



# Mineral content of sorghum genotypes and the influence of water stress



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## ABSTRACT

Sorghum is a source of several minerals whose content may vary depending on the genotype and the production environment. The objective of this study was to screen sorghum genotypes for mineral content and to investigate the effect of water stress on it. A large variability was observed in the mineral content of 100 sorghum genotypes grown in environments without (WoWS) and with water stress (WthWS). The water stress decreased Mn, P, Mg and S contents in 100, 96, 93 and 56% of genotypes, respectively. The genotypes and other factors seemed to have more impact than water stress on K, Ca, Cu, Fe and Zn levels. In 100 sorghum genotypes, 2 were classified as excellent sources of Fe and 25 of Zn, in both environments. The best two genotypes to Fe content were SC21 and SC655 and to Zn were SC320 and SHAN-QUI-RED which showed great potential for use in biofortification.

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## 1. Introduction

Minerals are inorganic elements widely distributed in nature and essential for growth and proper development of the human organism. The mineral deficiencies in diets may impair mental and physical development, decrease work output and contribute to morbidity from infections, especially among children, pregnant and lactating women (Hussain, Larsson, Kuktaite, & Johansson, 2010; Kayodé, Linnemann, Hounhouigan, Nout, & van Boekel, 2006; Ng'uni, Geleta, Johansson, Fatih, & Bryngelsson, 2011).

An appropriate diet can usually supply minerals. However, the diets of populations subsisting on cereals, or inhabiting regions where soil mineral imbalances occur, often lack some of them. The elements most frequently lacking in human diets are Fe, Zn and I, although other elements, such as Ca, Mg, Cu and Se can be deficient in the diets of some populations (White & Broadley,

2005). In Brazil, a high prevalence of anemia in the early years of life, especially in disadvantaged regions, has been frequently reported (Borges et al., 2007).

Low cost and relatively simple strategies have been proposed and adopted in an attempt to reduce the occurrence of mineral deficiencies such as, provision of medical supplements, fortification of foods and post-harvest change in eating habits (Davidsson & Nestel, 2004; Osendarp, West, & Black, 2003). Several biofortification projects have emerged as an alternative to contribute for the reduction of mineral deficiencies, especially iron and zinc. The objective of these projects is to increase the nutrient density in staple crops, mainly through agronomic intervention and genetic selection (White & Broadley, 2005). There is considerable genetic variation within crop species that is suitable for sustainable biofortification strategies. However, to ensure success in this research and development, a multidisciplinary approach is necessary, and the screening and selection of breeding lines or accesses for higher contents of essential nutrients is a preliminary and basic stage of development.

Cereals grains are the most common foods used in biofortification programs because they have been the major source of calories for human diets (Taylor, Taylor, & Kini, 2012; White & Broadley, 2005). In this sense, several studies have related expressive mineral levels in wheat, rice, maize and sorghum (Bänziger & Long, 2000; Hussain et al., 2010; Kayodé et al., 2006; Martino et al., 2012; Ndukwe, Edeoga, & Omosun, 2015; Ng'uni et al., 2011;

*Abbreviations:* WthWS, with water stress; WoWS, without water stress; Embrapa, Brazilian Agricultural Research Corporation; IGD, Institute of Genome Development; USDA, United States Department of Agriculture; ANOVA, Analyzes of Variances; ICRISAT, International Crops Research Institute for the Semi-Arid-Tropics.

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Proietti, Mantovani, Mouquet-Rivier, & Guyot, 2013; Queiroz et al., 2011; Zhang et al., 2010). In sorghum, the most usually abundant mineral is K, followed by P and Mg and the most abundant micro-element is Fe (Afify, El-Beltagi, Abd El-Salam, & Omran, 2012; Pontieri et al., 2014).

*Sorghum bicolor* L. Moench is an important cereal in the world and can grow under adverse environmental conditions, such as very dry, saline and hot areas, where the production of other cereals is uneconomical (Dicko, Gruppen, Traore, van Berkel, & Voragen, 2006). The cereal is used for food in Africa and Asia and for animal feed and ethanol production in the Americas and Australia. There is an increased interest in also using sorghum for human consumption due to the fact that it is gluten-free (Pontieri et al., 2013) and has other health benefit properties, such as cholesterol-lowering, anti-inflammatory, slow digestibility and inhibition of human esophageal and colon cancer cell growth (Awika, Yang, Browning, & Faraj, 2009; Carr et al., 2005; Moraes et al., 2012).

In Brazil, Embrapa Milho e Sorgo (Brazilian Agricultural Research Corporation) and partner institutions have been conducting breeding programs seeking the selection of sorghum genotypes with improved quality for human consumption. There is a large collection of sorghum accessions that have not been characterized for food quality characteristics at Embrapa. Thus, there is a great potential to be explored for the use of some of these genotypes to develop biofortified sorghum cultivars. Furthermore, there has been no investigation about the effect of water stress on the mineral levels of these genotypes.

It is widely known that grain mineral contents are influenced by genotype, environment and interactions between genotype and environment (Hussain et al., 2010; Ng'uni et al., 2011; Ray, Shipe, & Bridges, 2008; Zhang et al., 2010). In this context, plants exposed to some kind of stress may show a wide range of mechanisms that involve morphological, physiological, and biochemical changes that are dependent on the inherent sensitivity of the particular genotype to stress (Cramer, Urano, Delrot, Pezzotti, & Shinozaki, 2011; Jogaiah, Govind, & Tran, 2013). According to Singh, Gupta, and Kaur (2012), wheat grain grown under water stress showed lower levels of Fe, but in relation to Zn, other factors also affected mineral content.

The main objective of this study was to screen sorghum genotypes for mineral content and to investigate the effect of water stress on content. In addition, this research aimed to identify superior genotypes to use in breeding programs to successfully develop biofortified cultivars with high iron and zinc density and availability.

## 2. Material and methods

One hundred sorghum accessions from the IGD (Institute of Genome Development) association panel (Casa et al., 2008) with high genetic variability were used in this study (Supplementary Table 1). Trials were planted at the Embrapa Milho e Sorgo research station, located in Nova Porteirinha, MG, at latitude 15°47'S, longitude 43°18'W and 516 m above sea level, in June 2010. The climate of this region is semi-arid, with regular rainfall and is used for drought tolerance tests evaluation. The soil was classified as dystrophic Red-Yellow Latosol. The genotypes were evaluated in two environments; without water stress (WoWS) and with post-flowering water stress (WthWS) in order to evaluate the effect of water stress on mineral content of sorghum grain. The experimental plots consisted of two rows three meters long, spaced 0.50 m between rows. Three hundred kg/ha of the NPK (nitrogen, phosphorus and potassium) formula 08-28-16 was applied at planting and twenty-five days after planting, 150 kg/ha of urea was applied. This is the recommended fertilizer rate for

the grain sorghum production system in this region. Supplemental water was applied by sprinkler irrigation for two hours once a week. In the WoWS environment, the irrigation remained until the grain-filling phase was complete and in the WthWS irrigation was suspended 50 days after planting, at the boot stage, that is just prior to the emergence of the panicle where the panicle is extended into flag leaf sheath. At maturity, in October 2010, the panicles were harvested and transported to Embrapa Milho e Sorgo in Sete Lagoas, Minas Gerais, where they were threshed and the grain was stored in a cold chamber at 10 °C until analysis.

### 2.1. Pericarp color, origin and race of the genotypes

The pericarp color of the genotypes was determined visually and the origin and race (Supplementary Table 1) was based on Casa et al. (2008), Morris et al. (2013), Sukumaran et al. (2012) and USDA (2013).

### 2.2. Levels of minerals in sorghum grain genotypes

The Long, Bänziger, and Smith (2004) methodology was used to remove any mineral contaminants from the field. The grain was washed for 10 s with running deionized water in a plastic sieve and was thoroughly dried with paper towel. After washing, the samples were transferred to paper bags and placed immediately in an oven with forced air circulation at 80 °C for 4 days. Following drying, the grain samples were ground in a cyclone mill (Marconi, Piracicaba, São Paulo, Brazil) to a particle size of 0.5 mm and the flour was packaged in polyethylene bottles until mineral analysis in the Laboratory of the Embrapa Milho e Sorgo, between April and May 2012.

The analyses of minerals P, K, Ca, Mg, S, Cu, Fe, Mn and Zn were determined according to the methodology proposed by Silva (1999). Acids and other chemicals were obtained from Sigma for use in the digestion process. All glassware and plastic ware were washed with deionized water, soaked in 2% HNO<sub>3</sub> overnight, rinsed with deionized water, and air-dried before use.

For quantitative analyses, the working standard solutions used for calibration were prepared by diluting a mono-element stock solution of 1000 mg mL<sup>-1</sup> Ca, Cu, Fe, K, Mg, Mn, P, S and Zn (Specsol, Jacareí, São Paulo, Brazil) and used to prepare multi-element analytical calibration solutions to desired concentration in 0.25 mol L<sup>-1</sup> HNO<sub>3</sub>. The ranges of the calibration curves (5 points) were selected to match the expected concentrations for all the elements of the sample studied by ICP-OES. High purity water (i.e., with conductivity approximately 18 MΩ cm<sup>-1</sup>) was used in all sample preparation and analysis steps.

The Inductively coupled plasma-optical emission spectrometer (ICP-OES) used was a Varian 720 ES (Varian, Santa Clara, CA, USA) with axial viewing configuration. The ICP-OES instrument was initialized and allowed to achieve thermal equilibrium over 30 min. Details of the operating conditions are summarized in Table 1. Emission lines utilized were shown in Table 2.

**Table 1**  
Operational conditions adopted for the elemental analysis of samples by ICP OES.

Operational conditions	
RF power (kW)	1.2
Gas	Argon
Plasma gas (L.min <sup>-1</sup> )	15.0
Auxiliary gas (L.min <sup>-1</sup> )	1.5
Nebulizer pressure (Kpa)	200.0
Pump rate (rpm)	15
View	Axial
Number of replicates	1
Nebulizer spray chamber	Sturman Master
Nebulizer type	V-Groove

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