study the digestibility of starch and a sensory test was carried out to know consumer acceptance. The soluble fiber (maltodextrin) affected the structure and quality of the cakes less than the insoluble fiber (potato fiber) and the use of soluble fiber in the formulation resulted in lower glucose release under in vitro conditions Moreover, the consumer did not found differences among the control cake and the cakes prepared with soluble fiber. Considering the results as a whole, soluble fiber may be used for partial replacement of fat in sponge cake formulations and constitutes an appropriate strategy for obtaining healthy sponge cakes.

Keywords: sponge cake, fiber, quality, structure, starch digestion

INTRODUCTION

Sponge cakes are a well-known product worldwide and are deeply rooted in the culture of each country. They are popular with consumers, who consider them delicious products with particular organoleptic characteristics (Matsakidou et al., 2010).

The major ingredients that give sponge cakes their specific properties include not only eggs, flour and sugar but also fat, which comprises approximately 15%-25% of the batter (Rodríguez-García et al., 2012). Fat contributes to air incorporation into the batter in the form of small bubbles, which will improve the stability of the batter minimizing the coalescence phenomena, thus increasing the volume of the cakes; fat also interferes with the continuity of the gluten, favoring the formation of a final product with a smoother, softer texture (Román et al., 2015). Nevertheless, it is the food component with the highest energy value and the high percentage of fat in sponge cakes gives them a high calorie content (Rodríguez-García, Salvador, et al., 2014; Zahn et al., 2010). Many studies have demonstrated the close connection between excessive fat consumption and the development of excess weight, obesity and certain cardiovascular diseases (Kratz et al., 2013; Mente et al., 2009). The World Health Organization (World Health Organization (WHO), n.d.) has warned that excess weight and obesity, considered as a typical problem of high-income countries, are becoming major public health problems in many parts of the world. The United Nations Food and Agriculture Organization (United Nations Food and

Agriculture Organization (FAO), n.d.) also states that good nutrition is the first line of defence against disease and requires special attention on the part of the food industry, starting with food design.

Nowadays, the nutritional value of food is becoming increasingly important, as well as the fact that the nutrients contained in them meet the specific needs of the individual. This is in agreement with the call of the WHO and the US Senate Commission on Nutrition's general dietary recommendations to limit the energy intake from total fat and raise the quantity of dietary fiber to a minimum of 22 g per day. In fact, there is increasing demand from consumers for low-fat, low-calorie, dietary fiber-rich products (Martínez-Cervera et al., 2012).

Dietary fiber is of increasing nutritional and clinical interest owing to its beneficial effects on health and is being used as an ingredient in a large variety of foods (Oh et al., 2014). Fiber can regulate intestinal function, protect the intestinal walls from contact with certain harmful substances, reduce cholesterol absorption and regulate blood glucose levels (Hardacre et al., 2015; Oh et al., 2014). The agreed definition of dietary fiber refers to carbohydrate polymers with ten or more monomeric units, which are not hydrolyzed in the small intestine of humans (Viebke et al., 2014) . Depending on chemical, physical, and functional properties, dietary fiber can be classified into soluble and insoluble fiber. Soluble dietary fiber (SDF) includes pectins, gums, inulin- type fructans and some hemicelluloses whereas insoluble dietary fiber (IDF) includes lignin, cellulose and some hemicelluloses. SDF is considered to have benefits on serum lipids, lowering the level of serum total cholesterol, while IDF is linked to laxation benefits (Quiles et al., 2016).

Several previous studies have investigated reducing the fat in sponge cakes or other bakery products by replacing it with different types of fiber, such as inulin (Rodríguez-García, Salvador, et al., 2014; Zahn et al., 2010), citrus pectin (Lim et al., 2014; Psimouli and Oreopoulou, 2013), peach fiber (Grigelmo-Miguel et al., 2001), cocoa fiber (Martínez-Cervera et al., 2011) or maltodextrin (Psimouli and Oreopoulou, 2013).These studies have found that fat replacement is feasible but affects the batter and cake properties depending on the type of fiber used for the replacement. Moreover, dietary fiber can influence the digestion of starch by reducing the starch breakdown and thus, reducing glucose release and absorption (Brennan, 2005).

Dietary fiber incorporated into starch-based foods can entrap starch granules and restrict the availability of water during gelatinization. As a result, the accessibility of starch granules to digestive enzymes is limited under human digestion, which results in the lowering of the glycemic index (Angioloni and Collar, 2011). However, *in vitro* studies show different results depending on the kind of dietary fiber used.

The aim of the present study was to investigate the functionality of soluble and insoluble fibers as replacers for 30% fat on the formulation of low fat cakes. Maltodextrin was used as soluble dietary fiber and potato fiber was used as insoluble fiber. The batter viscosity was measured and micro-baking was simulated to assess the evolution of air bubble growth. Texture profile analyses were also performed and the crumb structure was examined.

Lastly, the digestibility of the starch was measured through in vitro digestion tests and consumer acceptance was assessed.

MATERIALS AND METHODS

Ingredients

Sponge cakes were prepared with the following ingredients: wheat flour (Harinas Segura S.L, Torrente, Valencia, Spain; composition provided by the supplier: 13.5%-15.5% moisture, 9-11% proteins); white sugar (AB Azucarera Ibérica S.L.U., Madrid, Spain); egg yolk and white, both as pasteurized liquids (Ovocity, Llombay, Valencia, Spain); skimmed milk powder (Corporación Alimentaria Peñasanta, S.A., Siero, Asturias, Spain); refined sunflower oil (Aceites del Sur-Coosur, S.A., Vilches, Jaén, Spain); sodium bicarbonate E-500ii and citric acid E-300 (Sodas y Gaseosas A. Martínez, S.L., Cheste, Valencia, Spain); salt; Fibersol-2, composed of 90% resistant maltodextrin (Matsutani Chemical Industry Co. Ltd., Hyogo, Japan, total dietary soluble fiber 90%); Vitacel KF200, a potato fiber-rich food ingredient (J. Rettenmaier and Söhne Gmbh + Co Kg, rich in insoluble fiber (55%), total dietary fiber 65%); and distilled water.

Batter and cake preparation

The four formulations studied (Table 1) were the control formulation (C) and three further formulations in which 30% of the sunflower oil was replaced by a soluble fiber (SF), an insoluble fiber (IF), or a 50/50 mixture of the two fiber ingredients (M). Extra distilled water was added at ratios of 1:1 Fibersol-2 to water and 1:4 Vitacel KF200 to water, as recommended by the suppliers of the fiber ingredients.

The batters were prepared using the ‘all in’ mixing procedure of Rodríguez-García et al (2014), with a few modifications. Firstly, all the liquid ingredients — egg white, yolk, milk and water — were placed in a Kenwood Major Classic mixer (Kenwood, Havant, UK). The solid ingredients — flour, sugar, Fibersol-2 and/or VitacelKF200, bicarbonate of soda, citrus acid and salt — were then added to the same bowl. The last ingredient added was the sunflower oil. To achieve homogeneous batters, all the ingredients were mixed for 30 s at 202 rpm, followed by 1 min at 260 rpm and 3 min at 320 rpm.

To bake each cake, 700 g of batter were poured into a 20 cm diameter Pyrex® mold and placed in a conventional oven (Electrolux, model EOC3430DOX, Stockholm, Sweden) that had been preheated to 180°C for 30 min. They were baked at 180°C for 47 min. After removing the cakes from the oven, they were left to cool for at least 1 hour and 30 minutes before they were examined. All the batters and cakes were prepared in triplicate and the tests were performed within 24 hours of their preparation.

Apparent viscosity

Batter viscosity was measured with a Haake Viscotester 6 R Plus viscometer (Thermo Scientific, Walthman, MA), using an R3 spindle at 6 rpm at room temperature. The samples were placed in a thermostatic bath to maintain a temperature of 25°C. The measurements were made in duplicate for each batter and in triplicate for each formulation.

Batter image analysis (micro baking simulation)

Microscope observation was performed during the micro baking simulation using a temperature-controlled stage (Analysa-LTS350, Linkam Scientific Instruments Ltd., Surrey, UK) under the lens of a light microscope (Nikon ECLIPSE 80i, Nikon Co. Ltd., Tokyo, Japan). A drop of the sample was placed in the concavity of the glass slide, which was placed on the temperature-controlled stage. During heating, the temperature ramp was controlled by a refrigeration system with a liquid nitrogen pump (Linkam). The temperature profile employed was 1.5°C/min from room temperature (25°C) to 105°C. The batter samples were observed at 4X magnification (x4/0.13∞/-WD 17.1 objective lens, Nikon). Photographs were taken with a camera (ExWaveHAD, model DXC-190) fitted to the microscope and connected to a computer. During the micro baking simulation, a video was recorded with photographs taken every 10 s in a 640x540 pixel format, using the microscope software (Linksys 32, Linkam). Three samples of each formulation were examined. The images were analyzed with ImageJ software (National Institute of Health, Bethesda, MD).

Macroscopic structure of the crumb

The cakes were cut in half through the center and scanned with an HP Scanjet G2710 (Hewlett-Packard, Palo Alto, CA, U.S.A) at a resolution of 300 dpi. Central sections of cake with a field size of 10x4 cm were analyzed. The cropped image was split into color channels, the contrast was enhanced and the image was thresholded and binarized with the aid of the ImageJ software program (National Institutes of Health, Bethesda, Maryland, USA). Total cell area within the crumb (%) and cell size (mm^2) were calculated. Four images of each formulation were analyzed.

Sponge cake texture

The textural properties were assessed with a TA-TXTplus texture analyzer (Stable Microsystems, Ltd., Godalming, UK), using the Texture Exponent Lite 32 program (version 6.1.4.0, Stable Microsystems). A texture profile analysis (TPA) was performed on four cubes (3x3x3 cm) cut from the center of the cake after removing the crust. The cubes were compressed to 40% of their original height at a test speed of 1 mm/s with a 5 s resting time between the two compression cycles. The trigger force was 5 g. The cubes were compressed with a 5 cm diameter cylindrical aluminum probe. After the two compression cycles, the following measurements were recorded: hardness, springiness, cohesiveness, chewiness and adhesiveness. Measurements were carried out in duplicate.

Field emission scanning electron microscopy (FESEM)

For each formulation studied, 0.5 cm sided cubes were cut, frozen at -80°C and lyophilized (Lyoquest 55, Telstar, Terrassa, Barcelona, España). The samples were then vacuum coated with platinum and observed under a field emission scanning electron microscope (model Ultra 55 FESEM, Zeiss, Oberkochen, Germany). Each formulation was analyzed in duplicate.

In vitro digestion

Digestion of the sponge cakes covered three stages: oral, gastric and intestinal.

For oral digestion, the protocol described by Smith et al (2015) was followed, with a few modifications. Consequently, 10 g of cake were crumbled by hand and 3,5 mL of saliva solution, previously incubated at 37°C, were added. This mixture was ground with a blender (Ufesa, U1EBB40001; BP-4500) for 15 s, then 70 mL of bidistilled water were added and mixed in by hand for 1 min to simulate mastication. The saliva solution was prepared as described by Mishellany-Dutour et al(2011).The following were dissolved in 1 L of bidistilled water: 5.208 g of $NaHCO_3$, 1.369 g of $K_2HPO_4.3H_2O$, 0.877 g of NaCl, 0.477 g of KCl, 0.441 g of $CaCl_2.2H_2O$, 2.16 g of mucin from porcine stomach type II (PGM Sigma M2378) and 8.70 g of α-amylase from porcine pancreas type VI-B (Sigma A3176).

In the gastric stage, to digest 25 g of sponge cake that had already been orally digested, 25 g of gastric fluid were placed in the digester, composed of a glass reactor with a thermostat-controlled jacket and continuous magnetic stirring fitted to a controlled temperature water circulator. The gastric fluid was preincubated at 37°C for 5 min at pH 2. The sample was added to the reactor, the mixture was adjusted to pH 2 with 2 M HCl, 0.006 g of pepsin (Sigma P7000) were added and the mixture was incubated for 1 hour at 37°C with stirring. The electrolyte solution that constituted the gastric fluid was prepared by dissolving the following in 1 L of distilled water: 3.1 g of NaCl, 0.11 g of $CaCl_2$, 1.1 g of KCl, 5.68 mL of 1 M $NaCO_3$. The pH was adjusted to 2 with 2 M HCl.

For the intestinal stage, the pH of the sample was raised to 6 with 1 M $NaCO_3$ to which pancreatin (Sigma P1750, 4xUSP) and bile salts (Sigma B8631) had been added. The pancreatin and bile salt solution was prepared with 0.1 g of pancreatin and 0.625 g of bile salts to 25 mL of 0.1 M $NaHCO_3$ (Rufián-Henares and Delgado-Andrade, 2009). Amyloglucosidase (A7095≥300 U/mL, Sigma) was then added at 0.2 mL/g of starch in accordance with Oh et al (2014) and Soong et al (2014). The pH was raised to 7.5 with 0.1 M $NaHCO_3$ and the mixture was incubated at 37°C for 3 h with stirring. Aliquots were removed at 0, 20, 60, 90, 120 and 180 min of digestion, immediately adding 1.4 mL of ethanol to stop the reaction (Bae et al., 2013), and centrifuged at 3000 rpm for 3 min. The glucose concentration was then measured with the GOPOD assay kit at 510 nm. For this measurement, 0.1 mL aliquots of the supernatant were taken, 3 mL of the GOPOD reagent were added, the sample was incubated at 40-50°C for 20 minutes and the absorbance was read at 510 nm.

The experimental data were fitted to the first-order equation proposed by Goñi et al (1997) [$C= C_{\infty} (1-e^{-kt})$], where C is the concentration at t time, C_{∞} is the equilibrium concentration, k is the kinetic constant and t is the chosen time.

Sensory Analysis

Consumers were recruited among students and employees of the Universitat Politècnica de València. A total of 82 untrained panelists (consumers) aged 22-63, were used for the study. Of the participants, 49% were women and 51% men.

The samples were assessed in a standardized tasting room equipped with individual booths. Each consumer received four pieces of cakes (C, SF, M and IF) coded by three digit random numbers. The pieces of cakes were served at room temperature in random order. Water was supplied to clean the consumers' mouths between each sample.

Consumer acceptance testing was done using a 9-box structured hedonic scale to score the "appearance", "texture", "taste" acceptability and "overall acceptance" of the product (from 1= "I dislike it extremely" to 9= "I like it extremely").

Statistical analysis

Analysis of variance (ANOVA) was used for statistical analysis of the results. The least significant differences (LSD) were calculated with a significance level of $p<0.05$. The Statgraphics Centurion XVI.II statistical program (StatPoint Technologies, Inc., Warrenton, VA, USA) was used for this purpose.

RESULTS AND DISCUSSION

Apparent viscosity

The batter viscosity results for the sponge cake formulations studied are expressed in mPa.s. On reducing the fat content by 30%, a significant ($p<0.05$) reduction was observed in the viscosity of the IF (7675.27 ± 53.53) and M (9032.95 ± 233.36) batters in comparison to the control (10732.04 ± 348.42), however no significant ($p>0.05$) difference was observed between SF (10724.73 ± 470.06) and control batters. This tendency has previously been reported by other authors (Rodríguez-García, Salvador, et al., 2014; Román et al., 2015; Zahn et al., 2010) who also obtained low viscosity values on replacing fat with soluble fibers and functional ingredients and lower viscosity values at higher rates of replacement.

Bearing in mind that the fat reduction level was constant in the present study, the ingredient with insoluble fiber led to the greatest reduction in viscosity, This rheological behavior is largely due to the greater quantity of water added to the insoluble fiber formulation (1:4) compared to the batter with soluble fiber (1:1), giving IF a higher ratio of liquid to solid ingredients, which led to lower viscosity values.

Light microscopy and image analysis of the batters

Figure 1A shows images of the batters of the different formulations (C, SF, M and IF) at different temperatures (30, 60, 90 and 100 ºC) during micro baking simulation.

Visual examination of the batter images showed a clear air bubble expansion effect due to the lower fat content and the addition of soluble and insoluble fibers together with water. In batter C, the size of the bubbles increased in a uniform, controlled way, distributing the bubbles evenly as the temperature rose.

In general, the reduction in fat and the addition of soluble and insoluble fibers allowed more bubbles to be incorporated during the mixing process (Figs. 1A and 1B). As the temperature rose, the bubbles naturally expanded. Bubble expansion was higher in the IF batter, which were the least stable at rising temperatures, with some of the bubbles losing their identity at 100°C as they coalesced with neighboring bubbles.

The images were analyzed to quantify the bubble size distribution during micro baking. Figure 1B presents histograms of the bubble size distributions at different temperatures. The C formulation batter incorporated fewer bubbles (Fig 1B) and showed a tendency to regular distribution of bubble sizes during heating, compared to the other batters. This behavior could be due to the greater apparent viscosity of this batter (C), which would help to make the air bubbles more stable, delaying their movement through the batter and slowing down their disproportionate growth and coalescence as observed previously by Rodríguez-García, Salvador, et al., 2014).

In general, a lower apparent viscosity of the replaced batters (SF, M and IF) may have allowed occluding more air during mixing; so, more number of bubbles per field is observed at the beginning of the micro baking process, particularly in the case of batters SF and M. During the micro baking process the air bubble sizes acquired an irregular distribution but towards the end of the heating scale (at 90ºC and 100ºC), the IF batter was found to have a higher percentage of larger bubbles.

The considerable reduction in the apparent viscosity of the IF batters and resulting reduction of air bubble stability in these samples increased the mobility, disproportion ratio, coalescence and size of the bubbles.

Macroscopic structure of the crumb

Figure 2 shows scanned, contrasted and binarized images of the different cakes (C, SF, M and IF).

Visual analysis of these cake images shows a practically uniform crumb macrostructure in the control cake (C). In contrast, a series of diffusion pathways appeared in the crumb of the reduced-fat sponge cakes. These pathways were less noticeable in the SF cake and more noticeable and numerous in the IF cake.

The images of the cakes were also analyzed to quantify the crumb macrostructure results (table 2). IF presented a significantly ($p<0.05$) higher cell size and a higher total cell area values compared to the other cakes. Consequently, IF presented a more aerated structure with bigger cells. These results agree with the tendency observed in the sponge cake batters during micro baking, as described in the previous section- IF batter was found to have a higher amount of larger bubbles at the end of the micro baking-. In turn, this is intrinsically affected by viscosity; thus, in IF crumb cake the rising percentage of air would be directly related to the low viscosity found in IF batter. Changes in the thermosetting mechanism, as a consequence of the extra water added, could also be responsible for the presence of the diffusion path ways in replaced cakes.

Cake texture

The results of the parameters obtained from the texture profile analysis curves for the sponge cakes under study are presented in Table 2.

The 30% fat reduction with fiber generated significantly ($p<0.05$) higher hardness values in SF, M and IF samples. IF was the hardest of the samples. Hardness values followed the trend IF>M>SF, and significant differences were observed among all of them This means that more force was required to compress the IF cake than the other formulations. Eslava-Zomeño et al (2016) obtained significantly higher hardness values for sponge cakes made with Optisol™ 5300 at different fat replacement ratios. Psimouli and Oreopoulou (2013) also found significantly higher hardness values in sponge cakes prepared with different carbohydrates as fat replacers..

The chewiness values showed a similar trend. The chewiness values of the control cake were significantly lower ($p<0.05$) than those of the other sponge cakes. When 30% of the fat was replaced by adding soluble and/or insoluble fibers to the formulations, chewiness increased significantly ($p<0.05$). IF was the chewiest cake. This means that greater energy was needed to chew the IF cake enough to be swallowable.

In general, the tendency for these parameters to increase could be related to the reduction in the batter viscosity of the respective formulations. In the control formulation batter, both the low number of bubbles and the distribution and homogeneous expansion of small bubbles influenced its low hardness values. In contrast, the greater variation in bubble size observed on adding the soluble and insoluble fibers, particularly the latter, increased the cake hardness considerably. Also, bearing in mind that one of the functions of fat is to make cakes smooth and soft, reducing the fat and adding soluble and insoluble fibers could be expected to increase the hardness of the cakes and, consequently, their chewiness.

The cohesiveness and adhesiveness values showed no significant ($p>0.05$) differences between the sponge cakes studied. The lower fat content and addition of soluble and insoluble fibers did not influence the work needed to compress the samples a second time compared to the first, nor did they alter the work needed to detach the compression probe from the sample.

The springiness values showed no significant ($p>0.05$) differences between C and SF or between M and IF, though the latter pair presented significantly ($p>0.05$) higher springiness.

The use of insoluble fiber in the formulation of the cakes seems to influence in the texture parameters as IF cake is the one with highest hardness, chewiness and springiness values.

Field emission scanning electron microscopy (FESEM)

The microstructure of the soluble fibers (Fibersol-2), insoluble fibers (Vitacel K200) and C, SF and IF cakes can be seen in the images obtained through field emission scanning electron microscopy (FESEM), shown in Figure 3.

The fibers showed considerable differences in structure. The soluble fiber was made up of numerous particles of varying sizes and shapes, although most were granular and presented a smooth appearance. The insoluble fiber had the typical rough appearance of plant cells, with visible cell walls (labelled pc) and transport tissues (labelled vc).

The structure of the control cake (C) can be seen to be composed of a gluten network, formed by the flour, which contained the other ingredients. The partially gelled starch granules were embedded in the gluten network and the oil acted as a lubricant, creating a continuous structure. The structures of the SF and IF cakes, with a 30% reduction in fat, were influenced by the characteristics of the respective fibers and presented a more irregular microstructure, since there was a smaller coating of oil. In the SF cakes, the partially gelled starch granules distributed irregularly through the cake matrix were very evident, as they retained their identity. In the IF cakes, the starch granules were deeply embedded in the matrix, giving rise to a more compact structure that can be related to their harder texture.

***In vitro* digestion**

Figure 4 shows the digestibility curves of cakes C, SF and IF after in vitro digestion. No significant ($p>0.05$) differences were observed between the samples at 20, 60, 90 and 120 minutes of digestion. However, SF presented significantly ($p<0.05$) lower values than the other cakes after 180 minutes.

The parameters obtained after fitting the curves using the first order model described by Goñi et al. (1997) are shown in Table 3. Although the kinetic constant (k), which indicates the rate of starch hydrolysis is augmented, it can be observed that the area under the hydrolysis curve after 180 min (AUC 180) and the equilibrium concentration (C∞) values were the lowest for SF, being significant ($p<0.05$) for C∞ values. AUC 180 is a comprehensive parameter for the starch hydrolysis, relating the glucose release over a hydrolysis period of 180 min (Gularte et al., 2012) and C∞ indicates the concentration at the equilibrium point, and a higher concentration of final product reflects increased digestibility of starch (Dura et al., 2014). Taking into account the results obtained for AUC 180 and C∞ values, the use of soluble fiber in the formulation of the cake would result in lower glucose release under *in vitro* conditions. This could be related to the field emission scanning electron microscope (FESEM) images, where the starch granules in the SF cake matrix were observed to be less gelled than those of the IF cake. Moreover, the soluble fiber with greater water absorption capacity would compete with the starch for the available water during sponge cake processing, leading to low starch gelatinization and consequently reducing the release of glucose during *in vitro* digestion in the case of SF cake.

Sensory acceptance

Table 4 presents the mean liking scores for the “appearance”, “texture”, “taste” and “overall acceptance” of the control cake and the cakes with the different type of fibers.

Statistical analysis showed that the control cake (C) and the cakes where fat was replaced by soluble fiber (SF) and the mix of fibers (M) did not differ significantly ($P < 0.05$) in all the attributes. However, IF cake obtained the lowest value when all the attributes were scored; being significantly ($P < 0.05$) lower than the other three samples for “texture” and “overall acceptance” attributes.

These results revealed that quality differences due to fat replacement by soluble fiber or by the mix of fibers were not perceived by consumers. However, the replacement by insoluble fiber gave place to significantly ($P < 0.05$) lower scores for “texture” and “overall acceptance” attributes. If the texture hedonic results are compared with the instrumental measurements it can be observed that the highest hardness and chewiness may have had an important negative influence in hedonic acceptability. In this context, IF was the hardest of the samples but its texture was the less liked by consumers.

CONCLUSIONS

Replacing 30% of the fat in a sponge cake formulation with ingredients that are rich in insoluble fiber and extra water caused a reduction in viscosity. As a result of their low viscosity, the batters with insoluble fiber incorporated a greater quantity of air bubbles during mixing, as observed in microbaking, and IF batter presented larger bubble size than the other formulations at the same temperatures. In the macroscopic analysis of the crumb, the IF cake also showed a larger quantity of diffusion pathways and a greater percentage of air. Replacing 30% of the fat in the sponge cake formulation with ingredients that are rich in soluble and/or insoluble fiber also caused increased hardness and chewiness. The IF cake was spongier because it contained a greater quantity of air but was also harder, which is related to the more compact matrix observed by FESEM. During in vitro digestion, SF showed a lower glucose release at 180 minutes. Regarding the sensory acceptance, the consumers did not find differences among C, SF and M cakes, however, IF cake was the less liked by consumers. Overall, considering the physicochemical, sensory and nutritional quality, soluble fiber may be used for partial replacement of fat in sponge cake formulations and constitutes an appropriate strategy for obtaining healthy sponge cakes.

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574 Table 1. Composition of the formulations studied (% flour base)

575

Ingredient*	C	SF	M	IF
Flour	100	100	100	100
Sugar	100	100	100	100
Egg yolk	27	27	27	27
Egg white	54	54	54	54
Milk	50	50	50	50
Oil	46	32.2	32.2	32.2
Soluble fibre	0	4	2	0
Insoluble fibre	0	0	2	4
Water	0	4	10	16
Sodium Bicarbonate	4	4	4	4
Citric acid	3	3	3	3
Salt	1.5	1.5	1.5	1.5

576

577 C: control cake; SF: cake with soluble fibre; M: cake with a mixture of soluble and insoluble
578 fibre; IF: cake with insoluble fibre

579

Table 2. Macroscopic structure of the crumb and textural properties of the cakes.

	Crumb Structure		Cake Texture				
Sample	Cell Size (mm^2)	Total Cell Area (%)	Hardness (N)	Chewiness (N)	Cohesiveness	Springiness	Adhesiveness (g·s)
C	1.0^a (0.1)	28.13^a (2.37)	4.98^a (0.46)	3.15^a (0.28)	0.71^a (0.01)	0.88^a (0.01)	2.55^a (1.55)
SF	1.0^a (0.2)	30.27^a (4.65)	5.62^b (0.41)	3.57^b (0.22)	0.72^a (0.01)	0.88^a (0.01)	3.17^a (1.01)
M	1.1^a (0.1)	32.45^a (2.11)	6.22^c (0.82)	3.98^c (0.49)	0.72^a (0.00)	0.89^b (0.01)	2.82^a (2.01)
IF	1.4^b (0.2)	38.52^b (3.69)	6.95^d (0.69)	4.46^d (0.43)	0.72^a (0.00)	0.90^b (0.01)	2.73^a (1.12)

Figures in brackets are standard deviations. $^{a, b, c, d}$ Means with different letters in the same column differ significantly ($p<0.05$). C: Control cake, FS: cake with soluble fibre; M: cake with a mixture of soluble and insoluble fibre; FI: cake with insoluble fibre.

Table 3. Kinetics of the in vitro starch digestibility.

Sample	AUC 180	C_∞ (g/100)	k (min^{-1})
C	2075.7[a] (15.6)	14.1[a] (0.4)	0.030[a] (0.003)
SF	1794.1[a] (279.3)	11.6[b] (1.5)	0.039[a] (0.007)
IF	1971.6[a] (64.6)	13.1[a] (0.3)	0.033[a] (0.001)

Values in brackets are standard deviations. [a, b, c, d] Means with different letters in the same column differ significantly ($p<0.05$).

C: Control cake, SF: cake with soluble fibre; M: cake with a mixture of soluble and insoluble fibre; IF: cake with insoluble fibre.

592 Table 4. Liking for appearance, texture, taste and overall
593 acceptance of cakes.

Sample	Appearance	Texture	Taste	Overall acceptance
C	7.00[a]	6.90[a]	7.00[a]	7.15[a]
SF	6.93[a,b]	6.78[a]	6.91[a,b]	7.04[a]
M	6.80[a,b]	6.73[a]	6.90[a,b]	7.03[a]
IF	7.03[b]	6.18[b]	6.46[b]	6.53[b]

594 Values are mean (n=82). [a, b, c, d] Means with different letters in the same column
595 differ significantly ($p<0.05$). C: Control cake, SF: cake with soluble fibre; M: cake
596 with a mixture of soluble and insoluble fibre; IF: cake with insoluble fibre.

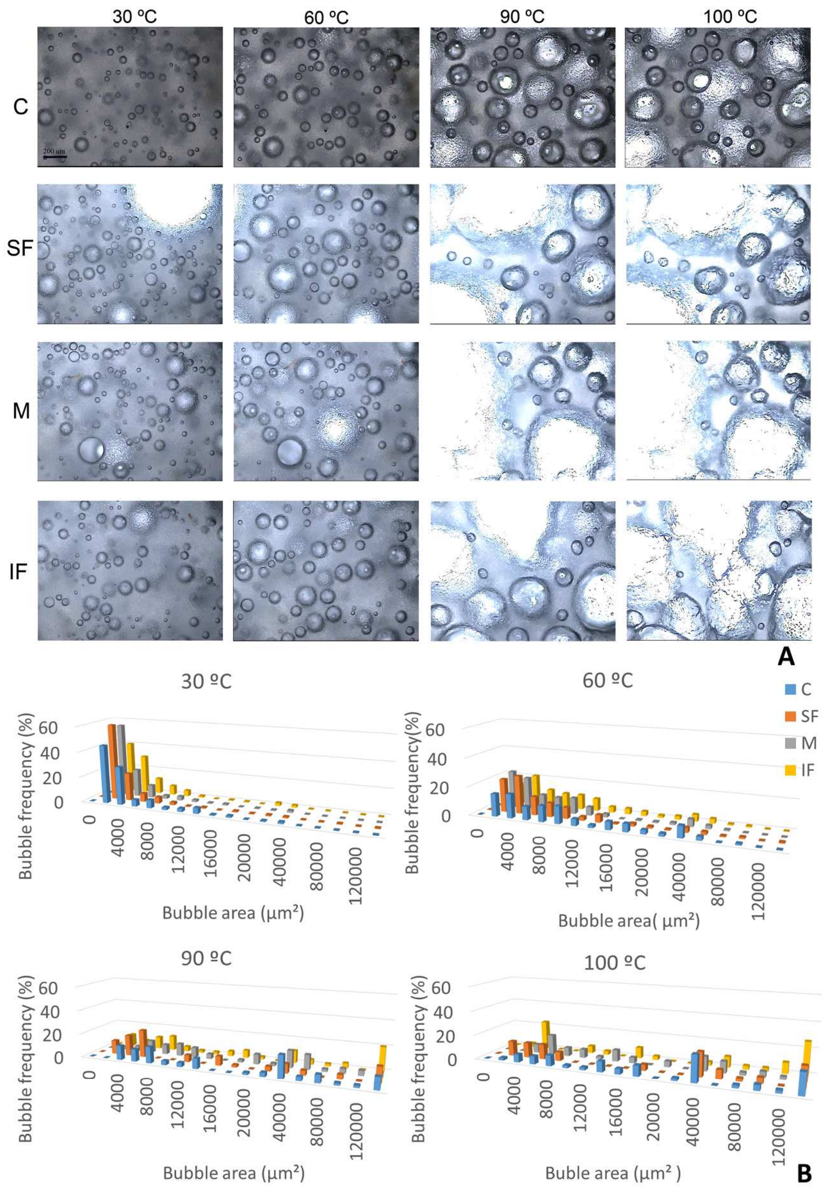

FIGURE 1. A: Light microscopy images of bubble expansion at different temperatures during microbaking. C: control cake; SF: sponge cake with soluble fiber; M: sponge cake with a mixture of soluble and insoluble fiber; IF: sponge cake with insoluble fiber. B: Histograms of bubble size distribution. C: blue, SF: orange, M: grey, IF: yellow.

175x250mm (300 x 300 DPI)

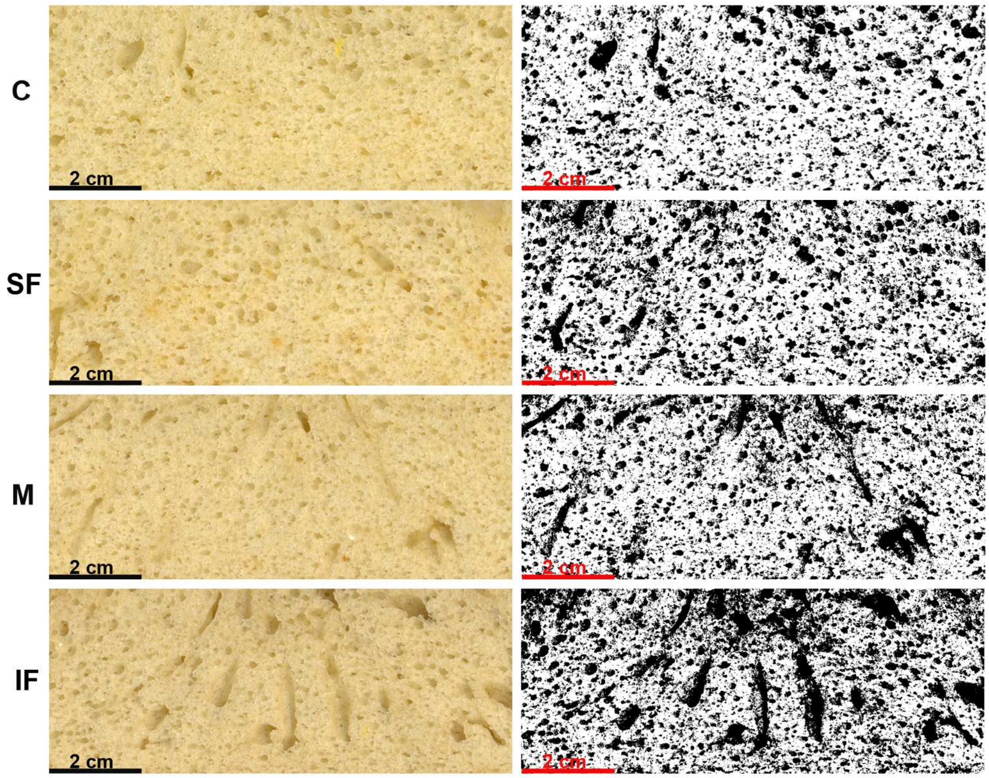

FIGURE 2. A: Scanned images of C, SF, M and IF sponge cakes, field size 4x10 cm, and corresponding binarised images (118 pixels/cm). C: control cake; SF: sponge cake with soluble fiber; M: sponge cake with a mixture of soluble and insoluble fiber; IF: sponge cake with insoluble fiber

175x142mm (300 x 300 DPI)

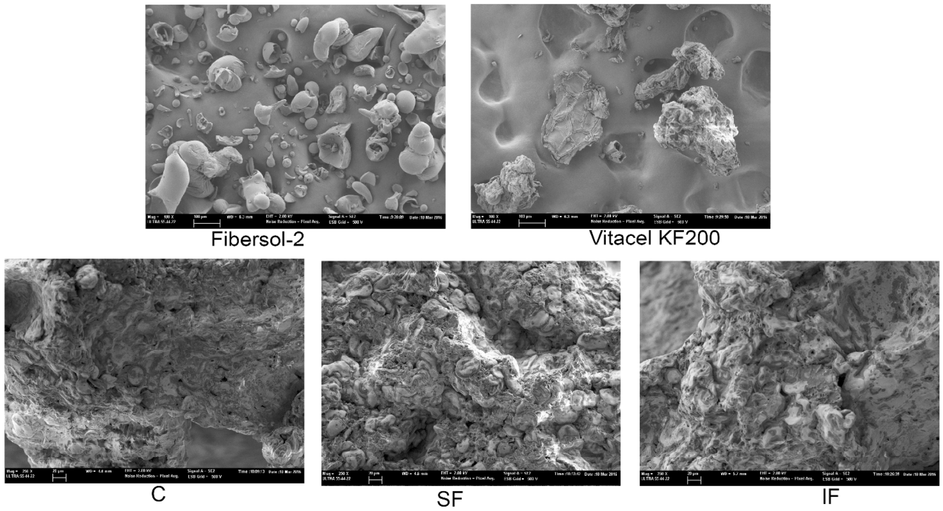

FIGURE 3. Field emission scanning electron microscopy (FESEM). Images of soluble fiber (Fibersol-2) and insoluble fiber (Vitacel KF200), magnification 100x, bar = 100 µm. Images of cakes C, SF and IF, magnification 250x, bar = 20 µm. pc: cell walls, vc: transport tissues. C: control cake; SF: sponge cake with soluble fiber; IF: sponge cake with insoluble fiber.

399x299mm (300 x 300 DPI)

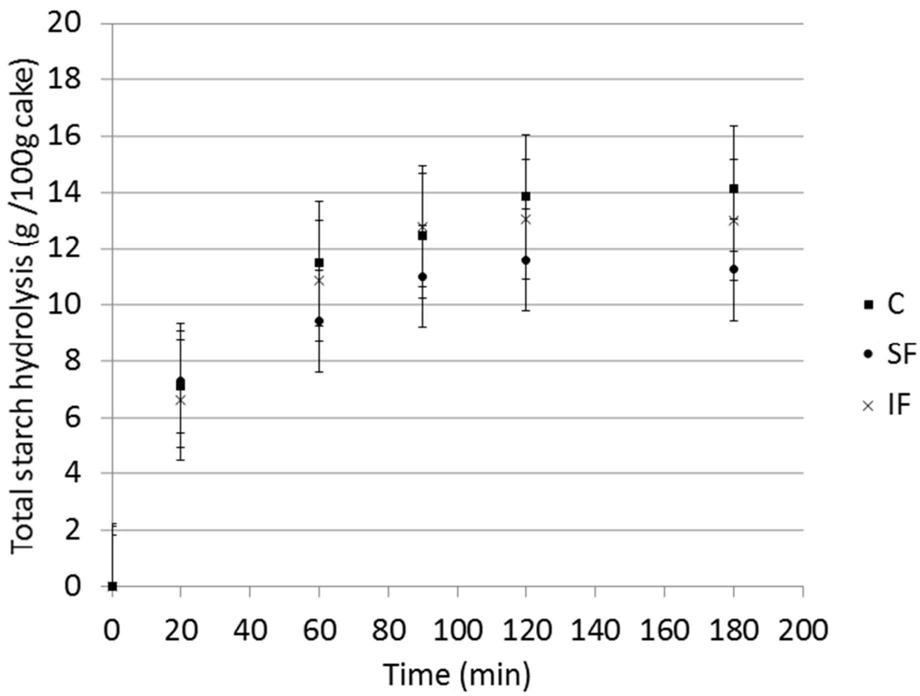

FIGURE 4. In vitro digestibility of starch of the cakes C, SF, M and IF. C: control cake; SF: sponge cake with soluble fiber; M: sponge cake with a mixture of soluble and insoluble fiber; FI: sponge cake with insoluble fiber.

210x170mm (96 x 96 DPI)

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PAPER TITLE	MANUSCRIPT ID NUMBER
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Elena Diez-Sánchez has made a substantial contribution to the design, acquisition and interpretation of data in the sections of the manuscript corresponding to digestion in vitro and sensory studies. She revised the manuscript and approved the version to be published.

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