



## *In vivo* protein quality of new sorghum genotypes for human consumption

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### ABSTRACT

The *in vivo* protein qualities were evaluated in flours from raw sorghum grains (RF) and flours from sorghum grains subjected to heat treatment in an oven (HTF) from the hybrids BRS 305, BRS 309 and BRS 310, developed by the Brazilian Agricultural Research Corporation (Embrapa). There were no differences in feed efficiency ratios among experimental groups. Heat-treated flour from BRS 309 and BRS 310 genotypes had higher protein efficiency ratios and net protein ratio values; however, they did not differ from those of flour from raw grain of BRS 310 genotype. Effects of heat treatment were observed in the BRS 309 genotype. Heat treatment did not affect true digestibility observed for the RF and HTF of the three genotypes. Lysine was the first limiting amino acid of the three sorghum genotypes. The HTF BRS 305 showed the lowest protein digestibility-corrected amino acid score value. Heat treatment improved the protein quality of genotype BRS 309; however, no differences were observed among the other genotypes.

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## 1. Introduction

Sorghum (*Sorghum bicolor* (L.) Moench) is the largest source of calories and protein for millions of individuals, mainly in Africa and Asia (Dicko, Gruppen, Traoré, Voragen, & Berkel, 2006; Mohammed, Mohamed, & Babiker, 2010). It is used to make porridges, beverages, and bakery products, among other traditional foods (Anglani, 1998; Méndez-Albores et al., 2009; Sanchez, 2003).

The protein content of sorghum corresponds to approximately 11.3% of the grain (USDA, 2010). The quality of its proteins and its amino acid composition can vary widely, depending on both genetics and location of cultivation (Deyoe & Shellenberger, 1965).

The protein quality of sorghum can be linked to phenolic compounds, such as tannins. These compounds are complexed to proteins, preventing their digestion and subsequent absorption (Dykes & Rooney, 2006). Cyanogenic compounds, such as dhurrin, show toxic potential when hydrolyzed by their glucosidases (Morant et al., 2008) and could impair animal growth.

Another determinant of the protein quality of the cereal is the technique of food processing. Cooking of sorghum for the production of porridge is the most commonly used preparation method (Duodu, Taylor, Belton, & Hamaker, 2003). *In vitro* studies have shown that wet cooking reduces the digestibility of sorghum compared to other cereals (Hamaker, Kirleis, Mertz, & Axtell, 1986; Mertz et al., 1984). According to some authors, the lower protein quality of this cereal (when cooked) is due to polymerization of the sorghum storage proteins, prolamins and kafirin, through the disulfide bond (Duodu et al., 2003; Hamaker et al., 1986), which also causes changes in protein secondary structure, from  $\alpha$ -helical to  $\beta$ -laminar (Emmambux & Taylor, 2009). However, very few studies have investigated the effect of dry heat treatment on the quality of sorghum protein. Recently, Correia, Nunes, Barros, and Delgadillo (2010) observed (in an *in vitro* model) that thermal processing methods, such as wet cooking, reduce the digestibility of sorghum and that dry heat and extrusion do not result in alterations.

The feeding potential of sorghum, Embrapa Maize and Sorghum has stimulated research to characterize genotypes with superior technological and nutritional quality, aiming to encourage the use of sorghum for human consumption. The aim of this study was to evaluate the *in vivo* protein quality of flours from raw sorghum grains (RF) and flour from sorghum grains subjected to heat treatment in an oven (HTF).

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**Table 1**  
Composition of experimental protein-free diet, with casein and with sorghum grain flours of genotypes BRS 305, BRS 309 and BRS 310 raw (RF) and subjected to heat treatment (HTF) (g 100 g<sup>-1</sup>).

Ingredients	Experimental diets							
	Protein-free	Casein	BRS 305 RF	BRS 305 HTF	BRS 309 RF	BRS 309 HTF	BRS 310 RF	BRS 310 HTF
BRS 305 RF	0	0	89.0	0	0	0	0	0
BRS 305 HTF	0	0	0	89.0	0	0	0	0
BRS 309 RF	0	0	0	0	75.2	0	0	0
BRS 309 HTF	0	0	0	0	0	75.2	0	0
BRS 310 RF	0	0	0	0	0	0	77.7	0
BRS 310 HTF	0	0	0	0	0	0	0	77.7
Casein	0	11	0	0	0	0	0	0
Maltodextrin	13.2	12.5	0	0	9.4	9.4	7.7	7.7
Sucrose	10	9.5	1.2	1.2	0	0	0	0
Soybean oil	7	7	4.7	4.7	4.8	4.8	4.6	4.6
Cellulose	10.2	10.2	0	0	3.3	3.3	2.6	2.6
Mineral mix	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5
Vitamin mix	1	1	1	1	1	1	1	1
L-cystine	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.3
Choline bitartrate	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Corn starch	54.6	45.1	0.3	0.3	2.5	2.5	2.6	2.6
Total	100.0	100.3	100.2	100.2	100.2	100.2	100.2	100.2

## 2. Material and methods

### 2.1. Sorghum genotype samples

Three sorghum genotypes were analyzed in this study, all of them developed and supplied by Embrapa Maize and Sorghum, Sete Lagoas, MG, Brazil: BRS 305 (light brown pericarp, with testa), BRS 309 (white pericarp, without testa) and BRS 310 (red pericarp, without testa). The cultivation of sorghum genotypes was conducted using spacing between rows of 0.70 m, mean density of 140,000 plants per hectare. Fertilization at sowing consisted of 300 kg ha<sup>-1</sup> of the 8-28-16 NPK formula +0.5% Zn. Other fertilization included 100 kg ha<sup>-1</sup> of urea, applied 40 days after germination. The planting was in Sete Lagoas, MG, Brazil, in February 2009.

### 2.2. Flour preparation, and determination of total phenolic and condensed tannins of sorghum genotypes

The sorghum grains were manually selected and sifted to remove dirt and impurities. To prepare the flours from grains subjected to heat treatment, the grains of the three sorghum genotypes were exposed to 105 °C in an oven with air circulation (Nova Ética®, model 400/6ND, Vargem Grande Paulista, São Paulo), as proposed by Souza et al. (2005), but modifying the exposure time to 30 min. Afterwards, raw and heat-treated grains were ground with pericarp in a knife mill (C.W. Brasender®, Dusburg, Germany) in order to obtain flour with a particle size of 850 µm.

Total phenolic content of the flours was determined, using the Folin–Ciocalteu method, as described by Singleton, Orthofer, Lamuela-Raventós and Lester (1999). The condensed tannins were measured, using the reaction vanillin/HCl, as described by Burns (1971) with modifications indicated by Maxson and Rooney (1972) and Price, Van Scoyoc, and Butler (1978).

### 2.3. Biological assay

Diet composition was based on guidelines from the American Institute of Nutrition for rodent growth (AIN-93G), according to Reeves, Nielsen, and Fahey (1993), with modification in the protein content to 9%. The diets were homogenized in an industrial mixer (Lieme®). After preparation, the protein content was determined for each diet by the semimicro Kjeldahl method (928.08), according to AOAC (2002). The diets were packed in plastic bags, properly labelled and stored in a refrigerator at 5 °C.

All experimental diets were formulated to provide the same energy density. The cellulose concentration was changed to 10.2%, depending on the fibre content of sorghum genotypes, to standardize the concentration of this nutrient among all diets (Table 1).

### 2.4. Animals

The experiment was carried out with forty-eight male rats (*Rattus norvegicus albinus*, Rodentia mammalia), Wistar, recently weaned, 23 days of age and weighing 51–60 g, from the vivarium of the Center for Biological Sciences and Health, of the Federal University of Viçosa.

The animals were randomly divided into eight groups of six animals, so that the difference between mean weights did not exceed 2.2 g, as recommended by AOAC (2002). The rats were kept in individual stainless steel cages and maintained at 22 ± 3 °C with a 12 h light/dark cycle.

The animal groups were fed the following diets: protein-free diet, casein, and diets with flour from raw sorghum (RF) and flour from heat-treated grains (HTF) of the genotypes BRS 305, BRS 309 and BRS 310 (Table 1). During the 14 days, animals received the experimental diets and deionized water *ad libitum*.

The study protocol was approved by the Ethics Committee of the Veterinary Department, Federal University of Viçosa, Brazil (Protocol n°28/2010).

### 2.5. Feed efficiency ratio

During the experimental period, animals were weighed on the 1st, 7th and 14th days and the feed efficiency ratio (FER) was determined, which represents the relationship between weight gain (g) and dietary intake by the animals (g).

### 2.6. Protein efficiency ratio and net protein ratio

The protein efficiency ratio (PER) was determined by the Hegsted method (1977), which relates the weight gain of animals (g) with the protein intake (g), modified for 14 days of experiment. The relative protein efficiency ratio (R-PER) was determined with the PER result of a casein diet established at 100%. Net protein ratio (NPR) was calculated according to Bender and Doell (1957), taking into account the weight gain of test group (W) and the weight loss of the protein-free diet group (WL) in relation to the test group's protein consumption (PC), according to the formula:

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