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# CONTENTS

<b>Industrial Process design for Microwave Pasteurization of Liquid Foods: a Computational Study</b> Huseyin Topcam and Ferruh Erdogan .....	166
<b>A Coupled CFD-Heat Transfer Model for In-Package Solid Food Pasteurization</b> Clarissa Detomi de Albuquerque, Sébastien Curet and Lionel Boillereaux.....	172
<b>Numerical Modelling of Airflow and Heat Transfer in a Vented Pallet of Cheese</b> Anh Thu Pham, Jean Moureh and Denis Flick.....	177
<b>Edible Oil Hydrogenation: Revisited Viscosity Prediction for Accurate Process Simulation</b> Pierre Albrand, Anne-Marie Billet, Carine Julcour and Vincent Gerbaud .....	181
<b>Optimal Control of Fructo-Oligosaccharide Production</b> J. Schorsch, M. Kinnaert, R. Fekih-Salem, L. Dewasme, C.C. Castro and A. Vande Wouwer .....	184
<b>Modeling and Simulation in Innovative Food Production and Processing Technologies</b>	
<b>Modelling and Validation of Time Temperature History and Enzyme Inactivation in the Continuous Flow Microwave assisted Pasteurization of Apple Juice</b> Érica S. Siguemoto, Carmen C. Tadini and Jorge A. W. Gut.....	189
<b>A Long-Short-Term Memory Network Model for Biscuit Baking</b> Alberto Tonda and Nathalie Perrot.....	194
<b>On the Quantification of the Impact of Natural Antimicrobials on the Growth Kinetics of <i>Listeria</i> in Complex Food Models</b> Katherine Costello, Madeleine Bussemaker, Eirini Velliou, Jorge Gutierrez-Merino, Maria Baka and Jan Van Impe.....	199
<b>Effect of Xhantan Gum on Physicochemical and Textural Properties of Gluten-Free Batter and Bread</b> Christian R. Encina-Zelada, José A. Teixeira, Fernando Monteiro, Ursula Gonzales-Barron and Vasco Cadavez.....	204
<b>Creating, Refining and Validating a Model Describing Spatiotemporal Dynamics in Cheese during Ripening</b> Thorsten Stefan .....	210

# EFFECT OF XANTHAN GUM ON PHYSICOCHEMICAL AND TEXTURAL PROPERTIES OF GLUTEN-FREE BATTER AND BREAD

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## KEYWORDS

Quinoa flour, Rheology, Texture profile analysis, Crumb image analysis

## ABSTRACT

The objective of this study was to evaluate the effect of xanthan gum (XG) on physicochemical, rheological and textural properties of gluten-free batter and bread. Batches of gluten-free bread used a base formulation of rice, corn and quinoa flour, and different levels of XG (1.5, 2.5 and 3.5%) and water content (WC; 90, 100 and 110%) in a full factorial design. Although in interaction with water, higher XG doses tended to produce batters of lower stickiness, work of adhesion, strength-cohesiveness; which, when baked, produced loaves of lower specific volume and baking loss; and bread crumb of lower  $A_w$ , pH, hardness, springiness, firmness, mean cell area, void fraction, mean cell aspect ratio, and higher firmness, consistency, cohesiveness, adhesiveness, chewiness, resilience, cell density, cell size uniformity and mean cell compactness ( $p < .001$ ). Gluten-free loaves of good appearance in terms of high specific volume, low crumb hardness, high crumb springiness, and open grain visual texture were obtained in formulations with 110% WC and XG doses between 1.5-2.5%.

## INTRODUCTION

To target the 0.5–1.0% of the world's population estimated to be affected by coeliac auto-immune disease (Gujral et al., 2012), a number of food products that avoid the use of wheat (gliadin), rye (secalins), barley (hordein) and oats (avidins) have been lately developed. There has been particularly an upsurge in the development of gluten-free bread.

Xanthan gum (XG), a high molecular weight polysaccharide produced by the bacterium *Xanthomonas*

*campestris*, is largely used in gluten-free breadmaking, because it can hydrate in cold water and produce a viscous solution with strong shear thinning flow behaviour. The xanthan molecule has a cellulosic backbone with side chains that wraps around the backbone and make it rigid, enabling its emulsifying and foaming properties (Naji and Razavi, 2014). In low amounts, XG has been found to increase loaf volume, and improve bread rheological and sensory properties (Hager and Arendt, 2013). Optimising textural and sensory attributes, researchers have recommended doses of XG in gluten-free bread, that, in general, range between 0.5% up to 7.0% (flour weight), yet this is strongly linked to bread formulation and water content. The objectives of this study were: (i) to assess the effect of both xanthan and water on the rheological properties of batter, and the physicochemical and textural properties of bread formulated with a mixture of rice, corn and quinoa flours; and (ii) to get an insight into the relationships among all the quality attributes measured.

## MATERIALS AND METHODS

### Bread elaboration

The effects of GG (three doses tested) and water content (WC; three levels tested) were evaluated using a full factorial design. Thus, nine batches of gluten-free bread, with two replicates, were produced using a base formulation of rice flour (50%), corn flour (50%) and quinoa flour (20%), sunflower oil (6% flour weight), white sugar (3% flour weight), refined salt (1.5% flour weight), instant yeast (3% flour weight), GG (1.5, 2.5 or 3.5% flour weight) and water (90, 100 or 110% flour weight). To make the batters, XG was purchased from TecPan (Portugal), while the other ingredients were purchased from a local supermarket. Demineralised water (pH=6.8) kept at 5°C overnight was used. All ingredients were mixed for 6 min in a professional food processor (SilverCrest SKMP-1200, Germany) equipped with a batter blade. Two hundred and eighty grams were then poured into in oiled and floured square

tins, and allowed to proof at 30°C and 85% of relative humidity for 60 min in a climatic chamber (Climacell 222, Germany). Afterwards, all moulds from the same batch were placed in a pre-heated convection oven (Princess, 2000 W, The Netherlands) for 60 min at 190°C. Bread loaves were un-moulded when reaching ambient temperature, and all analyses were performed after 24 h. The rheology of batter and bread were characterised using a texture analyser TA-XT plus (Stable Micro Systems, UK) fitted with specific fixtures, namely, the SMS/Chen-Hoseney stickiness rig for dough stickiness analysis; the 35 mm-diameter perspex flat rig, and a standard size back extrusion cylindrical container (50 mm diameter, capacity of 115 g approx.) for the back-extrusion analysis; and a 36 mm-diameter aluminium probe (model P/36R) for the texture profile analysis.

### Batter rheology

Several batter rheological properties were studied, including: 1) stickiness analysis (Agrahar-Murugkar et al., 2015), which produced measurements of: stickiness (STIba, g), work of adhesion (ADHba, g.s), and strength-cohesiveness (SCOba, mm); and 2) back extrusion analysis (Juszczak et al., 2012), which rendered measurements of: firmness (FIRba, g), consistency (CONba, g.s), cohesiveness (COHba, g) and viscosity index (VISba, g.s).

### Bread crumb quality properties

The bread loaf specific volume (SVObr, ml/g) was calculated as loaf volume divided by loaf weight 24 h after baking, while the baking loss (BLObr, %) was computed as [initial loaf weight before baking - the loaf weight after 24 h baking x 100] / initial loaf weight before baking. Such physical properties were measured in triplicate. Water activity (Aw) and pH of bread crumb were quantified in quintuplicate as in Machado-Alencar et al. (2015) and Hashemi et al. (2016), respectively. Texture profile analysis (TPA) (Martinez and Gomez, 2017) was employed to obtain the bread crumb rheological properties of hardness (HARbr, g), adhesiveness (ADHbr, g.s), springiness (SPRbr, mm), cohesiveness (COHbr, g), chewiness (CHEbr, g.mm), resilience (RESbr, dimensionless) and firmness (FIRbr, g).

### Bread crumb image analysis

Slices of bread were scanned (Canon Pixma MG-2550, Vietnam) with -10% brightness and +15% contrast with a resolution of 350 dpi, and a 4.0 cm x 4.0 cm field-of-view from the centre of the image was cropped and saved in png format for posterior analysis. Several grain crumb features were computed using the automated thresholding technique proposed in Gonzales-Barron and Butler (2006) and Gonzales-Barron and Butler (2008), coded in Matlab software (ver. R2015a, The Mathworks, USA). These were: mean cell area (MCA, mm<sup>2</sup>); cell density (CD, number of cells/mm<sup>2</sup>); uniformity (UNI, dimensionless), calculated as the rate between the number of cells <=5 mm<sup>2</sup> and number of cells > 5 mm<sup>2</sup>; void fraction (VFR, dimensionless), calculated as the proportion of the space occupied by the pores/cells; mean cell compactness (COM, dimensionless), with compactness defined as the ratio of the area of the cell to the area of a circle having the same perimeter; and mean

cell aspect ratio (ARA, dimensionless), with aspect ratio defined as the ratio of the major axis to the minor axis of a cell.

### Statistical analysis

Data were analysed using the R software version 3.3.1 (R Core Team, 2017). Analyses of variance were applied to assess the effect of XG and WC on the response variables: pH, Aw, BLObr, batter stickiness and batter extrusion. On the other hand, for the response variables: specific volume, TPA, firmness and features of bread crumb acquired by image analysis, a linear mixed model was used assuming that the measurements taken from the same loaf were clustered. Statistical analyses were conducted using the packages “plyr”, “ggplot2”, “lme4” and “lmerTest” for the linear models; and the packages: “rmisc”, “rcomdmisc”, “plyr”, “ggplot2”, “car”, “multcompView” and “lsmeans” for the linear mixed models.

## RESULTS AND DISCUSSION

### Batter properties

Both XG and WC affected the rheology of gluten-free batter, as per the results of the batter stickiness analysis (Figure 1) and the back-extrusion analysis (Figure 2). The batter stickiness properties of STIba, ADHba and SCOba, ranged from 28.81 to 55.90 g, from 3.19 to 4.57 g□s and from 2.42 to 3.67 mm, respectively. At a constant XG level, higher WC consistently (p<.001) increased the STIba, ADHba and SCOba measures, while at a constant WC level, higher XG contents consistently (p<.001) decreased those stickiness measures (Figure 1). This occurs because xanthan gum has the capacity to bind large amounts of water into the dough matrix.

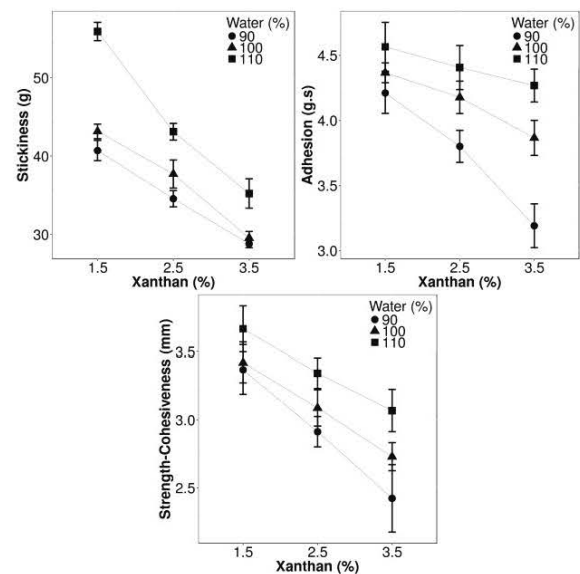


Figure 1: Effect of Xanthan Gum and Water Content on the Gluten-Free Batter Stickiness Properties of STIba (top left), ADHba (top right) and SCOba (bottom)

The back extrusion analysis values ranged for FIRba from 359.08 to 1990.04 g, for CONba from 3494.17 to 17363.37

g□s, for COHba from -260.59 to -1098.89 g and for VISba from -1249.64 to -4691.52 g□s. Higher XG contents increased ( $p<.001$ ) FIRba and CONba, and decreased ( $p<.001$ ) COHba and VISba while higher WC levels had an opposite effect, reducing ( $p<.001$ ) FIRba and CONba, and increasing ( $p<.001$ ) COHba and VISba (Figure 2). Using the same level of water (110%), Sciarini et al. (2010) obtained lower batter firmness values, from 50.8 to 1252 g, for a mixture of corn/soy (90:10) and rice/soy (80:20), respectively. They concluded that rice/soy mixtures required higher force to extrude, because soy proteins have the ability to absorb cold water, resulting in a decrease of free water in the batter mixture. In our case, quinoa proteins may have lent a greater cold water absorption capability, hence producing batters of higher firmness than those with soy proteins.

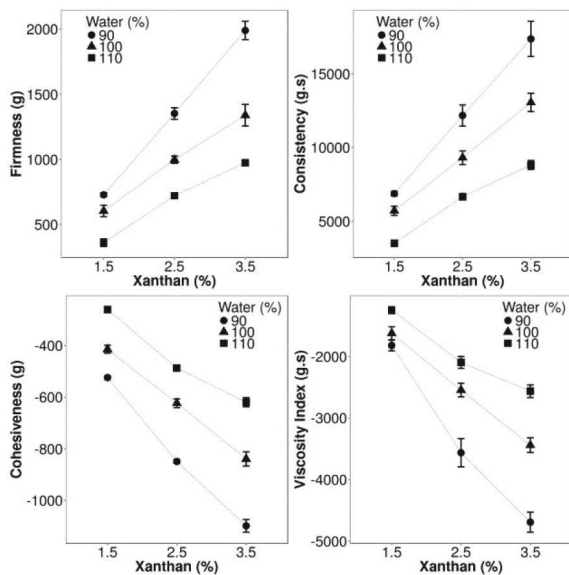


Figure 2: Effect of Xanthan Gum and Water Content on the Gluten-Free Batter Back-Extrusion Properties of FIRba (top left), CONba (top right), COHba (bottom left) and VISba (bottom right)

Compared between 1.5 and 2.5% of xanthan in a GF fresh filled pasta batter, Sanguinetti et al. (2015) observed that a higher dose of XG resulted in a more cohesive, less adhesive and more elastic batter. Sabanis and Tzia (2010) and Turkut et al. (2016) reported that higher consistency values and viscosity index in their GB batters led to lower specific volume. This finding was corroborated in the present study, where 3.5% XG doses produced loaves of lower specific volume (1.69 ml/g) compared to those obtained from treatments with 1.5% xanthan (1.78 ml/g) (Figure 3).

#### Bread crumb physicochemical properties

Moisture loss during baking ranged from 11.9 to 14.5%, increasing ( $p<.001$ ) with higher WC, and decreasing ( $p<.001$ ) with higher XG content (Figure 4). However, although higher XG amounts reduce baking loss, it can negatively affect the volume of loaves (Figure 4). The specific volume of bread ranged from 1.58 to 1.91%, and

decreased ( $p<.001$ ) with higher XG content, and increased ( $p<.001$ ) with higher WC.

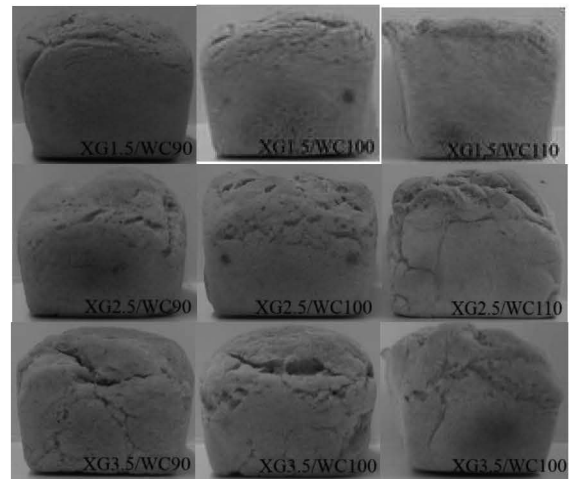


Figure 3: Photographs of Gluten-Free Bread Loaves Produced with Varying Xanthan Gum (XG) and Water Content (WC) Showing Height and Crust Appearance

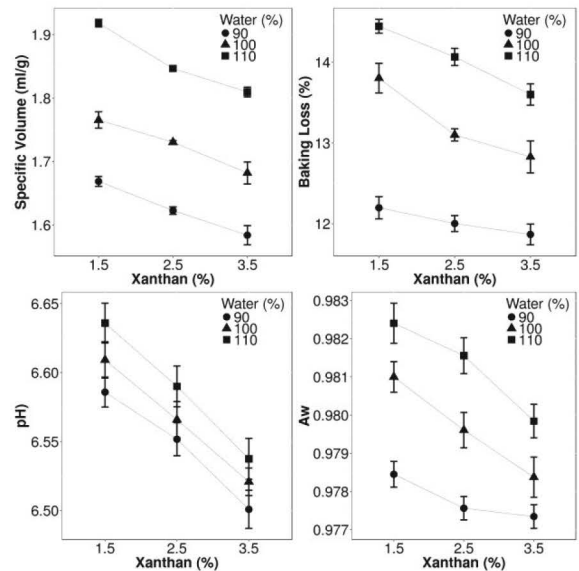


Figure 4: Effect of Xanthan Gum and Water Content on the Gluten-Free Bread Physicochemical Properties of SVObr (top left), BLObr (top right), pH (bottom left) and Aw (bottom right)

The positive impact of high WC was readily evident in the specific volume of breads, as explained by de la Hera et al. (2014), due to the plasticizer effect of the water which contributes to the extensional properties of the batter during mixing allowing the hydration of the particles. Nonetheless, Han et al. (2012) reported that excessive water can cause overexpansion during baking resulting in large volume breads or collapsed loaves. Onyango et al. (2011) explained that, as gas leaks out of the bubbles, it forces its way through the weakly connected particles and channels formed by gas pressing the particles apart. Since, in our experiments, small-sized bread loaves were mostly associated to lower baking losses; we can conclude that,

regardless of the XG dose, batters with low WC tended to proof insufficiently, resulting in bread loaves of lower volume. In relation to the other physicochemical properties, bread acidity was also influenced by the addition of XG ( $p < .001$ ) and WC ( $p < .001$ ), with more acidic crumbs produced by higher doses of XG. As expected, the amount of free water in the crumb decreased with the addition of higher concentrations of XG ( $p < .001$ ) and lower WC amounts ( $p < .001$ ).

### Bread crumb textural properties

Having produced firmer and less viscous batters, lower WC levels consequently yielded tougher breads (Figure 5). According to the TPA, loaves with the lowest WC content (90%) were significantly harder (5130 g), less cohesive (0.477) and less springy (0.833) than the treatments with 100 and 110% WC. The addition of higher amounts of water can improve crumb texture, since the 110% WC treatments produced softer breads, with lower values of HARbr (1995 g) and CHEbr (911 g·mm), ADHbr (-12.80 g·s – in absolute value), and higher values of SPRbr (0.884 mm), COHbr (0.517) and RESbr (0.217), compared to those obtained from the lower WC treatments of 90 and 100%. Although, to a lower extent than WC, XG also had an impact on bread crumb texture.

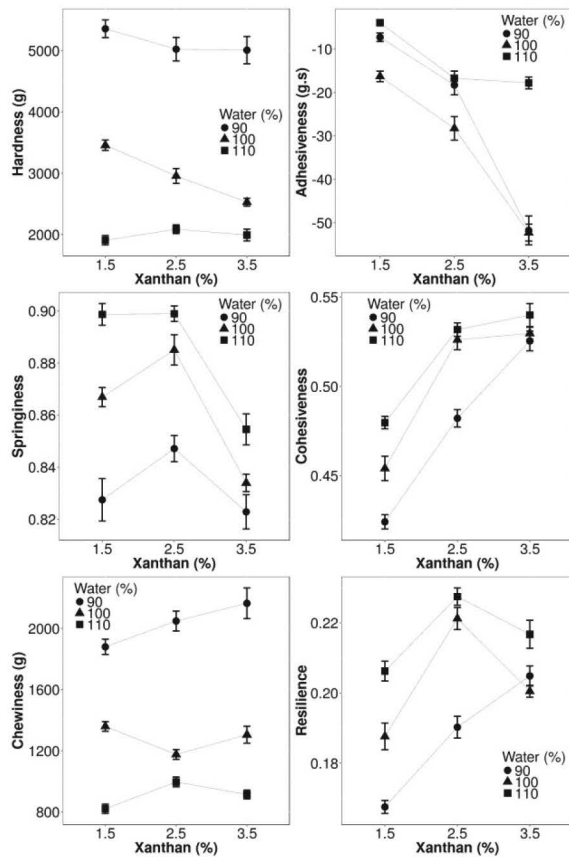


Figure 5: Effect of Xanthan Gum and Water Content on the Gluten-Free Bread Instrumental Textural Properties of HARbr (top left), ADHbr (top right), SPRbr (middle left), COHbr (middle right), CHEbr (bottom left) and RESbr (bottom right)

Loaves formulated with the lowest XG dose (1.5%) produced crumb with higher values of HARbr (3574 g) and SPRbr (0.864 mm), and lower values of ADHbr (-9.14 g·s – in absolute value), COHbr (0.453), CHEbr (1353 g·mm) and RESbr (i.e., 0.187), compared to those obtained from 2.5 and 3.5% XG. Apart from hardness, resilience and springiness are important quality properties as they characterise crumb elasticity or the ability of the material to return to its shape after stress (Onyango et al., 2011), which is a desired attribute empirically assessed by the consumer. In our results, RESbr and SPRbr were found to be linked, having both a quadratic trend with XG dose, and optimum (higher) values were around 2.5% XG. In addition, bread of these desired properties were obtained at the highest WC of 110%. Apart from the separate effects of XG and WC, statistical analysis evidenced interactions ( $p < .001$ ) between XG and WC on all of TPA measurements, being the strongest for resilience.

### Image analysis features of crumb grain

Digital images (Figure 6) of gluten-free bread crumb grains, showed visual differences among the nine formulations. While higher WC formulations produced more open grain textures, lower WC formulations produced the opposite, closer crumbs of smaller pores. Such differences were statistically corroborated in all of the image grain features analysed. Values were in the range of 0.73 – 1.89 mm<sup>2</sup> for MCA; 0.200 – 0.319 cells/mm<sup>2</sup> for CDE; 10.54 – 63.09 for UNI; 0.217 – 0.339 for VF; 0.755 – 0.776 for COM; and 1.666 – 1.770 for ARA.

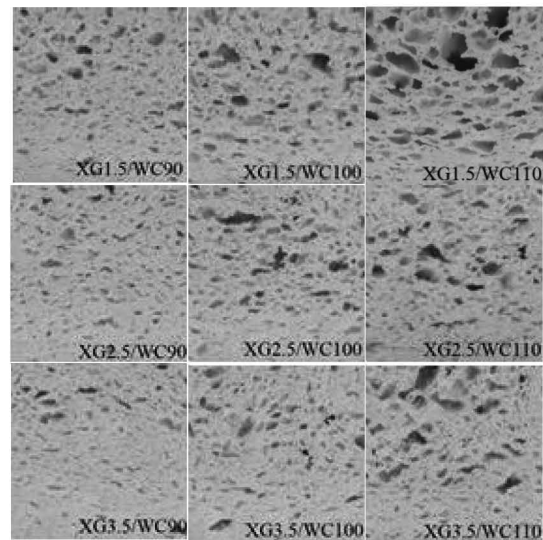


Figure 6: Crumb Grain of Gluten-Free Bread Produced by Varying Xanthan Gum (XG) and Water Content (WC)

According to Figure 7, when XG increased, MCA, VF and AR values decreased ( $p < .001$ ) while CDE, UNI and COM increased ( $p < .001$ ). This means that at a constant level of water in the formulation, increasing XG doses only produces smaller bread loaves. These smaller loaves tend to have a compact visual texture, formed of a greater number of cells, but of smaller size. Because of this, the cell size uniformity is greater but the void fraction is lower. Smaller



loaves of denser of more compact texture also have the characteristic of having more rounded cells (i.e., higher cell compactness) and less elongated cells (i.e., lower aspect ratio). Loaves of lower XG contents (1.5%) presented crumbs with larger pores, with higher values of MCA (1.35 mm<sup>2</sup>), VF (0.29) and AR (1.75), and lower values of CD (0.25 cells/mm<sup>2</sup>), UNI (20.5) and COM (0.76) compared to those obtained with higher XG levels of 2.5 and 3.5%.

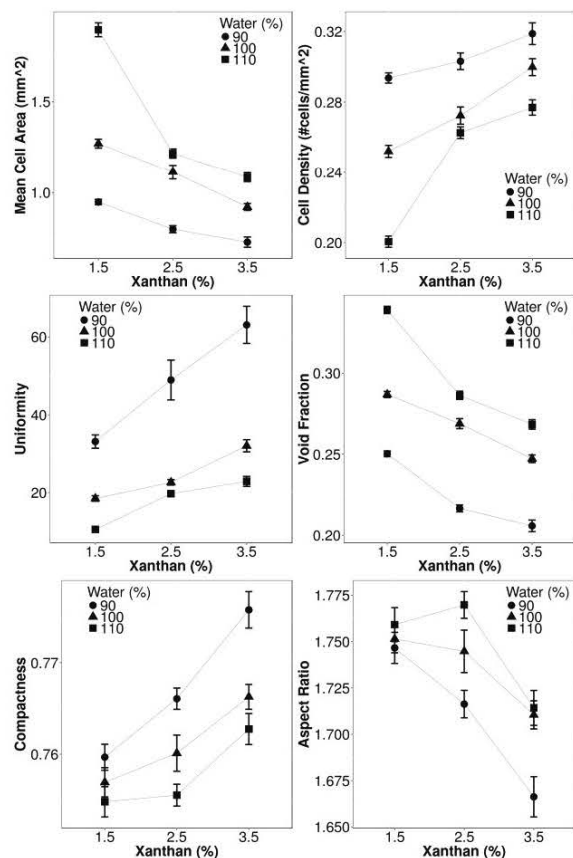


Figure 7: Effect of Xanthan Gum and Water Content on the Gluten-Free Bread Crumb Grain Features of MCA (top left), CD (top right), UNI (middle left), VFR (middle right), COM (bottom left) and ARA (bottom right)

On the other hand, when higher proportions of WC were added, the opposite was observed; this is, values of CD (0.25 cells/mm<sup>2</sup>), UNI (17.5) and COM (0.758) were lower ( $p < 0.001$ ), while values of MCA (1.38 mm<sup>2</sup>), VF (0.30) and AR (1.75) were higher when breads were formulated with 110% WC, in comparison with those obtained with the lower WC treatments of 90 and 100%. This signifies that, when loaves undergo a better proofing, expanding more during fermentation and baking, facilitated by the greater amount of water, the final visual texture of the bread crumb has altogether a different crumb grain. As breads were formulated with a constant XG dose and increasing WC levels, crumb grains appeared more open; in other words, crumb made of cells of greater size and less compact and more elongated shape. This in turn leads to a less uniform grain (since the number of large cells is greater) and a higher void fraction.

A denser bread crumb grain can also be effectively evaluated by the measurements of cell size uniformity and mean cell compactness or aspect ratio. Notice that higher values of UNI and COM were obtained with higher amounts of XG (3.5%) and lower values of WC (90%), corresponding in both cases to a denser structure (Figure 7). Thus, when during proofing and baking, batter expands more and steadily, a greater number of large cells is produced, therefore bringing down the ratio small-to-large cells (UNI), while due to coalescence, the large cells tend to be more elongated and less compact, thereby bringing down the values of COM.

From the nine formulations, the more open crumb grain was attained by the formulation with 1.5% XG and 110% WC (2.25 mm<sup>2</sup>; Figure 7), whereas the formulation with 3.5% XG and 90% WC produced the smallest mean cell size (0.56 mm<sup>2</sup>), characterising the denser structure obtained, which was also reflected by the lowest specific volume of this formulation (1.59 ml/g; Figure 3). For de la Hera et al. (2014), if gluten-free breads are elaborated with excessive water, large holes can appear in the crumb, as was also attested in our experiments for the formulation XG1.5/WC110 (Figure 6). In this study, using XG as the only batter thickener, it was possible to obtain crumbs with open grain structures at a high level of water (110%) and a low level of XG (1.5%). The presence of larger cells can also be linked to a spongier crumb structure, which is a desirable quality property yet not typically found in gluten-free breads. Similar to our findings were those reported by Schober et al. (2005) who encountered that sorghum bread with a fine crumb structure was tougher than bread with a coarse and open crumb structure.

#### 4. CONCLUSION

Higher amounts of xanthan gum, in gluten-free bread formulated with a mixture of rice, maize and quinoa flours, have in principle the capacity to retain more water; however, at a constant water level, higher doses of xanthan gum produce less viscous and sticky batters but of increased firmness, that translates into baked loaves of smaller volume with a more cohesive and less springy crumb texture. The highest water content of 110% and XG between 1.5 and 2.5% produced loaves of good quality in terms of high specific volume, low hardness, high springiness, low cell density, low cell size uniformity, high void fraction and high mean cell area.

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