

## Dynamic rheology and microstructure of starch gels affected by triticale genomic composition and developing stage

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**Abstract.** Starches of developing triticale grains, differing in genome composition (complete AABBRR or substituted AABBDR), were evaluated in terms of starch granule distribution, dynamic rheological behaviour and microstructural characteristics on several days after anthesis. The starch granules were of an oblate spheroid shape for A-granules, and of a spherical shape for B-granules. However, those obtained from the complete triticale showed a larger diameter size. An X-ray diffraction analysis revealed the common A-type pattern of cereal starches from early development stages. A dynamic rheological analysis showed that the storage and loss moduli reached maximum levels in the temperature range of 71–86°C and dropped at around 90°C. Starches from the complete triticale showed lower phase transition temperatures, compared to those obtained from the substituted genotype (56.1±0.3 and 60.3±0.8°C, respectively). Scanning electron microscopy showed that the gels made with the starch of complete triticales were of a less dense sponge-like structure.

**Keywords:** triticale developing grains, starch, gels, rheology, microstructure

### INTRODUCTION

Starch is the major polysaccharide and carbon reserve of plants (Hizukuri *et al.*, 1981). Starch in the cereal endosperm is synthesized and accumulated to enhance plant and grain development. However, it is the grain-filling period that determines the final weight and quality of mature grains (Zhao *et al.*, 2003).

The major components of starch are glucose polymers, amylose and amylopectin. Amylose is a linear oligosaccharide made of D-glucose units bonded to each other through  $\alpha$ -1,4-glycosidic bonds with a degree of polymerization (DP) in the range of 500–6000 glucose residues. Amylopectin is a very large and highly branched chain molecule with a DP ranging from  $3 \times 10^5$  to  $3 \times 10^6$  glucose units, and it consists of  $\alpha$ -1,6-linked D-glucose units attached to  $\alpha$ -(1,4)- bonds (Jeon *et al.*, 2010).

Changes in the glucan chain length distribution or the degree of crystallinity can alter starch physico-chemical characteristics (Copeland *et al.*, 2009). In the starch granule, amylose and amylopectin are arranged in the alternating amorphous and crystalline regions, respectively, forming what is known as the growth rings. Amylose molecules associated with large branches of amylopectin comprise the amorphous region of granules whereas short branches of amylopectin comprise the crystalline region, as a result of which a higher proportion of amylopectin in starch granules results in a higher crystallinity degree (Cheetham and Tao, 1998). Amylose exhibits the most useful functions as a hydrocolloid which forms gels and films. In contrast, amylopectin with long chains interferes with the interaction of amylose chains, preventing retrogradation, and leads to viscosity loss (Chung and Liu, 2009).

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Starch in cereal grains is synthesized in amyloplasts by four types of enzymes, *i.e.* ADP-glucose pyrophosphorylase, starch synthase, as well as branching and debranching enzymes (Cao *et al.*, 2012). The action of these enzymes is influenced by cultivar and environmental conditions, which in turn affects the amount and size of the synthesized starch granules as well as the chain-length (Ulbrich *et al.*, 2015), the amylose: amylopectin ratio (Altenbach *et al.*, 2003; Cornejo-Ramírez *et al.*, 2016; Jiamjariyatam *et al.*, 2015) and the fine structure of amylopectin (Jane *et al.*, 1999). All these properties influence the functional properties of starches, thus contributing to the final quality of food and non-food products. However, there is a continuous demand for new cultivars with improved agronomic characteristics, such as better yield and kernel size, improved nutritional properties and optimal performance under extreme environmental conditions. While the breeding of new and better cultivars through genetic crosses is required to overcome their demand, it is important to ensure that the expected improved quality has been reached through agronomic and rheological performance tests.

Triticale (*X Triticosecale* Wittmack) is a hybrid cereal crop created from a cross between durum wheat (*Triticum durum*) and rye (*Secale cereale*). Triticale varieties have been developed as complete (with genomic constitutions AABBRR and AABBDDRR) or substituted (with genomic constitutions AABDDR and AABDDDDR). The first substituted triticale, called Armadillo, was obtained as a result of a spontaneous crossing of triticale with an unknown Mexican semi-dwarf bread wheat. Taking advantage of the highly heritable good agronomic characteristics resulting from the fortuitous crossing, Armadillo materials were distributed by CIMMYT among plant breeders from all over the world, in order to be used for providing high fertility, better hectolitre weights and grain yield, insensitivity to photo-perforation, dwarfism, early maturity and good nutritional quality to the less promising triticales (Varughese *et al.*, 1987). With further genetic improvements and research, both types of triticales have shown phenotypes and agronomic differences. Complete triticales are more adaptable and resistant to limiting conditions, showing higher productivity, both in fertile areas and in adverse conditions. However, substituted triticales have given rise to more varieties, since they are more stable and of better bakery quality (Mellado *et al.*, 2008).

However, few studies have been conducted to compare and contrast the differences in certain physicochemical properties of starches among different triticale genotypes during grain development. To our knowledge, no research has been carried out that would focus on the dynamic rheological properties of starches affected by triticale genomic composition and grain developing stage. The aim of this research was to determine and compare the dynamic rheological and microstructural gel-forming properties of starches isolated from developing grains of two triticale

genotypes on different days after anthesis. The results might yield further insights into whether immature and mature, complete and substituted, triticale grains could be best suited for commercial production.

## MATERIALS AND METHODS

Seed samples of a complete triticale (Eronga variety, AABBRR genome) and a substituted triticale (Yoreme variety, AABDDR genome) were employed in this study. Grain samples of the two genotypes of triticale were planted during the 2012-2013 planting cycle at the same time and under the same environmental conditions, at the agriculture experiment station of the Agriculture and Livestock Department of the University of Sonora, Mexico (208 m above the sea level, with an average annual temperature of 24.2°C and an annual average rainfall of 280 mm). A completely randomized complete block design with three replications (plots) for each triticale genotype was used.

Triticale spike samples were collected on selected days after anthesis (DAA). Eight spikes were cut from plants within each plot and shelled manually. Only developing and mature grains from the middle part of the spikes were collected and stored at -70 °C until analysis.

Starch of triticale samples collected on different DAA was obtained from single kernels using the method proposed by Bettge *et al.* (1995) with modifications. Briefly speaking, the embryo and bran of the kernels were manually removed with a razor blade; the remaining parts of the kernels were lightly crushed between two sheets of wax paper and transferred to a standard 1.5 ml Eppendorf polypropylene microcentrifuge tube. To prevent lumping, the solids were steeped twice with 500 µl of petroleum ether for 30 min at room temperature with occasional gentle agitation. After centrifugation at 10,000 g for 2 min and removal of the solvent, the precipitate was steeped twice in 500 µl of 100 mmol l<sup>-1</sup> NaCl at room temperature for 30 min with occasional gentle agitation. Gluten was formed by kneading the endosperm inside the microcentrifuge tube, with the use of a Teflon micro-spatula, and then manually removed. The starch was washed twice with 500 µl of deionised water to remove excess of salt, following which it was centrifuged at 10,000 g for 2 min, re-suspended in 1 ml of acetone and dried at ambient temperature.

The size distribution of defatted starch granules was measured using laser diffraction in a Coulter LS 100Q (Beckman Coulter, Miami, FL, USA). Big starch granules (> 6 µm) are commonly referred to as A-type starch granules, whereas the small ones (≤ 6 µm) are known as B-type starch granules. For analysis, a small quantity (20-30 mg) of starch was vortexed in 2 ml of deionised water and pumped through the optical chamber. A GB500 standard, consisting of a glass bead sample (500 µm nominal mean particle size), was used as a reference.

The crystallinity percent of triticale starches was determined using the X-ray diffraction methodology proposed by Song and Jane (2000). Starch samples were equilibrated in a 100% relative humidity chamber for 24 h at 27°C. X-ray diffraction patterns (XRD) were obtained with nickel foil-filtered Cu K $\alpha$  radiation, using a Rigaku RINT-2000 diffractometer (Tokyo, Japan), at 27 mA and 50 kV. The scanning region or the two-theta angle ( $2\theta$ ) ranged from 5 to 50° with a 0.05° step size and a count time of 2 s. The crystalline percent values were calculated according to Hayakawa *et al.* (1997), by means of the following equation: Crystallinity percent =  $Ac / (Ac + Aa) \times 100$ , where  $Ac$  and  $Aa$  correspond to the crystallinity and amorphous areas, respectively, on the X-ray diffractograms.

Rheology is a convenient tool to study starch gelation since a physical gel is formed during starch gelatinization. The gelatinization process can be analyzed by monitoring the rheological properties using a rheometer. The gelatinization properties of the starches isolated from the developing grains of the two triticale genotypes were studied using a stress-controlled Discovery Hybrid Rheometer HR-2 (TA Instruments, New Castle, DE, USA) equipped with two parallel plates (Peltier Plate Steel-104556) with 40 mm in diameter. A volume of 1.5 ml of 10% starch-water suspensions was routinely placed on the centre of the bottom plate while the upper plate was immediately lowered until the final gap of 1 mm was reached. The edges of the plates were covered with paraffin oil in order to prevent dehydration of the samples by evaporation. Starch samples were examined in the angular frequency sweep scan range of 0.01-60 Hz at a constant strain of 1%. The start temperature was 25°C, and a ramp temperature of 5°C min<sup>-1</sup> was used until the final temperature of 90°C was reached. After 30 s of releasing the stress, the samples were cooled to 25°C at a cooling rate of 5°C min<sup>-1</sup>. After 300 s of soaking time, the gels were frozen at -20 °C, lyophilized and stored for further analysis. The rheological parameters elastic modulus ( $G'$ ) and viscous modulus ( $G''$ ) were obtained at the frequency sweep range indicated above. The phase transition temperature  $T_0$ , *i.e.* the crossover where the  $G'$  and  $G''$  values started to increase at the same temperature and representing the sol-to-gel transition, was determined in the dynamic modulus spectrum ( $G'$  and  $G''$ ) during heating (Ulbrich *et al.*, 2015).

Scanning Electron Microscopy (SEM) was used to analyze the microstructure of the triticale starch gels using the method proposed by Jane *et al.* (1994). The lyophilized starch gels were placed on a 13 mm silver tape, metal-shadowed with gold/palladium (60/40) (Sputter coater SPI-Module; West Chester, PA, USA) and mounted on a brass disk. Starch samples were analyzed using a magnification of 1500 X under low vacuum, using a JEOL JSM-5400LV scanning electron microscope (Peabody, MA, USA), at an acceleration voltage of 15 kV.

All data were obtained from three independent experiments and expressed as mean  $\pm$  standard deviation of triplicates. Data were subjected to one-way analysis of variance (ANOVA) following general model procedures. A comparison of sample means was performed by means of the Tukey's test with the SAS program (SAS, 2005). In all cases, the mean values were considered significantly different at  $p \leq 0.05$ .

## RESULTS AND DISCUSSION

The granule size distribution of the starch strongly influences its physicochemical properties and functionality. In this study, the size distributions of starch granules of developing triticale grains were measured using laser diffraction. Table 1 shows the size distributions of starch granules of complete (Eronga) and substituted (Yoreme) triticale developing grains. Both triticale genotypes showed a bimodal starch granule distribution similar to that reported previously (Cornejo-Ramírez *et al.*, 2015) with oblate spheroid shape for A-granules and spherical shape for B-granules (Bechtel *et al.*, 1990). In general, A- and B-type starch granules of both triticale genotypes increased in size during grain development, with differences in size distribution values as well as in granules volume percent observed at all stages of development. However, A- and B-type starch granules of the complete triticale showed bigger sizes than those obtained from the substituted genotype throughout the course of the developing period of grains. The data also show that the A- and B-type starch granules of the complete triticale grew faster and reached larger diameters than the granules of the substituted triticale. A similar behaviour was observed in wheat starch granules during development (Bechtel *et al.*, 1990; Kim *et al.*, 2003).

Starch molecules arrange themselves in the grain in crystalline granules. Amylose molecules comprise the amorphous region of granules, whereas amylopectin molecules with short branches comprise the crystalline region. Crystallinity of starch granules has been directly associated with amylopectin concentration (Cheetham and Tao, 1998) and inversely related to onset temperature ( $T_0$ ), peak temperature ( $T_p$ ) and enthalpy ( $\Delta H$ ) (Kim *et al.*, 2003). There are three types of crystalline structures, *i.e.* A-type found in cereal starches, B-type found in tubers and C-type present in legumes (Singh *et al.*, 2003). In our study, the starches presented the typical A-type X-ray diffraction pattern (Fig. 1) observed in cereal starches (Kim *et al.*, 2015). The diffractograms of triticale starches showed peaks at 15, 17.1, 18 and 23° from the early stage until their maturity.

The starch samples displayed the same X-ray diffraction pattern because their chemical composition and physicochemical properties are similar. Also, intensities of each peak of the starches isolated from developing grains on 16, 22 and 31 DAA are similar, whereas the intensities of the peaks of these starch samples are different from those obtained from mature grains. This suggests that the

**Table 1.** Starch granule size distribution and volume (%) of A- and B-type starch granules obtained from complete (Eronga) and substituted (Yoreme) triticales during grain development on different days after anthesis (DAA)

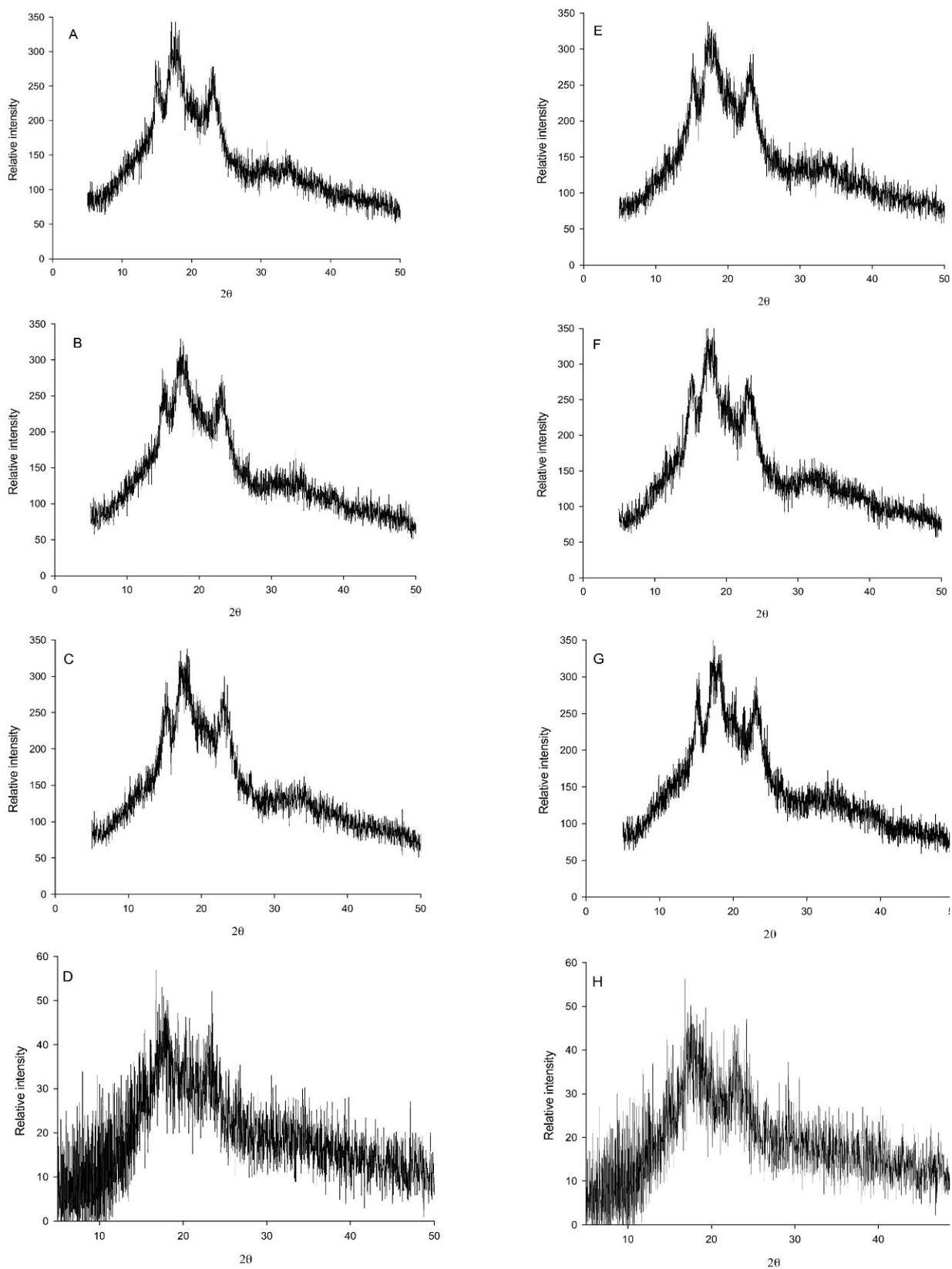
DAA	A-type		B-type	
	Size ( $\mu\text{m}$ )	Volume (%)	Size ( $\mu\text{m}$ )	Volume (%)
Complete triticale (Eronga)				
16	6.7-23	86.3 $\pm$ 0.06aB	0.96-3.7	13.7 $\pm$ 0.06bC
22	8.5-30	85.5 $\pm$ 0.20aC	0.98-4.5	14.5 $\pm$ 0.20bB
31	14-36	87.6 $\pm$ 0.20aA	1.5-5.2	12.4 $\pm$ 0.20bD
40	18-40	80.4 $\pm$ 0.03 <sup>b</sup> D	2.0-11.0	19.6 $\pm$ 0.03bA
Substituted triticale (Yoreme)				
16	6.7-20	86.7 $\pm$ 0.05aB	0.94-3.4	13.3 $\pm$ 0.05bC
22	9.0-24	88.0 $\pm$ 0.09aA	0.96-3.6	12.0 $\pm$ 0.09bD
31	11.2-33	86.3 $\pm$ 0.30aC	1.0-4.2	13.7 $\pm$ 0.30bB
40	12-38	74.6 $\pm$ 0.05aD	1.2-6.0	25.4 $\pm$ 0.05bA

Mean values are expressed in percentage  $\pm$  standard deviation. Values followed by the same lowercase letter in the same row and capital letter in the same column are not significantly different ( $p \leq 0.05$ ,  $n=3$ ).

amylopectin shapes of starches from mature grains are different from those of starches isolated from immature grains. Based on the results provided by the X-ray diffraction analysis of inorganic crystals, carried out by Inoue and Hirasawa (2013), the total area, and the longitude and latitude of crystals affect the X-ray peak intensities. As shown by Cornejo-Ramírez *et al.* (2016), reduction in the intensity of the X-ray diffraction patterns of triticale starches might be due to reduction in the volume of the A-type starch granules at the end of the maturation period. Additionally, the crystallinity of those starches increases due to a compaction of the amylopectin chains to form the crystalline areas.

A comparison of the crystallinity values observed along the whole developing period showed that the complete and substituted triticale starches were similar, reaching final values of 36.1% on 40 DAA (Table 2). Furthermore, an analysis of the crystallinity values of each triticale throughout the developing period showed that the complete triticale did not exhibit significant differences in crystallinity from 16 to 31 DAA, but on 40 DAA displayed a value significantly higher than those observed at early stages. In contrast, the starch granules of the substituted triticale showed continuous changes in crystallinity until the end of the maturity period. The final values in crystallinity of the triticale starch granules were similar to those observed in the starch of wheat (Zhang *et al.*, 2013), as well as barley and triticale (Ao and Jane, 2007), though they were higher than those reported for waxy wheat (Yoo and Jane, 2002). Differences in percent crystallinity values of starches from several sources have been attributed to the amylose (Yoo and Jane, 2002), lipid (Finnie *et al.*, 2009), and amylopectin contents and amylopectin chain length, as well as the orientation of amylose double helices (Zhang *et al.*, 2013).

The gelatinization process triggers several changes in the physicochemical properties of starches which be measured with a rheometer. At the sol-gel transition, the starch solution loses its liquid properties and begins to turn into a solid substance. In this study, the gelatinization mechanism of starches from complete (Eronga) and substituted (Yoreme) developing triticale grains was studied by means of a dynamic mode rheological analysis. Oscillatory measurements were performed within the linear viscoelasticity region and the results provided information on the dynamic mechanical properties of the starch gels evaluated as the elastic modulus  $G'$  and the viscous modulus  $G''$ . Figure 2A,B shows the evolution of the elastic ( $G'$ ) and viscous ( $G''$ ) moduli of starch granules from the complete and substituted triticales, respectively, as a function of temperature. The changes in  $G'$  and  $G''$  of the 16 to 40 DAA starch dispersions displayed similar behaviour. In general, for both triticale genotypes the  $G'$  and  $G''$  moduli values reached maximum levels in the temperature range of 71-86°C, and dropped with further heating at around 90°C. Both moduli showed the same profile; however,  $G'$  displayed higher values than  $G''$  in both triticale genotypes, although those of the complete genotype were higher than the substituted genotype. Higher values of  $G'$  and  $G''$  have also been observed in corn (Kaur *et al.*, 2008) and wheat (Ulbrich *et al.*, 2015) starch dispersions. The effect of frequency on the elastic modulus  $G'$  and viscous modulus  $G''$  of the starch of developing complete (Eronga) and substituted (Yoreme) triticale grains are shown in Fig. 2C, D. All samples showed a solid-like system. The values of  $G'$  and  $G''$  moduli of all triticale starches were independent of frequency, which is typical of 'true gels' (Sang *et al.*, 2008). Also, all starch samples showed  $G'$  values higher than those of  $G''$ , and were higher



**Fig. 1.** X-ray diffraction of the starch of complete (AABBRR) triticale on 16, 22, 31, and 40 DAA (A, B, C and D, respectively) and substituted (AABBDR) triticale on 16, 22, 31, and 40 DAA (E, F, G and H, respectively).

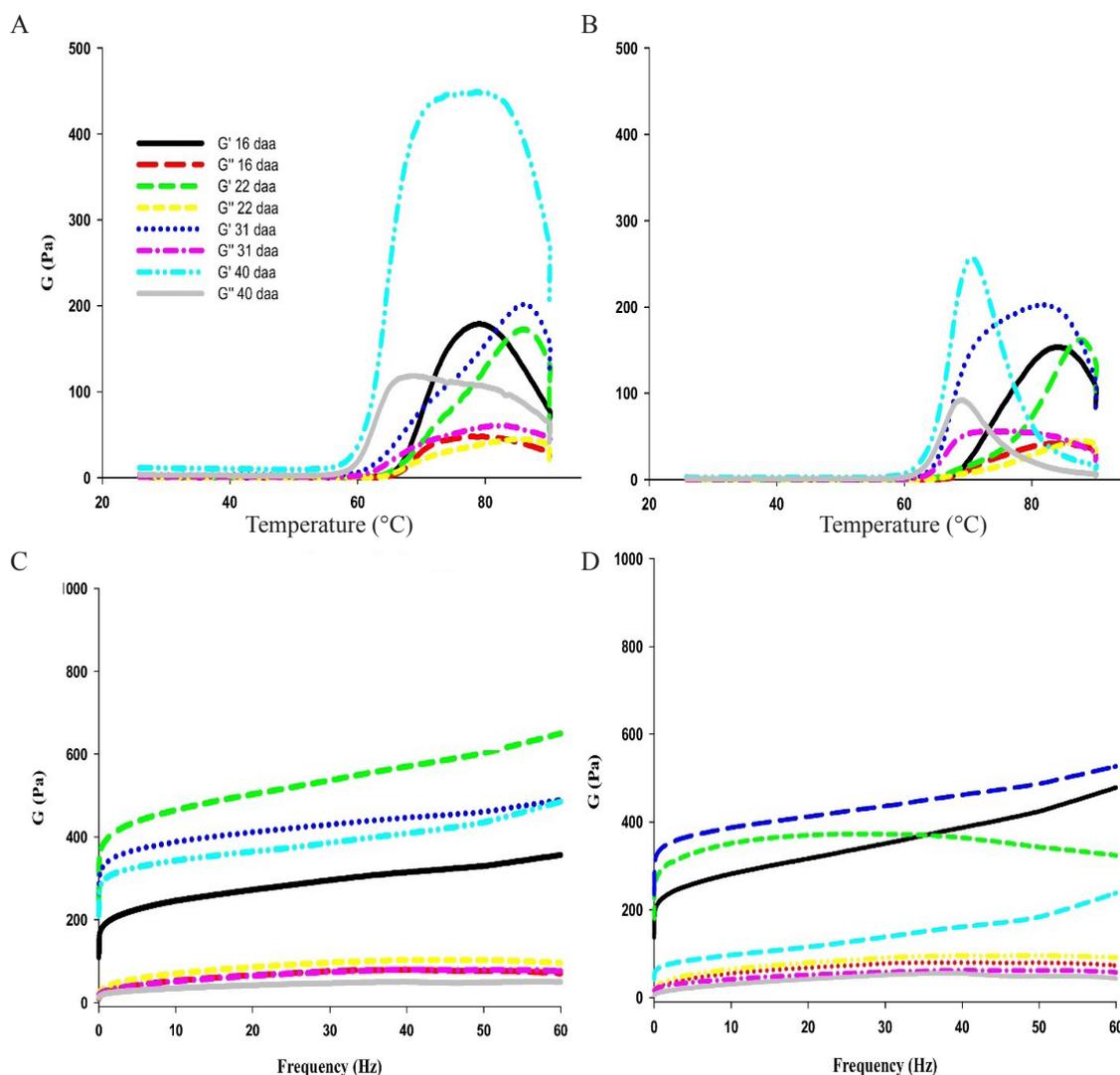
**Table 2.** Crystallinity percent of starch granules from complete (AABBRR) and substituted (AABBDR) developing triticale grains on different days after anthesis (DAA)

DAA	Crystallinity (%)	
	Complete triticale (Eronga)	Substituted triticale (Yoreme)
16	26.5 ± 0.7aB	27.0 ± 0.6aC
22	27.3 ± 0.4aB	28.1 ± 0.3aBC
31	28.6 ± 0.7aB	29.2 ± 0.4aB
40	36.1 ± 0.1aA	36.8 ± 0.3aA

Explanations as in Table 1.

in the complete triticale than in the substituted one, suggesting a gel-like behaviour similar to that observed in the starch of corn (Kaur *et al.*, 2008), sorghum (Sang *et al.*, 2008) and wheat (Ulbrich *et al.*, 2015). Furthermore, the starch of both triticale genotypes exhibited very similar  $G''$  values, suggesting that its viscous contribution to the gel structure was similar throughout its synthesis in the developing grains.

The sol-gel transition temperature  $T_0$  was determined at the crossover point of  $G'$  and  $G''$ . Table 3 shows the  $T_0$  values of starches from complete (Eronga) and substituted (Yoreme) developing triticale grains. The starches of the developing complete triticale grains showed significantly lower  $T_0$  values than those of the substituted triticale throughout the developing period. The  $T_0$  values of complete and substituted triticale were the highest on 16 DAA



**Fig. 2.** Changes in elastic  $G'$  and viscous  $G''$  modulus during heating (the angular frequency sweep scan range of 0.01-60 Hz; constant strain 1%; ramp temperature  $5^{\circ}\text{C min}^{-1}$ ; start temperature  $25^{\circ}\text{C}$ ; final temperature  $90^{\circ}\text{C}$ ) of 10% starch suspensions of the complete triticale (AABBRR) and the substituted triticale (AABBDR) by the effect of temperature (A and B) and frequency (C and D) using a stress-controlled Discovery Hybrid Rheometer HR-2 equipped with two parallel plates with 40 mm in diameter.

**Table 3.** Sol-to-gel phase transition temperature ( $T_0$ ), degree of polymerization of amylose and apparent amylose content of starches extracted from complete (AABBRR) and substituted (AABBDR) developing triticale grains on different days after anthesis (DAA)

DAA	Transition temperature ( $T_0$ , °C)	Degree of polymerization of amylose*	Apparent amylose (%)*
Complete triticale			
16	63.9±0.4aB	375±49a	13.5±0.2b
22	63.6±0.5aB	405±36a	11.7±0.7a
31	59.5±0.7bB	811±42a	16.5±0.8a
40	56.1±0.3cB	1549±42a	23.5±0.8a
Substituted triticale			
16	66.3±0.3aA	254±38b	14.1±0.2a
22	65.2±0.2aA	287±43b	11.3±0.7a
31	62.7±0.3bA	756±41b	15.9±0.6b
40	60.3±0.8cA	1313±62b	22.1±0.7b

Values are expressed as mean  $\pm$  standard deviation. Values followed by the same: lowercase letter and capital letter in the same column are not significantly different for similar DAA ( $p \leq 0.5$ ). \*Data taken from Cornejo-Ramírez *et al.* (2016).

(63.9  $\pm$  0.4 and 66.2  $\pm$  0.3°C, respectively) and 22 DAA (63.6  $\pm$  0.5 and 65.2  $\pm$  0.2°C, respectively), and decreased with time, reaching the lowest values on 40 DAA (56.1  $\pm$  0.3 and 60.3  $\pm$  0.8°C, respectively). The highest  $T_0$  values were observed in starches of the substituted triticale from the 16 DAA and to 22 DAA with no significant differences. However, the transition  $T_0$  values determined in this research for both triticale starches were lower than those reported for acid-modified wheat (69.4°C), potato (80°C) and pea (80°C) (Ulbrich *et al.*, 2015). Yet, they fell within the range of values reported for normal corn (65.7°C) (Kaur *et al.*, 2008) and normal wheat (46-57.1°C) starches (Sasaki *et al.*, 2000). The differences in  $T_0$  of starches from diverse cereal cultivars might be due to variations in the granular structures, degree of polymerization and molecular architecture of amylopectin (Singh *et al.*, 2003).

The higher percent crystallinity of the starch makes the granules more resistant to gelatinization because the longer chains of amylopectin require higher temperatures to dissociate completely, while the opposite occurs for amylose (Yamin *et al.*, 1999). Also, a starch with high amylose content has a more amorphous region and, therefore, a less crystalline region, as a result of which low temperatures are needed to achieve gelatinization (Sasaki *et al.*, 2000). In our study, the starches from the substituted triticale genotypes showed, in general, higher crystallinity values throughout the developing period, although no significant statistical differences were observed.

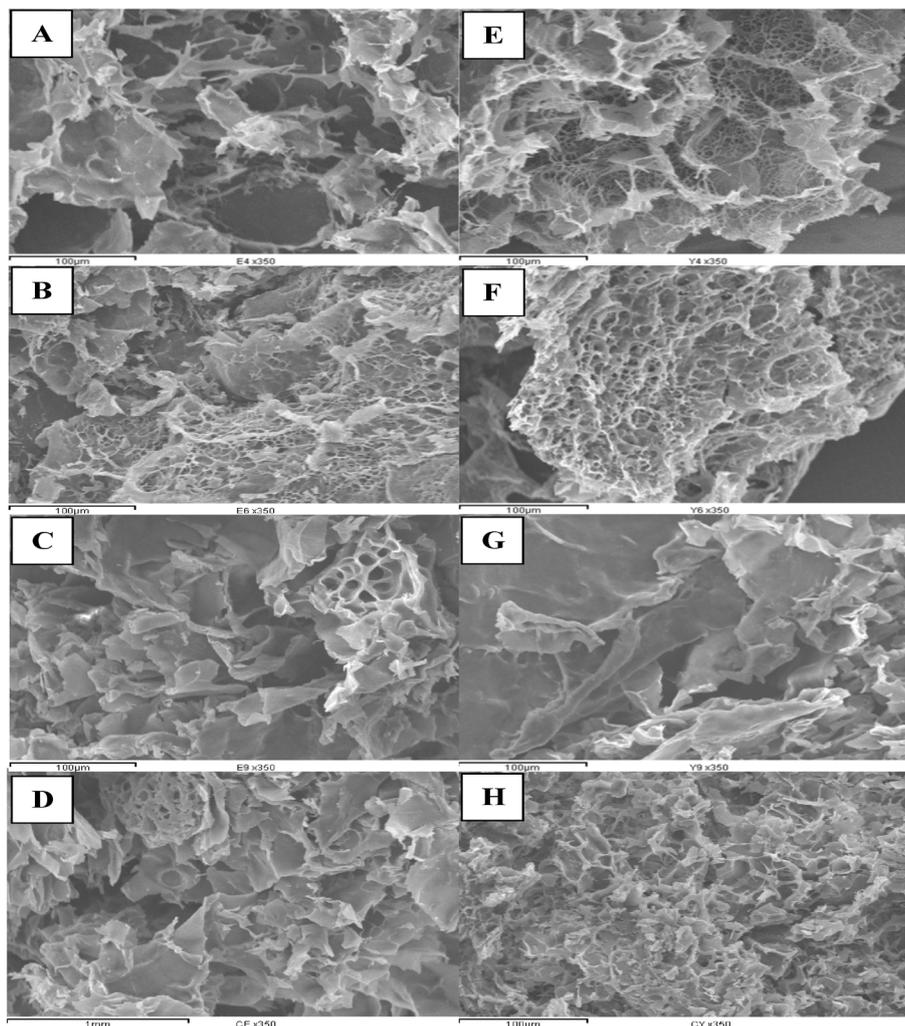
The  $T_0$  values were strongly influenced by genotype, considering that the starches of the developing grains of the complete triticale Eronga showed lower transition tempera-

tures ( $T_0$ ) and were related to higher amylose contents and higher amylose DP, in contrast to those of the substituted triticale Yoreme (Table 3).

The gels made with the starch of triticale developed on 16 DAA showed large air cells of sponge-like structures (Fig. 3), which has also been observed in waxy corn (Kaur *et al.*, 2008) and corn starch (Alishahi *et al.*, 2015). However, both gels displayed some differences. The starch gel of complete triticale (Eronga) showed an internal structure more similar to the gels of corn starch, while the starch gel of the substituted triticale (Yoreme) displayed an internal structure showing small air cells within large ones.

The gels made with the triticale starch samples on 22 DAA formed a closed network, creating small cells of sponge-like structures, similar to the internal structure of the gels made with normal corn starch (Kaur *et al.*, 2008). The internal structure displayed by all these gels might be due to higher DP of amylose (Table 3) that could make a thick matrix, capable of filling the space between the remnants, resulting in a porous structure due to the formation of voids, similar to a honeycomb-like structure. The differences in the gel microstructures might be due to the differences in the physicochemical characteristics of the developing starch granules to make the gels. The starches of complete and substituted triticale differed in terms of the DP of amylose (Cornejo-Ramírez *et al.*, 2016), size and volume percent of A- and B-starch granules (Table 1) at this stage of development.

The gels made with the starch samples of the triticale collected on 31 DAA formed a network with larger cells of sponge-like structures, though not as large as those of the gel network made with the starch formed on 16 DAA. The internal structures of the gels were similar to those made



**Fig. 3.** Scanning electron micrographs of starch gel samples prepared from starches of developing grains of a complete triticale (AABBRR) on 16, 22, 31, and 40 DAA (A, B, C and D, respectively), and of a substituted triticale (AABBDR) on 16, 22, 31, and 40 DAA (E, F, G and H, respectively) using a magnification of 350x.

with waxy corn starch (Kaur *et al.*, 2008); however, the starch gel of the complete triticale showed similar areas to the gels made with wheat starch-wheat fibre (Sun *et al.*, 2015) and corn starch (Alishahi *et al.*, 2015).

The gels made with the triticale starch samples at maturity (40 DAA) showed differences among them in their internal structures. The gel made with starch of complete triticale displayed an internal structure with a minor dense honeycomb structure, with reduced internal pores, and a large lamellar structure similar to the gels made with wheat starch (Sun *et al.*, 2015). Therefore, large starch gels have the advantage in competing for water molecules forming a strong network structure that enhances the mechanical properties of noodles, vermicelli and sheet jelly (Alishahi *et al.*, 2015). In contrast, the gel made with the starch of substituted triticale displayed more similarities to the gels prepared with wheat starch (Sun *et al.*, 2015) and normal corn starch (Kaur *et al.*, 2008). The corn starch gels showed low peak viscosity,  $G'$ ,  $G''$  and  $T_p$  and less pronounced changes in the rheological parameters within the frequency

sweeps, low stability of pastes at low temperatures and low resistant to retrogradation. The gels made with substituted triticale showed the same rheological behaviour as the corn starch gels, and they could have low cohesiveness and high values of springiness, hardness, gumminess and viscosity (Sun *et al.*, 2015).

## CONCLUSIONS

1. The results showed that the starches of the complete genotype displayed different gel properties than those obtained from the substituted genotype, due to differences in the molecular structure of their amylose and amylopectin, synthesized on different DAA during grain developing.

2. The complete triticale developed bigger and more crystalline A- and B-type starch granules than the substituted triticale. Although no significant differences in crystallinity were observed between the starches collected in different DAA, the substituted triticale showed higher crystallinity values, which coincided with the low amylose content reported in a previous study.

3. Dynamic rheology determinations revealed that the complete triticale displayed higher values of elastic  $G'$  and viscous  $G''$  moduli than the substituted triticale.

4. Starches of the complete triticale collected on several DAA showed lower phase transition temperature values than those obtained from the substituted one, suggesting lower gelatinization temperatures.

5. The microstructure of gels prepared from starches extracted from the complete triticale grains displayed a denser internal structure.

**Conflict of interest:** The authors have no conflict of interest to declare.

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