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Effects of Cereal Malts Used as Improver on Physico-chemical, Nutritional and Sensory Characteristics of Wheat and Red Sorghum Composites Breads

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Abstract The aim of this study is to optimize composite breads of wheat and whole red sorghum flour, using natural improvers. Three types of local malted cereals were used as natural improvers. The red sorghum flour was fermented with EPSs producing LAB strain prior to use. The technological characteristics of the composite flours were determined using an alveograph. The physico-chemical and nutritional characteristics of the composite breads were determined using standard methods and their sensory profiles were evaluated by a panel of 35 consumers. The alveograph results showed an increase in dough resistance, deformation and a decrease in extensibility and elasticity with the level of incorporation of red sorghum flour. No significant difference (p>0.05) was observed based on the physicochemical analyses with the three types of local cereal malts, except with the incorporation of 15% of the red sorghum flour, dry matter and water content. Significant difference (p>0.05) wasn't observed with the three types of cereal malts in the macronutrient contents of the composite bread samples. However, significant differences were observed according to the incorporation levels. An increase in protein content (11.65±0.91 - 15.76±0.54%/DM); crude fat content (1.80±0.08 - 2.65±0.16%/DM) and a decrease in carbohydrates content (84.82±0.19 - 79.72±0.47%/DM) was observed. Regarding the content of mineral elements, significant differences (p<0.05) were observed in the use of the three types of malt for the incorporation rate of 15% (Zn content), 30% (K and Mg content) and 50% (K content) of red sorghum flour. The free amino acid profile revealed six (06) free amino acids (Gly, Pro, Val, Met, Ile, and Lys) of which four are essential ones. Breads containing 30% of whole red sorghum flour was the most appreciated by consumers.

Keywords: *composite bread, cereal malts, improver, red sorghum, sensorial characteristics*

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1. Introduction

Sorghum (*Sorghum bicolor* L. Moench) is one of the oldest cereals of African origin; probably from Ethiopia, from where it is spread around the world [1]. Sorghum is consumed in various forms around the world like baked bread, porridge, tortillas, couscous, gruel, steam-cooked products, alcoholic, and non-alcoholic beverages etc [2]. This cereal is suit for processing into starch, flour, grits, and flakes, and it is used to produce a wide range of industrial products. It can also be malted and processed into malted foods, beverages, and beer [2]. Bread is a bakery product which main ingredients are water, flour, salt, yeast, sugar, and fat all mixed and fermented to form a viscoelastic dough before baking [3,4]. It is an important and mostly consumed staple cereal-based food globally and it contains useful nutrients such as starch, protein, fiber, vitamins, and minerals [5,6,7]. In addition, classical bread, made from wheat flour, interest is growing as a possible functional food due to its wide-spread distribution, convenience, sensorial properties, among others [7,8]. Usually, classical bread is considered a good source of energy and not an important element of nutrition. Its high content of sugar has been a concern in public, because it is often linked with risks of some diseases such as cardiovascular disease, diabetes and obesity [9]. The consumption of bread has become more and more spread in sub-Saharan Africa due to changes in food habit and urbanization. However, to meet the supply and this growing interest remains a great challenge [10]. Therefore, breads with health claims have gained more attention in this context, showing an opportunity to expand innovating bread production [7,11].

Wheat flour, the main ingredient in bread, is avoided by gluten intolerant patients as it contains high gluten content, low fiber and high glycemic level, and also contributes, to many disorders and diseases like diabetes, obesity, and atherosclerosis [12]. Due to such undesirable effects, breads with claiming free gluten, made from other cereal products is interesting in public health point of view. Thus, sorghum, could be an excellent substitute candidate. It is a gluten-free cereal [2]. Sorghum is an important source of many bioactive compounds such as flavonoids, which have been found to inhibit tumour development [2]. Sorghum sugars are released more slowly than wheat, hence it could be suitable for diabetics [2]. African food security, passes through raw material production and the use of indigenous grains such as millets or sorghum in manufacturing foods [13]. Imported wheat flour, especially in its refined form, is used to make breads and other bakery products. A strategy to reduce wheat import is to utilize composite flour containing wheat and sorghum in bakery products. There are also technological challenges to replacing wheat flour with millet or sorghum flour, which diminishes bread quality particularly with respect to loaf volume, texture, mouth-feel and staling rate [13,14]. Gluten proteins are essential for structure building; they form a network in the solid matrix and allow gas retention and expansion, which improve the bread volume [13,15]. Starch retrogradation and water molecule migration are the major causes of bread staling [16]. The gluten network can slow the movement of water, thereby maintaining softness and reducing bread staling [16]. The absence of gluten functionality in red sorghum restricts the high level of substitution in the wheat bread formula. One option is to employ hydrocolloids to mimic the properties of gluten. To do this, dextran produced by lactic acid bacteria (LAB) can act as a hydrocolloid in bread making [13,17]. In the practice, exopolysaccharide (EPS) -producing LAB are most often used to ferment gluten-free cereal flour dough into a slimy sourdough [13]. Some studies reported that the use of malt flour in controlled conditions improve loaf volume and crumb texture [18,19]. These positive effects were attributed to the natural enzymes expressed during the germination process that might decrease or completely replace the quantity of commercial enzymes added to bread formulation. This technique makes it possible to enrich cereals with hydrolytic enzymes such as beta and alpha-amylases, in sugars, in free amino acids and in vitamins, thus improving the technological and nutritional quality of the derived products [20,21,22].

The aim of this study is to evaluate the effect of local cereals malts flours used as natural improvers on the physico-chemical and nutritional characteristics of composite bread produced based on red sorghum and wheat flour with different incorporation of red sorghum flour.

2. Material and Methods

2.1. Vegetal Material

The vegetal material were local cereal grains including sorghum (*Sorghum bicolor* (L. Moench) and millet (*Pennisetum glaucum).* These cereals were purchased from national institute of environmental and agriculture research (INERA/Saria at Koudougou, Burkina Faso). Three varieties of local cereals grains (30 kg each one) were purchased, including «IKMP5" for millet grains, "Framida" for red sorghum grains and "Kapèlga" for white sorghum.

2.2. EPSs-producing LAB Strain Used as Starter Culture

The EPSs producing LAB strain (A16) used as starter culture for red sorghum dough fermentation was obtained from traditional fermentation process of *Massa* [23]. This strain was previously characterized as EPS producer and identified as *Weissella confusa/cibaria* using 16S rRNA gene sequencing [23].

2.3. Characteristics of Cereal Malts Used as Natural Improver

Three types of cereals malts were used as improver (Table 1) for composite bread production. It was red sorghum, white sorghum and millet malts. These malts were produced in controlled conditions at the IRSAT/DTA microbiology laboratory (Ouagadougou, Burkina Faso) [22]. The choice of these malts was mainly based on their diastatic power, α and β-Amylases activities.

Table 1. Characteristics of cereals malts used as improver

Cereals malts samples	Diastatic power (UPD)	α-Amylases (CERALPHA units)	β-Amylases (CERALPHA units)	Water content (%)	Dry matter (%)
SRn72	251.22 ± 6.8	221.65 ± 7.1	29.57 ± 2.6	9.44 ± 1.5	90.56 ± 1.5
SBn72	266.17 ± 8.9	237.33 ± 11.5	28.84 ± 3.2	9.86 ± 2.8	90.14 ± 2.8
PMn48	239.85 ± 19.3	201.91 ± 19.7	37.94 ± 1.5	8.39 ± 1.5	91.61 ± 1.5

SRn72: Red sorghum malt 72 h of germination; **SBn72:** White sorghum malt 72 h of germination; **PMn48:** Pearl millet malt 48 h of germination. [22].

2.4. Preparation of EPSs Producing LAB Inoculum and Sourdough

The EPSs producing LAB, *W. confusa*/cibaria, strain A16 (previously stored in MRS-broth + glycerol at -20 °C) was subcultured onto MRS (Man Rogosa and Sharp) agar (Liofilchem, Spain) and incubated for 48 h at 37°C. One colony was then subcultured in 10 mL of MRS-broth and incubated for 24 h at 37°C. A volume of 0.1 mL of culture broth was subcultured in MRS-broth (10 mL) and then incubated for 16 to 18 h at 37°C. The culture broth obtained was distributed in sterile cryotubes (1 mL/tube), then centrifuged (MIKRO 220R, Germany) at 8000 *g* for 10 min. The supernatant of each tube was removed and the pellet (cells) retained. Amount of 1 mL of sterile diluent [0.1% (w/v) peptone (Difco), 0.85% (w/v) NaCl (Sigma), pH 7.2 ± 0.2] was added to this pellet; after stirring, a further centrifugation was carried out at 8000 *g* for 10 min. The supernatant was again removed and the pellet was preserved. One millimeter (1 mL) of sterile distilled water was added to the pellet and, after stirring, the cells suspension which constitutes the inoculum was stored in the refrigerator at 4 °C. The concentration of viable cells of the inoculum was determined by enumeration on MRS agar. The inoculum (10^6 CFU/ml) was used for the fermentation of the red sorghum dough which has been the main element for the composite bread production. The dough was inoculated with *W. confusa*/cibaria strain (A16) at 1% (v/w then incubated in an oven at 25°C for 24 h to obtain a red sorghum sourdough.

2.5. Composite Bread Production Process

For the preparation of composite bread (Figure 1), the total amount of water used for kneading was between 62-63% (v/w) of the total flour mass. The dough was produced by mixing all the ingredients (yeasts, salt, sugar, cereal malt flour, wheat flour and the red sorghum dough fermented with the EPS-producing LAB strain A16) in a kneader for 4 min at low speed and 10 min at high speed. The dough obtained was put in to balls of 250 g and let to rest for 10 min. These balls are then mechanically molded into French baguette-type loaves of bread and let to rest in a proofing chamber for 45 min (30 °C, Relative Humidity 75%).

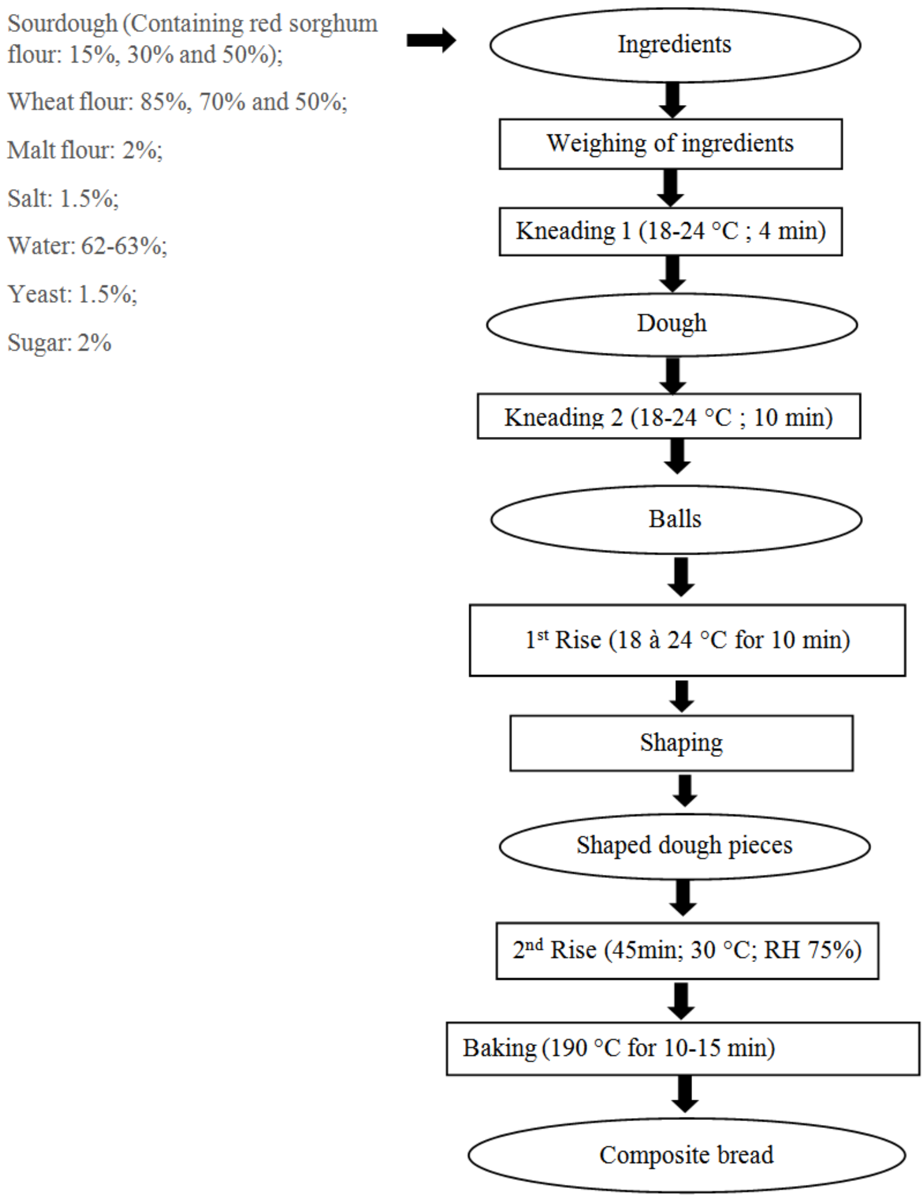

Figure 1. Composite bread production diagram

After the dough pieces had rested, the loaves were incised with a blade and baked in a rotary electric oven (RAMA hos, China), equipped with an automatic water injection system, with preheating at 240°C and baking at 190°C for 15 min. After baking, the loaves were cooled at room temperature. Composite breads without malt flour were used as control breads. Three formulations of composite bread (Table 2) were done using three types of cereals malt flour (red sorghum, white sorghum and millet malt flour).

Table 2. Composite bread formulations

Composite bread recipe	Formulation 1 (%)	Formulation 2 (%)	Formulation 3 (%)
Red sorghum flour	15	30.0	50
Wheat flour	85	70.0	50
Cereals malt flour	2.0	2.0	2.0
Yeast	1.5	1.5	1.5
Sugar	2.0	2.0	2.0
Salt	1.5	1.5	1.5

2.6. Physico-chemical and Nutritional Analyses

The strength and elasticity of the dough based on red sorghum and wheat flours were measured using a chopin alveograph (Alveolab, French) according to the french standard NF ISO 5530-4: 1992

The pH of the breads samples was measured with an electronic pH-meter (Model HI 8520; Hanna Instrument, Singapore). For each sample, 10 g of product were mixed with 20 mL of distilled water prior to pH measurement. For titratable acidity determination, 5 g of sample suspended in 30 mL of ethanol (90°) was mixed during 1 h and centrifuged for 5 min at 3500 g. From the supernatant, 20 mL was transferred to a 50 mL measuring flask and was titrated with NaOH 0.1 N using 1% phenolphthalein as indicator. The titratable acidity was calculated according to [24]. Water content was determined by drying the sample at 105 °C ± 2 °C for 24 h according to NF V03-707: 2000. Ash content was determined by incineration at 650 °C overnight according to international standard ISO 2171: 2007. Crude protein content (N×6.25) was determined by the Kjeldahl method [25] after acid digestion according to NF V03 50: 1970. Crude fat content was determined by soxhlet extraction using n-hexane according to ISO 659: 1998 and [26]. Total carbohydrates content was determined by spectrophotometric method at 510 nm using orcinol as reagent [27]. The energy value was calculated according to the Atwater method [28]. The determination of mineral elements was carried out by flame atomic absorption spectrometry (Perkin-Elmer model 303) according to [29]. The crude fiber content was determined by the formic insoluble method according to [30]. The free amino acid profile was carried out by high performance liquid chromatography (HPLC) using Waters PICO-TAG method [31] which consisted of three steps: hydrolysis of samples, sample derivatization pre-column and HPLC-reverse phase analysis. Amino acids identification and concentrations were determined with the Empower software by comparing retention times obtained with retention times of standards. The values were expressed in g/100g of dry matter.

2.7. Sensory Evaluation of Composite Bread Samples

The sensory evaluation consisted of evaluating the sensory profile of composite bread samples. A test of differentiation of the composite bread samples compared to the control sample (composite bread without malt flour) and a test of the classification of the composite bread samples were also performed according to [32]. The tasting panel included 35 consumers composed of men and women with a minimum age of 15 whom were familiar with composite bread samples.

2.8. Statistical Analyses of the Data

All the data (except those for sensory analyses) were submitted to Analysis of Variance (ANOVA) with the statistical software XLSTAT-Pro 7.5.2: 2016 and the means were compared using the test of Newman-keuls to the probability level $p < 0.05$. The curves were obtained using Microsoft Excel 2013. Sensory evaluation data were analyzed using the Chi^2 test with the statistical software SPSS.

3. Results

3.1. Characteristics of Flours

It emerged from the results of Table 3 that the characteristics of the dough from the composite flour (Wheat-red sorghum) in terms of tenacity (P), extensibility (L), swelling (G) and baking force or work (W) varied from 77 to 148 mmH_2O, from 13.4 to 67 mm, from 2.7 to 18.5 cm3 and from 141 to 309.10^{-4} J, respectively, according to the incorporation rate of red sorghum flour. These results showed that when the incorporation rate of red sorghum flour increased, the quality of the dough in terms of strength and elasticity decreased.

Table 3. Characteristics of flours (Wheat, Wheat-Red sorghum) through the alveograph

Parameters	Wheat flour (From CANADA)	Composite flour (Wheat-Red sorghum)		
Incorporation rate of red sorghum flour	**0 %**	**15 %**	**30 %**	**50 %**
P (mmH_2O)	122	148	135	77
L (mm)	130	67	40	13.4
G (Cm^3)	25.4	18.5	14.1	2.7
W (10^{-4}J)	430	309	197	141
P/L	0.9	2.2	3.4	5.7

P: Tenacity; **L:** Extensibility; **G:** Swelling; **W:** Baking force or work.

3.2. Physicochemical Characteristics of Composite Bread Samples

The results of the water content of the composite bread (Table 4) ranged between 31.70±0.09 and 35.00±0.49 % for composite bread samples containing 15% of red sorghum flour. For the breads containing 30% of red sorghum flour, the values ranged from 29.17±0.29 to 31.66±4.33% and for those containing 50% of red sorghum flour, the values ranged from 27.30±0.01 to 29.33 ±1.85%. The pH of composite breads varied from 5.38±0.01 to 5.44±0.03, from 5.15±0.02 to 5.32±0.06 and from 4.86±0.09 to 5.07 ±0.09, respectively for breads containing 15%, 30% and 50% of red sorghum flour (Table 4). Regarding the titratable acidity of the composite breads, its content increased with the incorporation rate of red sorghum flour. Indeed, the bread samples containing 50% red sorghum flour presented a higher acidity (ranged from 0.49±0.01 to 0.52±0.01%), followed by samples containing 30% of red sorghum (ranged from 0.33±0.01 to 0.43±0.02%) and finally those containing 15% of red sorghum flour (ranged from 0.19±0.01 to 0.22±0.02%) (Table 4). Composite breads without malt (controls) showed the lowest values compared to breads containing malt flour. The ash content of the samples ranged from 1.62±0.05%/DM to 1.74±0.02%/DM for the composite breads containing 15% of red sorghum flour (Table 4). For those containing 30% of red sorghum flour, the ash content ranged between 1.85±0.01%/DM and 2.05±0.03%/DM. For the breads containing 50% of red sorghum flour, ash content ranged from 1.97±0.08%/DM to 2.04±0.04%/DM. The ash content increased with the incorporation rate of red sorghum flour. For the crude fiber content, values ranged from 1.71 ± 0.76%/DM (bread samples without malt) to 2.39 ± 0.17%/DM (bread samples using malt flour). These crude fiber contents increased with the incorporation rate of red sorghum flour (Table 4). No significant difference (p>0.05) was observed in the use of the three types of malt for the physico-chemical parameters except with the incorporation of 15% and 30% of the red sorghum flour for the water content, dry matter, pH and titratable acidity.

Table 4. Physico-chemical characteristics of composite bread samples

	Water content (%)	Dry matter (%)	pH	Titratable acidity (%)	Ash (%/DM)	Fiber (%/DM)
			15% incorporation of red sorghum flour			
PSRr1	35.00±0.49^a	65.01±0.49^c	5.39±0.09^a	0.22±0.02^a	1.74±0.01^a	2.02±0.17^a
PSRb1	31.70±0.09^c	68.30±0.09^a	5.38±0.01^a	0.19±0.01^a	1.62±0.05^a	2.70±0.01^a
PSRm1	33.72±0.26^b	66.28±0.26b^b	5.44±0.03^a	0.21±0.01^a	1.68±0.02^a	1.82±0.31^a
PSRt1	33.01±0.03b^b	66.99±0.03^b	5.42±0.04^a	0.19±0.01^a	1.72±0.07^a	1.71±0.76^a
P-value	**0.005**	**0.005**	**0.821**	**0.293**	**0.392**	**0.437**
			30% incorporation of red sorghum flour			
PSRr3	29.17±0.29^a	70.84±0.29^a	5.16±0.01^b	0.43±0.02^{ab}	1.93±0.12^a	2.23±0.51^a
PSRb3	30.27±0.49^a	69.79±0.43^a	5.32±0.06^a	0.37±0.01 ^{a}	1.92±0.06^a	1.86±0.13^a
PSRm3	31.56±0.69^a	68.44±0.69^a	5.24±0.02^{ab}	0.33±0.02^b	2.05±0.03^a	1.78±0.35^a
PSRt3	31.66±4.33^a	68.34±4.33^a	5.15±0.02^b	0.33±0.01^b	1.85±0.01^a	1.97±0.27^a
P-value	**0.831**	**0.831**	**0.058**	**0.043**	**0.349**	**0.812**
			50% incorporation of red sorghum flour			
PSRt5	28.89±0.11^a	71.11±0.11^a	4.99±0.05^a	0.51±0.00^a	1.97±0.08 ^{a}	2.07±0.04^a
PSRb5	29.24±0.09^a	70.76±0.09^a	4.92±0.08^a	0.49±0.01^a	1.98 ±0.02^a	1.98±0.04^a
PSRm5	27.30±0.01^a	72.71±0.01^a	4.86±0.09^a	0.52±0.01^a	2.00 ±0.03^a	2.31±0.16^a
PSRr5	29.33 ±1.85^a	70.67±1.85^a	5.07±0.09^a	0.50 ±0.05^a	2.04 ±0.04^a	2.39±0.17^a
P-value	**0.006**	**0.463**	**0.397**	**0.822**	**0.782**	**0.195**

PSR: Red sorghum bread**; m:** millet malt**; r:** red sorghum malt**; b:** white sorghum malt**; t:** control (without malt) **1:** 15% incorporation of red sorghum**; 3:** 30% incorporation of red sorghum flour; **5:** 50% incorporation of red sorghum flour**.** Values with the same letters in the column are not significantly different at the 5% level according to Newman-keuls's test.

Table 5. Macronutrient composition of composite bread samples

	Protein (%/DM)	Crude fat (%/DM)	Carbohydrate (%/DM)	Energy value (kcal/100g)
		15% incorporation of red sorghum flour		
PSRr1	12.63±0.62^b	1.98±0.03^{ab}	83.65±0.59^a	402.93±0.10^a
PSRb1	11.65 ±0.91^b	2.10±0.12^a	84.63±0.84^a	404.01±0.79^a
PSRm1	12.42 ±0.04^b	1.85±0.02^{ab}	84.05±0.00^b	402.51±0.04^a
PSRt1	11.66±0.19^b	1.80±0.08^b	84.82±0.19^a	402.12±0.67^a
P-value	**0.070**	**0.126**	**0.088**	**0.204**
		30% incorporation of red sorghum flour		
PSRr3	12.9±0.24^a	2.16±0.10^a	82.99±0.03^{ab}	403.11±0.01^a
PSRb3	13.6±0.31^a	1.87±0.20^a	82.61±0.16^b	401.70±1.28^a
PSRm3	14.33±0.54^a	2.04±0.04^a	81.58±0.47^{ab}	401.99±0.09^a
PSRt3	12.49±0.17^a	2.13±0.10^a	83.54±0.27^a	403.22±0.48^a
P-value	**0.945**	**0.459**	**0.840**	**0.396**
		50% incorporation of red sorghum flour		
PSRt5	15.62±0.24^a	2.65±0.16^a	79.89±0.03^a	404.14±0.01^a
PSRb5	13.69±0.31^a	2.60±0.09^a	81.73±0.16^a	405.05±1.28^a
PSRm5	15.76±0.54^a	2.52±0.17^a	79.72±0.47^a	404.60±0.09^a
PSRr5	13.50 ±0.17^a	2.46±0.05^a	81.89 ±0.27^a	405.34±0.48^a
P-value	**0.461**	**0.752**	**0.344**	**0.544**

PSR: Red sorghum bread**; m:** millet malt**; r:** red sorghum malt**; b:** white sorghum malt**; t:** control (without malt) **1:** 15% incorporation of red sorghum flour**; 3:** 30% incorporation of red sorghum flour; **5:** 50% incorporation of red sorghum flour**.** Values with the same letters in the column are not significantly different at the 5% level according to Newman-keuls's test.

3.3. Macronutrient Composition of Composite Bread Samples

Results from Table 5, showed that the protein content of the bread samples varied from 11.65 ±0.91%/DM (PSRb1) to 15.76±0.54%/DM (PSRm5). The protein content increased according to the incorporation rate of red sorghum flour, leading to an increase in this content in composite bread in general and pearl millet malt in particular. The fat content also increased from 1.80±0.08%/DM (PMt1) to 2.65±0.16%/DM (PMt5). This increase correlated with the rate of incorporation of the red sorghum flour. As for the carbohydrates content, it decreased with the increase in the incorporation rate of red sorghum flour, from 84.63±0.84%/DM (PSRb1) to 79.72±0.47%/DM (PSRm5). The highest value was recorded with 15% incorporation of red sorghum flour, especially for the PSRb1 sample. The energy value varied from 402.51±0.04 kcal/100g (control bread) to 405.34±0.48 kcal/100g for the PSRr5 sample (sorghum bread with 50% incorporation of red sorghum flour using red sorghum malt as improver). No significant difference ($p>0.05$) was observed in the use of the three types of malt for the macronutrient contents except for the incorporation rate of 15% (crude protein, crude fat and carbohydrates) and 30% (carbohydrates) of red sorghum flour. On the other hand, differences were observed according to the levels of incorporation of red sorghum flour.

3.4. Minerals Composition of Composite Bread Samples

Considering the types of malt flour used, no significant difference ($p>0.05$) was observed for iron (Fe) content for breads samples containing 15% and 30% of red sorghum flour (Table 6). Values ranged from 2.18±0.00 mg/100g to 2.84±0.02 mg/100g; from 5.31±0.66 mg/100g to 5.76±0.26 mg/100g and from 7.94±0.91 mg/100g to 8.44±0.35 mg/100g respectively for the incorporation rate of 15%, 30% and 50% of red sorghum flour. Significant difference ($p<0.05$) was found for the incorporation of red sorghum flour at the rate of 15% depending on the type of malt used base on the content in zinc (Zn). These values are ranged respectively from 1.15±0.00mg/100g to 1.38±0.00 mg/100g. However, for 15% and 30% incorporation of red sorghum flour, no significant difference was observed ($p>0.05$). The potassium (K) content range from 204.36±0.00 mg/100g to 210.30±0.00 mg/100g for 15% of red sorghum flour breads from 229.71±0.67 mg/100g to 232.52±0.37 mg/100g for 30% of red sorghum flour breads from 241.45±0.35 mg/100g to 262.11±0.46 mg/100g for composite breads containing 50% of red sorghum flour. Sodium (Na) content ranged from 404.66±3.45 mg/100g to 469.79±6.28 mg/100g for bread samples containing 15% of red sorghum flour, from 427.15±15.87 mg/100g to 453.62±0.8 mg/100g for those with incorporation of 30% of red sorghum flour and ranged from 446.78±0.07 mg/100g to 450.83±8.15 mg/100g for samples with incorporation rate of 50% of red sorghum flour. Magnesium (Mg) content of the composite bread samples (wheat-red sorghum) are ranged from 30.16±4.16 mg/100g to 32.28±17.84 mg/100g; from 39.82±0.01 mg/100g to 42.71±0.01 mg/100g and from 51.92±0.24 mg/100g to 52.52±0.70 mg/100g, respectively for breads samples containing 15%, 30% and 50% of red sorghum flour. The results of the minerals composition obtained from composite bread samples showed an increase in the values according to the increase of incorporation rate of red sorghum flour. Regarding the content of mineral elements, significant differences ($p<0.05$) were observed in the use of the three types of malt for the incorporation rate of 15% (Zn content), 30% (K and Mg content) and 50% (K content) of red sorghum flour.

Table 6. Minerals elements composition of composite bread (Wheat-red sorghum) samples

Bread samples	Fe (mg/100g)	Zn (mg/100g)	K (mg/100g)	Na (mg/100g)	Mg (mg/100g)
15% incorporation of red sorghum flour					
PSRr1	2.54±0.43^{a}	1.32±0.00^{ab}	207.46±0.00^{a}	404.66±3.45^{a}	32.28±17.84^{a}
PSRb1	2.18±0.00^{a}	1.15±0.00^{b}	204.36±0.00^{a}	469.79±6.28^{a}	30.16±4.16^{a}
PSRm1	2.84±0.02^{a}	1.36±0.00^{a}	210.30±0.00^{a}	433.61±0.44^{a}	31.55±1.12^{a}
PRSt1	2.42±0.00^{a}	1.38±0.00^{a}	207.90±0.00^{a}	461.29±0.00^{a}	31.06±0.00^{a}
P-value	**0.441**	**0.029**	**0.966**	**0.070**	**0.581**
30% incorporation of red sorghum flour					
PSRr3	5.31±0.66^{a}	1.57±0.00^{a}	232.52±0.37^{a}	453.62±0.87^{a}	40.05±0.16^{c}
PSRb3	5.28±0.55^{a}	1.45±0.00^{a}	229.71±0.67^{b}	446.47±1.08^{a}	39.82±0.01^{c}
PSRm3	5.76±0.26^{a}	1.42±0.00^{a}	230.69±0.09^{b}	436.92±2.17^{a}	42.71±0.01^{a}
PSRt3	5.59±0.02 ^{a}	1.49±0.09 ^{a}	231.27±0.045^{ab}	427.15±15.87^{a}	41.43±0.06^{b}
P-value	**0.850**	**0.250**	**0.046**	**0.249**	**0.000**
50% incorporation of red sorghum flour					
PSRr5	8.08±0.02^{a}	1.59±0.02^{a}	250.16±1.30^{b}	448.05±0.37^{a}	52.52±0.70^{a}
PSRb5	8.44±0.35^{a}	1.58±0.05^{a}	260.32±0.16^{a}	449.98±3.5^{a}	52.26±0.13^{a}
PSRm5	7.94±0.91^{a}	1.52±0.01^{a}	241.45±0.35^{c}	446.78±0.07^{a}	52.51±0.06^{a}
PSRt5	8.07±0.01^{a}	1.56±0.00^{a}	262.11±0.46^{a}	450.83±8.15^{a}	51.92±0.24^{a}
P-value	**0.897**	**0.357**	**0.000**	**0.911**	**0.671**

PSR: Red sorghum bread**; m:** millet malt**; r:** red sorghum malt**; b:** white sorghum malt**; t:** control (without malt) **1:** 15% incorporation of red sorghum flour**; 3:** 30% incorporation of red sorghum flour; **5:** 50% incorporation of red sorghum flour**.** Values with the same letters in the column are not significantly different at the 5% level according to Newman-keuls's test.

Table 7. Free amino acids profile of composite bread (wheat-red sorghum) samples

Bread samples	Gly (%/DM)	Pro (%/DM)	Val (%/DM)	Met (%/DM)	Ile (%/DM)	Lys (%/DM)
15% incorporation of red sorghum flour						
PSRr1	0.000	1.298	0.000	0.000	0.000	0.000
PSRb1	0.000	1.401	0.000	0.000	0.000	0.082
PSRm1	0.000	1.360	0.000	0.000	0.000	0.000
PSRt1	0.007	0.575	0.163	0.037	0.000	0.135
30% incorporation of red sorghum flour						
PSRr3	0.000	1.922	0.000	0.000	0.000	0.088
PSRb3	0.000	1.997	0.000	0.000	0.000	0.166
PSRm3	0.000	1.784	0.000	0.000	0.000	0.000
PSRt3	0.000	0.438	0.067	0.059	0.014	0.138
50% incorporation of red sorghum flour						
PSRr5	0.000	0.945	0.000	0.000	0.000	0.000
PSRb5	0.000	0.836	0.000	0.000	0.000	0.080
PSRm5	0.000	0.927	0.000	0.000	0.000	0.000
PSRt5	0.000	0.526	0.000	0.025	0.043	0.127

PSR: Red sorghum bread**; m:** millet malt**; r:** red sorghum malt**; b:** white sorghum malt**; t:** control (without malt) **1:** 15% incorporation of red sorghum flour**; 3:** 30% incorporation of red sorghum flour; **5:** 50% incorporation of red sorghum flour**.**

3.5. Free Amino Acid Profile of Composite Bread Samples

The profile of free amino acid in composite breads (Table 7) showed the presence of six (06) free amino acids (Gly, Pro, Val, Met, Ile, and Lys). Among these amino acids, four are essential amino acids (Val, Met, Ile, and Lys). Proline is present in all the bread samples with a lowest content of 0.43%/DM in the PSRt3 bread and a highest content of 1.99%/DM in the bread produced with white sorghum malt at 30% incorporation of red sorghum flour (PSRb3). Isoleucine amino acid was present only in the control samples of bread using an incorporation of 30% and 50% of red sorghum flour (PSRt3 and PSRt5). Glycine amino acid is present only in control bread sample with an incorporation of 15% of red sorghum flour. From the result, it emerged that Met and Val contents were found in control bread samples produced using an incorporation of 15%, 30% and 50% of red sorghum flour (PSRt1, PSRt3 and PSRt5). Lys amino acid was found in the control bread samples and the bread samples using white sorghum malt as improver. Most of free amino acids content decreased with the increase of incorporation rate of red sorghum flour (Table 7).

3.6. Sensory Evaluation Results of Composite Bread Samples

Sensory evaluation is summarized in Figure 2. The consumers found that the breads made from wheat-red sorghum flour presented a brown and golden crust. Thus, 64.10% of the consumers reported that the brown color of the crust increased according to the increase of incorporation rate of red sorghum flour. It also emerged that the PSRb3 and PSRm3 bread samples (30% incorporation of red sorghum flour) presented the best aromas according to 61.73% of the consumers. The evaluation of the texture showed that the breads were slightly crispy according to 54.49% and 50% of consumers, respectively for incorporation rates of 15% and 50% of red sorghum flour. The tart taste sensation of the samples was pronounced with increasing rate of incorporation of red sorghum flour which is fermented into red sorghum sourdough. However, 81.41% of consumers found that the PSRm1 bread and the control bread had a low acid taste for a red sorghum flour incorporation rate of 15%. When the incorporation rate of red sorghum flour increased, 75% and 53.57% of consumers found that the taste became sour for 30% and 50% incorporation rates of red sorghum flour, respectively. Mouthfeel was rated as pleasant by 64.75% and 54.29% of consumers for the 15% and 30% incorporation rate of red sorghum flour, respectively. All breads produced with malts had a good mouthfeel for both of these incorporation rates (15% and 30%). For the 50% incorporation of red sorghum flour, consumers found the mouthfeel to be fair and the bread very hard. For the hedonic test (Figure 3), it emerged that 28.2% of the consumers found that the bread produced with millet malt (15% incorporation rate of red sorghum flour) was very pleasant. Breads produced with red and white sorghum malt (69.2% of consumers), millet malt (23.1% of consumers) and control bread (64.1% of consumers) were also pleasant. The incorporation rate of 30% of all the breads produced with malts was the most appreciated by the panel. Breads with millet and red sorghum malt were considered very pleasant with a proportion of 53.1% of consumers. Overall, the malts used had a positive effect on consumers' appreciation. For the 50% substitution, all the breads were considered pleasant with a proportion of 65.7% of consumers for the PSRr5 bread sample and the control bread, 62.9% for the PSRm5 bread and 45.7% for PSRb5 bread.

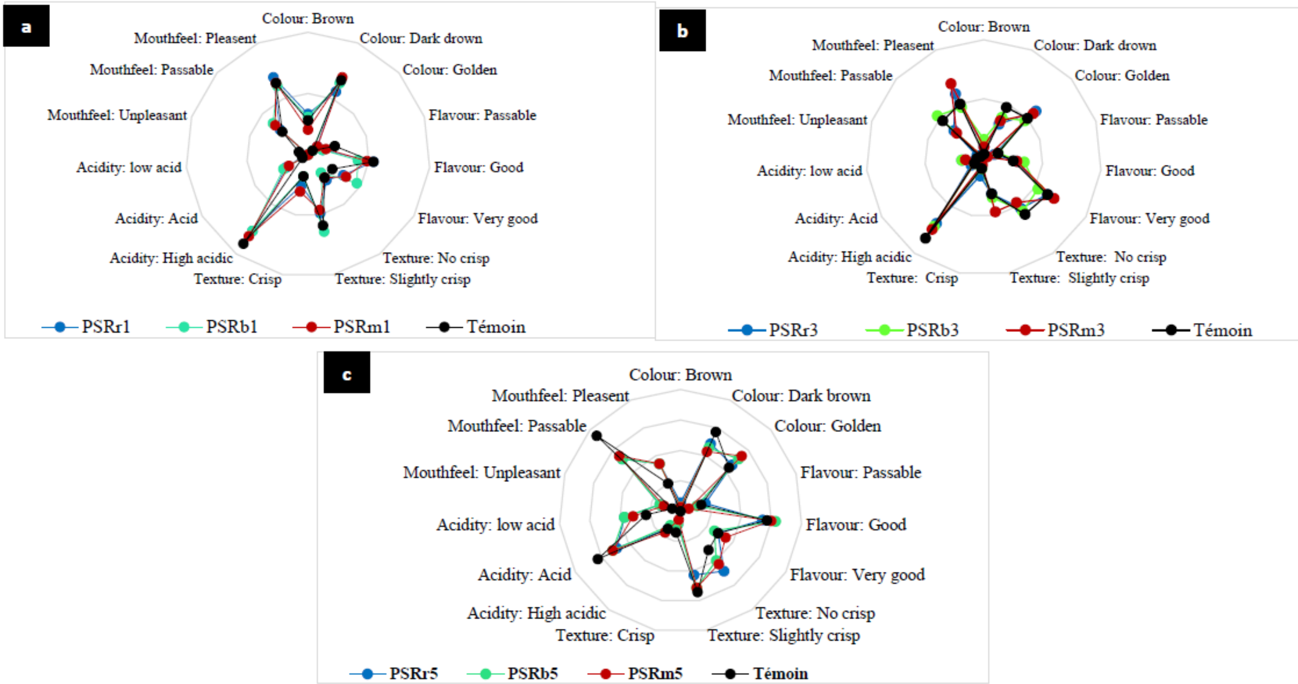

Figure 2. Sensory profile of composite bread made with wheat-red sorghum flour (**Legend: A:** Wheat-red sorghum bread (15% incorporation of red sorghum flour); **B:** Wheat-red sorghum bread (30% incorporation of red sorghum flour); **C:** Wheat-red sorghum bread (50% incorporation of red sorghum flour))

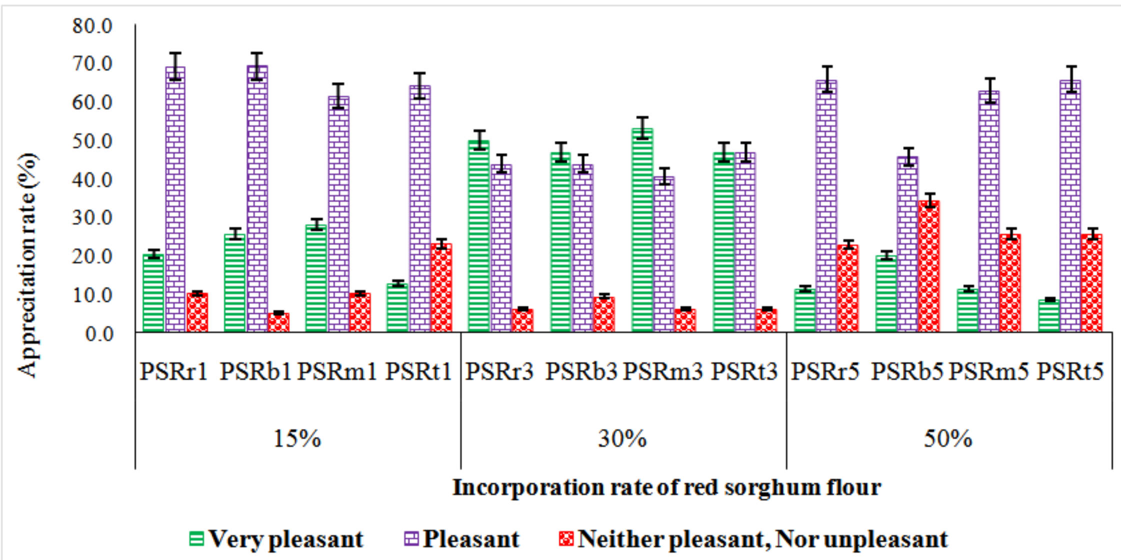

Figure 3. Hedonic characteristics of composite bread made with wheat-red sorghum flour (**PSR:** Red sorghum bread**; m:** millet malt**; r:** red sorghum malt**; b:** white sorghum malt**; t:** control (without malt) **1:** 15% incorporation of red sorghum flour**; 3:** 30% incorporation of red sorghum flour; **5:** 50% incorporation of red sorghum flour)

4. Discussion

The incorporation of whole red sorghum flour in composite breads resulted in an increase in the resistance of the dough to deformation, a decrease in extensibility and elasticity. The extensibility of a dough is dependent on both the extension rate due to viscous flow and the elastic properties of the dough. This last property influence the amount of stress required to stretch the dough [33]. High tenacity / extensibility (P/L) indicates a resistant and inextensible dough, while low P/L indicates a weak and extensible dough. P/L is often used industrially together with the baking force (W) to assess flour quality, as P/L indicates the shape of the alveogram and thereby the balance between tenacity and extensibility [33]. The W is one of the industrially most applied alveograph parameters, as it is used for prediction of processing behaviour of flour cultivars. The incorporation of 30% of red sorghum flour leads to good baking strength comparable to that obtained with 100% of some wheat flours with low W. On the other hand, beyond 30% incorporation of red sorghum flour, the volume of loaves during resting step decreases, which leads to breads of high density with a low volume. This could be justified by the absence of gluten in red sorghum flour. The absence of gluten functionality in red sorghum just like millet restricts the high level of incorporation of red sorghum flour in composite bread formula. In fact, gluten, starch and water are major components of bread crumb [13]. Gluten proteins are essential for structure building, as they form a network in the solid matrix and allow gas retention and expansion, which improve the bread volume [13]. To overcome this, EPS-producing LAB (A16) was used to produce EPS during the fermentation of red sorghum dough. In fact, EPS can act as a hydrocolloid in bread making to mimic gluten properties [13,17]. The flours characteristics obtained from alveograph in this study were superior to those found (P= 54, mmH_2O, L= 36 mm and W= 76 10-4J) by [34] who produced bread with the incorporation rate of 30% of sorghum flour.

It also emerged from this study that the pH value decreased significantly ($p<0.05$) with the increase in the incorporation rate of red sorghum sourdough. This leads to an increase in the acidity content. This could be explained by the fact that during the fermentation of red sorghum dough by the LAB strain A16 for the production of EPS, there was acidification leading to a decrease in pH. These results corroborate those of [35] and [13]. Indeed, LAB are well known to produce antimicrobial molecules such as organic acids (lactic, acetic, formic and caproic phenolic), carbon dioxide, hydrogen peroxide, ethanol and bacteriocins during fermentation [36]. As the amount of red sorghum sourdough used for substitution of wheat flour increased with the incorporation rate, the titratable acidity increased and the pH decreased. Moisture content decreased with the increasing of incorporation rate, while dry matter content increased. This would probably be due to red sorghum flour which has low water absorption like millet flour compared to wheat flour [4,7]. These results corroborate those of [13] who found that replacing wheat flour with 50% unfermented millet flour significantly reduced the Farinograph water absorption. The decrease in moisture content of composite bread could also be attributed to the denaturation of protein which resulted into more interactions between proteins and polysaccharides through electrostatic forces [37]. This led to intermolecular network, water entrapment of water and lower free water content which is associated with decreased moisture content in foods [37]. The fiber content increased with the incorporation rate of red sorghum flour, in particular at 30% incorporation rate. An explanation of this result is due to the fact that the red sorghum flour was obtained from whole red sorghum grains which are therefore rich in dietary fiber [2,1]. These results are similar to those of [1] who found a crude fiber content of 2.0% in sourdough bread from sorghum flour; and those of [38] who found a crude fiber content between 1.76 to 2.91% in bread consisting of wheat bread enriched with rice bran. These results are also in line with those of [7] who studied the production of composite bread based on wheat and millet flour. The red sorghum fibers like whole millet fibers are also helpful in reducing the risk of gall stones. Food rich in insoluble fibers can speed up the transit of undigested food through the colon and also reduce the secretion of bile acids which prevents the formation of gall stones [39]. As a low glycemic index, insoluble fibers help in the slow releases of carbohydrates into human organism thus proving energy over a longer period and keeping blood glucose under check [40]. The results highlighted also an increase in protein and fat contents. An increase in the incorporation rate of red sorghum sourdough in the composite bread increased the protein and fat content. This variation of the protein contents may be due to the proteolytic activity of the fermenting strains [41]. [1] showed that the moisture content, crude protein, crude fat, crude fiber, and ash in the sourdough bread samples generally improved when compared with the raw material (sorghum flour) used for the baking. However, a decrease in the carbohydrate content related to an increase in the incorporation rate of millet flour was observed. These results could be explained by the fact that red sorghum flour was added as a fermented dough. During fermentation, lactic acid bacteria use a part of the carbohydrates to produce lactic acid resulting in a decrease of carbohydrate content and an increase of the acidity of the composite bread. These relative low carbohydrate contents could be an advantage for the consumption of this type of bread by diabetics. The results of the energy value are higher than those found by [40] during the production of composite bread based on wheat-millet flour with different incorporation rates (379.55 Kcal/100g) and wheat bread without added millet flour (Normal Bread: 312.7 Kcal/100g). An increase in the content of mineral elements (Fe, K, Zn and Mg) was observed in relation to the increase in the rate of incorporation of red sorghum flour. As for the other mineral elements, bread being produced with whole red sorghum flour, this would explain the fact that the content of these mineral elements increases with the incorporation rate of red sorghum flour. Indeed, the germ fraction of red sorghum is rich in minerals [2]. Sourdough bread from sorghum flour production, [1] showed that the bread samples had good mineral composition which indicates that the fermentation had positive influence on the sourdough. Six types of free amino acids have been found, including four essentials amino acids (Val, Met, Ile, and Lys). The proportions of amino acids in the composite bread were relatively low. This could be due to the fact that red sorghum, like other cereals, has some limitations due to its low content in protein and some essential amino acids, such as lysine [42]. Amino acids are important biological components needed in the human body for biosynthesis, neuro-transmission and other metabolic activities [43]. The sensory evaluation results showed that the color of most of the composite breads was brown and golden, gradually becoming darker with the increase in incorporation rate of red sorghum flour. These results corroborated those of [1] in sourdough bread production from sorghum flour, who showed that the taste panel members commented that the sourdough looked like cake because of its dark brown appearance, crumb and crust. This darkening could be explained by the effect of heat leading to the billion reaction in the presence of amino acids, reducing sugars and humidity levels during cooking [44]. The colour resulted from the use of red variety of sorghum for the flour processing. The brownish bread appearance could also be related to the increase in fibre content [45]. The consumers found that breads samples presented a good aroma. The main compounds that influenced breads aroma are volatile organic compounds derived from the fermentation and baking stages [34]. The production of EPS in the fermented red sorghum dough improved the texture of the mixed bread, making it slightly crispy and less brittle. Several studies on composite and whole grain breads have confirmed the usefulness of dextran in improving the technological and sensory quality of composite breads [46,47]. The composite bread obtained with the 30% incorporation was the most appreciated by the panelists because of its texture, its slightly acidic taste and a good feeling in the mouth.

5. Conclusion

The results found in this study showed that the baking strength of the composite flours decreases considerably with increasing levels of red sorghum flour incorporation. The physico-chemical and sensory characteristics of the composite bread revealed that the use of sorghum and millet malt as natural improvers in bread-making contributed to improve the availability of macronutrients and micronutrients. Overall, the results of the sensory evaluation showed that the composite bread samples presented a good aroma, a good texture, a slightly acidic taste and a pleasant mouthfeel. The 30% incorporation was the most appreciated by the panelists. The production of composite bread based on wheat-red sorghum, using local cereal malts as a natural improver, could be an alternative to replace imported improvers in the production of bread. This could contribute to reducing the importation of wheat and improvers in developing countries and an increase in the income of producers through the increase in the production of local cereals such as red sorghum which will be used in the production of bread to replace wheat.

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Statement of Competing Interests

The authors have no competing interests.

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