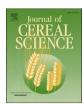
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# Novel biofortified sorghum lines with combined waxy (high amylopectin) starch and high protein digestibility traits: Effects on endosperm and flour properties



Mohammed S.M. Elhassan <sup>a</sup>, M. Naushad Emmambux <sup>a</sup>, Dirk B. Hays <sup>b</sup>, Gary C. Peterson <sup>c</sup>, John R.N. Taylor <sup>a, \*</sup>

- a University of Pretoria, Institute for Food, Nutrition and Well-being and Department of Food Science, Private Bag X20, Hatfield 0028, South Africa
- <sup>b</sup> Texas A&M University, Molecular and Environmental Plant Sciences, 2474-TAMU, College Station, TX 77843, USA
- <sup>c</sup> Texas A&M AgriLife Research and Extension Center, 1102 East FM1294, Lubbock, TX 79424, USA

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#### ABSTRACT

Novel biofortified sorghum lines have been developed with both waxy starch (high amylopectin) and high protein digestibility traits. Eight sorghum lines with different combinations of waxy, non-waxy, high- and normal-protein digestibility traits were studied in relation to flour properties. Lines with the high protein digestibility trait had loosely packed starch granules and floury endosperm, irrespective of whether they were waxy or non-waxy. In terms of thermal properties, combined waxy-high digestibility lines had the highest onset endothermic temperature as well as endothermic energy compared to non-waxy, normal protein digestibility lines. The waxy-high protein digestibility lines had higher paste viscosity and formed much softer and less sticky pastes than the non-waxy, normal protein digestibility lines. Flours of the combined waxy-high protein digestibility sorghum lines had much higher solubility than the non-waxy- normal protein digestibility lines. At 30 °C flour solubility, waxy-high protein digestibility sorghum lines flour was similar to commercial wheat bread flours. The high flour solubility, high pasting viscosity and soft paste of sorghum lines with combined waxy and high protein digestibility traits indicate that their flours may have better properties for making dough-based food products than normal non-waxy, normal protein digestibility sorghums.

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#### 1. Introduction

Sorghum is the fifth most important cereal crop in terms of production, about 58 million tonnes in 2011, with Africa being the major producing region accounting for >40% of world production (FAOSTAT, 2013). Sorghum is highly suited for cultivation in the semi-arid and sub-tropical regions of Africa as it is one of the most drought-tolerant cereal crops (Srinivas et al., 2009). Further, sorghum does not elicit an adverse reaction in coeliacs (Ciacci et al., 2007). However, despite its high production and its applicability in gluten-free foods, the commercial utilization of sorghum is limited, particularly as flour for dough-based food products, and especially in bread. This is largely due to the fact that its flour does

Abbreviations: DSC, Differential scanning calorimetry; SEM, Scanning electron microscopy; WAI, Water Absorption Index; WSF, Water Soluble Fraction.

E-mail address: john.taylor@up.ac.za (J.R.N. Taylor).

not form gas-holding dough (reviewed by Taylor et al., 2006).

This drawback of sorghum presents a major challenge in Africa, where increasing dependence on wheat to produce bread to meet the needs of the rapid expanding urban population is negatively affecting the continent's economic situation. According to the International Food Policy Research Institute, in 2013 African countries were spending US \$12 billion to import 40 million tons of wheat (IFPRI, 2013).

The major reason for the inferior bread-making properties of sorghum is that kafirin, the sorghum prolamin protein, does not exhibit the visco-elastic properties of wheat gluten in normal dough systems (Taylor et al. (2014). However, Goodall et al. (2012) showed that flour of sorghum with a high protein digestibility trait (Weaver et al., 1998), resulting from modified kafirin prolamin protein body development (Oria et al., 2000), produced better quality sorghum-wheat composite doughs and breads than normal sorghum flour. This was attributed to formation of an improved dough protein network. Furthermore, the starch granules in the

<sup>\*</sup> Corresponding author.

corneous endosperm of sorghum are surrounded by hydrophobic matrix proteins (Munck, 1995), comprising the kafirin protein bodies and glutelins (Taylor et al., 1984). This matrix of hydrophobic proteins may reduce the extent of water absorption and solubilisation of sorghum starch (Chandrashekar and Kirleis, 1988; Ezeogu et al., 2005). In turn, this may lead to inadequate functionality of sorghum flour because in wheat flour starch water holding is related to dough functionality (Dexter et al., 1994).

Novel biofortified sorghum lines which combine the high protein digestibility trait with the waxy (high amylopectin) starch trait, hence having high starch digestibility (Rooney and Pflugfelder, 1986), have recently been developed by Texas A&M University through conventional breeding (Jampala et al., 2012). These lines were developed from a cross between a waxy endosperm parental line and the high protein digestibility line described above, which was developed by Purdue University (Weaver et al., 1998). However, research into the end-use functionality of these sorghum waxy-high protein digestibility sorghum lines has been very limited.

The objective of this work was to examine the effects of sorghum lines with the waxy and high protein digestibility traits individually and in combination on characteristics related to flour functionality with the aim of determining the potential of these novel biofortified sorghum lines for making good quality doughbased food products.

#### 2. Materials and methods

#### 2.1. Sorghum samples

Eight sorghum lines, developed and bred through conventional plant breeding by Texas A&M University, were studied. All the lines were of the white tan-plant type. They comprised: two non-waxynormal protein digestibility lines coded 199 and 200, two waxynormal protein digestibility lines (coded 175 and 179) and one non-waxy-high protein digestibility line (coded 106), and three waxy-high protein digestibility lines (coded 109, 142 and 146). Sorghum lines coded 109, 142 and 146 were obtained via crossing lines Tx2907 and P850029 (Jampala et al., 2012). Tx2907 was released from the Texas A&M AgriLife Research sorghum breeding program as a waxy and normal protein digestibility sorghum (Miller et al., 1996). P850029, a high protein digestibility line, was developed at Purdue University from a population derived from the high lysine line P721Q (Weaver et al., 1998). The lines were increased at Halfway, Texas in 2012. The lines were blocked using rows of photosensitive lines as pollen breaks to avoid cross pollination.

MR Buster, a red, non-tannin, non-waxy, normal protein digestibility commercial hybrid sorghum was used as a standard. It was cultivated in Botswana and kindly supplied by the National Food Technology Research Centre in Botswana.

#### 2.2. Grain endosperm and protein body structure

Twenty sound grains from each sorghum line were dissected longitudinally. Endosperm texture was recorded by stereo light microscopy. By reference to sorghum endosperm type illustrations (ICC, 2011), the relative proportion of corneous endosperm to floury endosperm was estimated for each line. Endosperm structure was evaluated using scanning electron microscopy (SEM). Whole sorghum grains were dissected longitudinally by scalpel after freezing in liquid nitrogen. Then samples mounted on aluminium stubs using poster gum and were sputter coated with gold and then viewed using a Zeiss Evo LS15 SEM (Carl Zeiss, Oberkochem, Germany) operated at an acceleration voltage of 8 kV. Protein body structure was investigated using Transmission Electron Microscopy (TEM) as described (Da Silva et al., 2011a,b).

#### 2.3. Flour preparation

Twenty g sound grains of each line were decorticated using a Tangential Abrasive Dehulling Device (TADD, Venables Machine Works, Saskatoon, Canada), removing approx. 20% by weight of the grain outer layers. A laboratory hammer mill (Mikro-Feinmuhle-Culatti MFC Grinder, Janke and Kunkel, Staufen, Germany) with a 500  $\mu m$  opening screen was used to mill the decorticated grain.

#### 2.4. Flour moisture content

Moisture content was determined by NIR (DA 7200 NIR analyser, Perten Instruments Springfield, IL) using the supplier's calibration for sorghum.

#### 2.5. Protein content

Protein content (N x 6.25), was determined by Dumas combustion according to AACC method 46–30 (AACC International, 2000).

#### 2.6. Starch amylose content

Amylose content was determined by the Megazyme amylose/amylopectin assay kit procedure (Megazyme Ireland International, Bray, Ireland). Amylopectin in the sample is specifically precipitated by the addition of the lectin concanavalin A (Con A) and removed by centrifugation. The concentration of amylose in the sample is determined colorimetrically using the glucose oxidase-peroxidase (GOPOD) reagent (Wong et al., 2009).

#### 2.7. In vitro protein digestibility

In vitro protein digestibility of the flours was determined according to the pepsin digestibility method of Hamaker et al. (1986)

**Table 1**Starch amylose content and in vitro pepsin protein digestibility of waxy and high protein digestibility sorghum lines and their controls (199 and 200) and normal red sorghum cultivar MR Buster.

Line	Starch type	Protein digestibility trait	Amylose (%)	Protein digestibility of raw sorghum flour (%)	Protein digestibility of cooked sorghum flour (%)
109	Waxy	High	3.9 <sup>a</sup>	$72.8^{\rm f} \pm 1.1$	$46.7^{\rm f} \pm 0.2$
142	Waxy	High	7.3 <sup>c</sup>	$58.1^{\circ} \pm 0.3$	$36.5^{de} \pm 0.0$
146	Waxy	High	5.8 <sup>b</sup>	$68.6^{e} \pm 0.2$	$48.1^{\rm f} \pm 1.7$
175	Waxy	Normal	12.1 <sup>d</sup>	$55.4^{ab} \pm 0.3$	$32.3^{ab} \pm 1.1$
179	Waxy	Normal	7.9 <sup>c</sup>	$54.3^{a} \pm 0.3$	$31.9^{a} \pm 1.1$
106	Non-waxy	High	23.2 <sup>e</sup>	$64.1^{d} \pm 0.0$	$38.7^{e} \pm 0.9$
199	Non-waxy	Normal	25.7 <sup>f</sup>	$55.4^{ab} \pm 0.1$	$32.7^{abc} \pm 0.1$
200	Non-waxy	Normal	27.8 <sup>g</sup>	$56.3^{b} \pm 0.1$	$35.4^{cd} \pm 1.2$
MR Buster	Non-waxy	Normal	30.8 <sup>h</sup>	$57.9^{c} \pm 0.2$	$35.0^{\text{bcd}} \pm 1.2$

Means with different superscript letters within a column are significantly different (p < 0.05).

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