

Effect of sorghum pre-processing (roasting and germination) on the replacement level and quality of sorghum-wheat bread: bread characteristics, digestibility, consumer acceptability and microbiological analysis

Vijay Singh Sharanagat¹ · Prabhat K. Nema¹  · Lochan Singh² · Saravanan Mani³ · Anand Kishore¹

Revised: 15 July 2023 / Accepted: 5 August 2023 / Published online: 27 September 2023
© Association of Food Scientists & Technologists (India) 2023

Abstract The present study was focused on the replacement of refined wheat flour (RWF) by control (CS) and processed sorghum flour [germinated (GS) and roasted (RS)] on the properties of flour/batter/dough (particle size, XRD, pasting, dynamic rheology, farinograph) and bread (physical, textural, digestibility, microbiological and sensory). Prominent variations adhered with sorghum processing, but decreasing patterns occurred for flour–water absorption, dough stability times, storage modulus, peak/final/break-down viscosities, bread-moisture content, specific volume,

porosity, and lightness. Flour's pasting temperature, dough development time, breadbulk density, hardness, gumminess, and bitterness increased. Composite flours mainly had weak nature compared to RWF. The baking loss was lower for 10–30% CS and GS incorporation than RS. Composite bread had higher in-vitro protein and starch digestibility (CS > GS > RS) than RWF. Three days storage life with acceptable quality scores was obtained for bread with CS and GS up to 20% and RS up to 30% incorporation.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s13197-023-05810-3>.

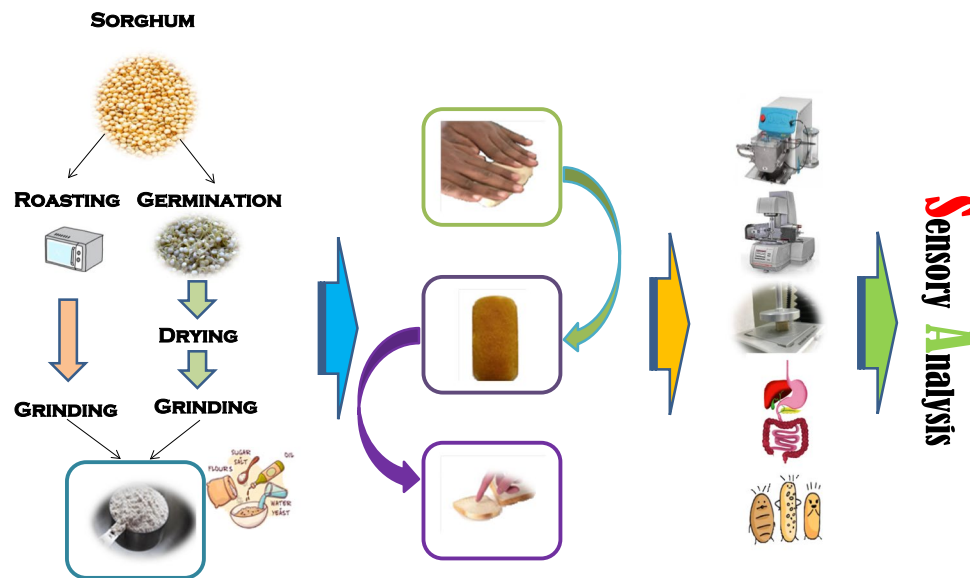
✉ Prabhat K. Nema
pknema@yahoo.co.in

¹ Department of Food Engineering, NIFTEM, Sonapat, Haryana 131028, India

² Contract Research Organization, NIFTEM, Sonapat, Haryana 131028, India

³ Department of Basics and Applied Sciences, NIFTEM, Sonapat, Haryana 131028, India

Graphical abstract



Keywords Low-gluten bread · Sorghum · Digestibility · Shelf-life

Abbreviations

| | |
|-----|------------------------|
| RWF | Refined wheat flour |
| CS | Control sorghum |
| GS | Germinated sorghum |
| RS | Roasted sorghum |
| RF | Rice flour |
| DDT | Dough development time |

Introduction

Bread is considered as a staple food, and refined wheat flour is the most commonly used flour for the preparation of bread. The gluten protein in wheat plays an important role in imparting desired structure, rheological behavior, and quality to the bread (Millar et al. 2019). However, a large proportion of the population suffers from gluten allergy resulting in severe and adverse health effects, which challenges researchers to look for suitable alternatives. One of the promising alternatives is a partial and complete replacement of wheat flour with flours of different food grains (cereal, legumes and pulses) to develop bread (Olojede et al. 2020; Marchini et al. 2021). However, the development of suitable bread from these alternative flours is also challenging as the non-wheat flour incorporation weakens the dough and adversely affects the bread quality, for example, specific volume is reduced, hardness increases, bread becomes denser etc. (Collar et al. 2014). To mitigate these drawbacks, different hydrocolloids and

starches known to improve the structural properties are introduced into the formulations, but reports suggest that hardness still increases and no significant enhancement was observed on specific volume (Coronel et al. 2021). Hence, it may be derived that the success achieved in supplementing the refined wheat flour in bread is currently limited (Mondor et al. 2014).

Sorghum is a resilient C4 crop that originated in North-eastern Africa and has become an important staple food for nearly 500–750 million people in the semi-arid tropics of Asia, Africa, and Latin America (ICRISAT 2018; Gebre et al. 2021). It is the fifth most important crop grown for its diverse economic uses, such as food, feed, and production of biofuels/ethanol (Raju et al. 2021), with approximately 35–42% accountability as food (Sharanagat et al. 2019; Gebre et al. 2021) and rest for fuel and feed. It is gluten-free in nature and rich in macronutrients, micronutrients, and bioactive compounds (Rashwan et al. 2021). Owing to its nutritional benefits and slow digestibility, the grain has become a promising food for people suffering from obesity, diabetes, and autoimmune allergic reactions (Rashwan et al. 2021). A huge potential thus lies for the food processing sector for the development of functional and low-gluten food (especially bread) from sorghum for the celiac population.

Studies on the development of bread (composite/gluten-free) from sorghum have mainly focussed on utilizing unprocessed grain flour (Olojede et al. 2020). However, a very few studies have reported modification of the sorghum flour properties before developing bread. The dimensions of flour

processing include extrusion (Jafari et al. 2017), thermal and hydrothermal (Torbica et al. 2021), germination (Phattanakulkaewmorie et al. 2011), fermentation (Ogunsakin et al. 2015), irradiation (Almaiman et al. 2021), etc. As observed, only a few studies evaluated the effect of germinated sorghum flour on dough and bread quality. Additionally, to the best of our knowledge, no study has reported using roasted sorghum flour to replace refined wheat flour or to develop bread. Thus, future research may be warranted to fulfill these highlighted gaps.

In general, it has been observed that roasting and germination alter the chemical composition of grain to reduce the anti-nutritional factor and increase the digestibility (Suhag et al. 2021; Campos-Vega et al. 2010) while offering positive effect on functional and antioxidant properties (Ouazib et al. 2016; Jogihalli et al. 2017). These modifications are attributed to the heat involvement in the roasting process (facilitating the inactivation of certain heat-labile anti-nutritional compounds) (Ouazib et al. 2016; Jogihalli et al. 2017) and enzyme-based breakdown of macronutrients in germination process (Kumar et al. 2020). Microwave roasting is considerably fast, requires less space, and saves approximately 25% more energy than the conventional oven (Nirmaan et al. 2020). The initial cost of a microwave oven is higher compared to a conventional oven, whereas the operational cost is low compared to the conventional oven method.

Despite these processes' advantages, studies on the utilization of modified sorghum flour by these processes have not been well conducted. Thus, the present study aimed to substitute refined wheat flour with germinated and roasted sorghum flour to develop functional low-gluten composite bread. The study will thus mitigate the existing gap by determining the effect of the incorporation of germinated and roasted sorghum flour on the dough and composite bread properties.

Material and method

Material

Sorghum grains (HJ-513) were procured from Hisar Agriculture University (Haryana, India). Refined wheat flour (RWF), rice flour (RF), salt, sugar, yeast (dry), and seed oil was purchased from a local market in Kundli, Haryana, India.

Processing of sorghum

Sorghum grains (HJ-513) were collected and cleaned for foreign impurities and broken parts. Cleaned grains were divided into three equal parts. The first part was soaked in water for 12 h and germinated at room temperature for 3 days, and dried in a hot air dryer (40 °C) for 24 h, second part was roasted using a laboratory microwave at 450 W for 10 min (Sharanagat et al. 2019) and third part was kept as control (unprocessed). The germinated (GS), roasted (RS), and control sorghum grains (CS) were ground and passed through 212-micron sieves and were kept in aluminium pouches until further analysis. Flour properties like particle size and XRD and composite flour properties like pasting and dynamic rheological analysis have been performed and explained in supplementary text 1.

Nomenclature of flours

Since sorghum flours were mixed with RWF and RF in different combinations to develop composite bread, different nomenclature was used for their identification. The same nomenclature has been used throughout the text as follows: RWF—Refine wheat flour, RF—Rice flour, CS—Control (untreated) sorghum flour, GS—Germinated sorghum flour, RS—Roasted sorghum flour. The 10, 20, 30, 40, and 50

Table 1 Composite bread formulation

| Ingredients | %Composition/100 g | | | | | | | | | | | | | |
|-------------|--|------|------|------|------|------|------|------|------|------|------|------|------|--|
| | RWF100 | CS10 | CS20 | CS30 | CS40 | GS10 | GS20 | GS30 | GS40 | RS10 | RS20 | RS30 | RS40 | |
| RWF | 100 | 80 | 70 | 60 | 50 | 80 | 70 | 60 | 50 | 80 | 70 | 60 | 50 | |
| CS | 0 | 10 | 20 | 30 | 40 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | |
| GS | 0 | 0 | 0 | 0 | 0 | 10 | 20 | 30 | 40 | 0 | 0 | 0 | 0 | |
| RS | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 10 | 20 | 30 | 40 | |
| RF | 0 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | 10 | |
| Yeast | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 | 3 | |
| Oil | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | 2 | |
| Sugar | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | 5 | |
| Salt | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 | |
| Water | As per the requirement approx 5% less as predicted by Farinograph analysis | | | | | | | | | | | | | |

RWF refine wheat flour, CS control sorghum flour, GS germinated sorghum flour, RS roasted sorghum flour, RF rice flour

associated with nomenclature represents the percentage of particular flour in composite flour.

Farinograph analysis

Farinograph analysis of the mixture of different flours (RWF, RF, CS, GS, and RS) without yeast and other ingredients was performed using a dough lab (DoughLAB 2500, Perten, Australia). The flour mixture (300 g, Table 1) was used with the fixed initial water content (50%). The instrument was allowed to add water (drop-wise) during analysis as per the requirement. Water required and dough development time was then analysed from the developed Farinograph.

Bread development and analysis

Breads were formulated by keeping RF constant to 10% and mixing CS, GS, RS, and RWF in different combinations (Table 1). Prepared dough samples were kept in the mould (oiled-1 mL) and allowed to ferment for 45 min at 35 °C followed by baking at 180 °C for 25–30 min (Electrical oven, PEO-1, Power-3.01 kW and Voltage-220 V). Prepared bread samples were allowed to cool at room temperature for 3 h before proceeding with further analysis.

Physical properties of bread

Water activity (aw) and moisture content of samples were determined through the water activity (aw) meter (Aqua LAB 4TE, USA) and AOAC (2006) standard method,

respectively. The baking loss was calculated by the difference in the weight of the loaf before and after baking. The ratio of volume (determined through the rape-seed displacement method) to mass ratio was used for calculating the specific volume (cm³/g) of bread loaf (AACC 2000). Density of sample was determined by the ratio of mass and volume. Apparent density of bread was measured using pycnometer using toluene displacement method and porosity was estimated as a ratio of bulk density and apparent density (Rahman et al. 2005).

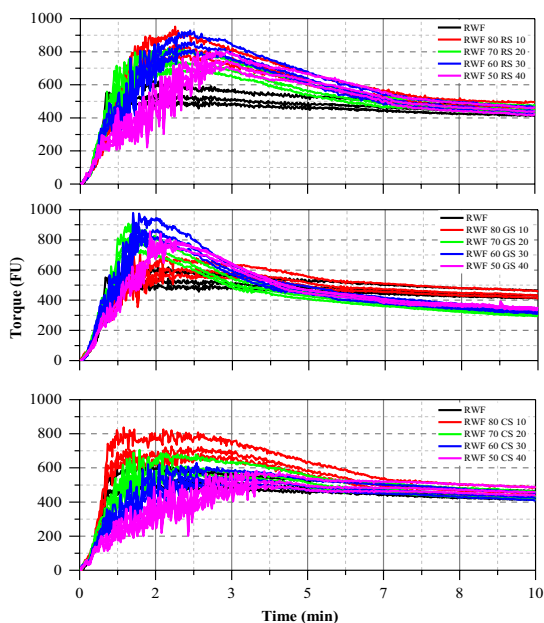
The color of the developed breads’ crust and crumb was analyzed using a handheld colorimeter (KONICA MINOLTA, CR-400, Japan). Browning Index (BI) was calculated using Eq. 1 (Dhua et al. 2021).

$$BI = \frac{100 \times (X - 0.31)}{0.17} \tag{1}$$

where X = $\frac{(a+1.75L)}{(5.64L+a-3.01b)}$, L: brightness, a: redness, b: yellowness.

Textural properties of bread

A crumb sample (20 mm × 20 mm) was cut from the center of the slices to obtain uniform slices of 12 ± 1 mm thickness. Texture profile analysis was performed using Texture Analyzer (Stable Microsystems, TA-HD Plus C, UK) equipped with a compression probe (75 mm diameter), by two sequential compression events under the following conditions: pre-test speed—1 mm/s, test



| Combination | Water used (ml/100g) | Development time (min) | Peak (FU) | Softening (FU) | Stability (min) |
|--------------|----------------------|------------------------|-----------|----------------|-----------------|
| RWF | 65.29 | 1.8 | 533.1 | 61.2 | 4.7 |
| RWF 80 CS 10 | 62.45 | 1.9 | 661.1 | 174.2 | 3.0 |
| RWF 70 CS 20 | 61.47 | 2.3 | 606.7 | 134.1 | 3.0 |
| RWF 60 CS 30 | 59.57 | 3.0 | 544 | 82.8 | 3.2 |
| RWF 50 CS 40 | 58.22 | 4.3 | 522.9 | 51.2 | 4.6 |
| RWF 80 GS 10 | 59.52 | 1.3 | 790 | 385.9 | 1.1 |
| RWF 70 GS 20 | 58.87 | 1.4 | 773.5 | 382.8 | 1.0 |
| RWF 60 GS 30 | 59.72 | 1.7 | 845.9 | 444.8 | 1.0 |
| RWF 50 GS 40 | 59.84 | 2.0 | 785.7 | 385.9 | 1.2 |
| RWF 80 RS 10 | 64.62 | 2.3 | 828.9 | 318.5 | 1.9 |
| RWF 70 RS 20 | 62.12 | 2.2 | 739.8 | 249.4 | 2.3 |
| RWF 60 RS 30 | 63.94 | 2.5 | 845.7 | 351.4 | 1.6 |
| RWF 50 RS 40 | 62.72 | 3.2 | 737.7 | 261.7 | 1.4 |

Fig. 1 Farinograph analysis of composite flour (RWF refine wheat flour, CS control sorghum, GS germinated sorghum, RS roasted sorghum)

speed—1 mm/s, post-test speed—2 mm/s, target mode—strain (50%) and trigger force—5 g. Parameters such as hardness, chewiness, springiness, and cohesiveness were calculated from the obtained data profile.

In-vitro protein digestibility of bread

The in-vitro protein digestibility of the bread samples was determined by the method described by Cornejo et al. (2015). The detailed experimental process has been given in supplementary text 2. The percent protein digestibility (Y) was calculated by using Eq. 2.

$$Y = 210.464 - 18.1x \quad (2)$$

where x is the change in pH after 10 min.

In-vitro starch digestibility of bread

The starch digestibility of the bread sample was determined using the method described in the STA20kit (Sigma). The detailed experimental process has been given in supplementary text 2.

Microbiological analysis and shelf life analysis

IS 5402 (2012) method was utilized to determine the total mesophilic count during the storage of the bread. Details methodology has been explained in supplementary text 2.

Sensory analysis

The sensory analysis of the bread was performed by the method described by Nasir et al. (2020). Details methodology has been explained in supplementary text 2.

Statistical analysis

All the analysis was performed in triplicate, and data was presented as mean \pm standard deviation. All the graphs were prepared in Origin Pro 2007, and data was statistically analysed using SPSS.

Results and discussion

Farinograph analysis

Figure 1 shows the farinograph attributes of RWF along with RWF composite flours possessing RF, CS, GS, and RS. The highest requirement of water was noted for the RWF flour in

comparison to other composite flours, while the lowest value was recorded for RWF50CS40 composite flour. The reduction in the water absorption capacity in composite flours over RWF was found to be 4.3–10.82% with CS, 8.34–9.83% with GS, and 1.02–4.85% with RS. This reduction observed for water absorption in composite flours over RWF may be attributed to the dilution of hydrophilic gluten content, mainly responsible for networking and cross-linking, and an increase in lipid and hydrophobic or endospermic (kaffarins) protein upon sorghum addition. The reduction in water absorption with an increase in millet flour was also reported by Sharma and Gujral (2019). Also, more water requirement in RS composite flours than the GS composite flours may be due to the higher loss of water molecules in RS due to involved heat treatment and damaged starch structure, which otherwise is absent in GS, where grains are first soaked in water for a stipulated time period followed by germination and drying.

The dough development time (DDT) for the RWF was found to be 1.8 min. Whereas the DDT increased with an increase in concentration of sorghum and among the composite flours, the highest and the lowest development time was observed for replacement levels of 40% and 10% of CS and GS, respectively. Incorporation of CS, GS, and RS led to 1.05–2.38, 0.7–0.9, and 1.2–1.7 fold change in DDT, respectively, i.e., higher DDT was observed for replacement with CS than RS and GS. Thus, there were some composite flours (RWF80GS10, RWF70GS 20, and RWF60GS30) whose development time was far lower to that of RWF. These changes in the DDT with respect to the form of sorghum used may be associated with the change in the protein and damaged starch content. It has been stated that protein hydration is largely affected by the presence of damaged starch, which bounds water molecules strongly while leaving lesser molecules for protein interaction. As a result, dough development takes longer time due to affected protein linkages and networking.

The flour strength may also be depicted through stability times whereby longer stability time duration shows flours suitability to hearth (oven based with no pan)/variety bread development and at times the higher duration of mixing. This factor is found to be dependent on the chemical composition of glutenins and on the presence/absence of cellulosic fiber. The trend of stability times (in min) in the present study was as follows: RWF (4.7) > RWF + CS (3.0–4.6) > RWF + RS (1.4–2.3) > RWF + GS (1.0–1.2). This suggests that RWF and all combinations of RWF + CS are better suitable for the development of hearth bread (Sourdough, French, Ciabatta, and Italian) where lean formulas are used (with few or no enriching ingredients), the loaf is placed directly on a hot surface (without pan) and the shape/size of bread is controlled. It may also be suggested that the GS and RS fall

Table 2 Effect of addition of sorghum flour on physical and textural properties of composite bread

| Sample (%) | Physical properties | | | | | Textural properties | | | | | | |
|------------|------------------------------|----------------------------|-----------------------------|---------------------------|----------------------------|----------------------------|---------------------------|------------------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| | MC | a_w | BL | SV | BD | AD | P | Hardness (N) | Cohesiveness | Springiness | Gumminess (N) | Chewiness (N) |
| RWF 100 | 34.83 ± 0.08 ^a | 0.95 ± 0.02 ^{ab} | 11.74 ± 0.15 ^{abc} | 3.15 ± 0.02 ^a | 0.32 ± 0.00 ^a | 0.68 ± 0.02 ^a | 0.53 ± 0.01 ^a | 7.60 ± 0.35 ^a | 0.64 ± 0.01 ^a | 0.82 ± 0.21 ^{ab} | 4.90 ± 0.27 ^a | 4.01 ± 0.80 ^a |
| RWF 80 | 34.71 ± 2.47 ^a | 0.95 ± 0.03 ^{ab} | 9.95 ± 1.49 ^a | 2.16 ± 0.01 ^{bc} | 0.46 ± 0.00 ^{bcd} | 0.79 ± 0.04 ^{abc} | 0.42 ± 0.02 ^{ab} | 13.34 ± 0.55 ^{abcd} | 0.56 ± 0.00 ^b | 1.02 ± 0.12 ^{bc} | 7.43 ± 0.28 ^{ab} | 7.53 ± 0.63 ^{ab} |
| CS 10 | 33.51 ± 0.69 ^{ab} | 0.95 ± 0.01 ^{ab} | 9.55 ± 2.43 ^a | 1.83 ± 0.03 ^d | 0.55 ± 0.01 ^{de} | 0.87 ± 0.03 ^{bcd} | 0.37 ± 0.03 ^{ab} | 16.18 ± 0.56 ^{bcd} | 0.57 ± 0.01 ^b | 0.85 ± 0.04 ^{ab} | 9.28 ± 0.14 ^{ab} | 7.86 ± 0.26 ^{ab} |
| CS 20 | 33.62 ± 0.22 ^{ab} | 0.95 ± 0.04 ^{ab} | 9.89 ± 0.43 ^a | 1.46 ± 0.02 ^c | 0.69 ± 0.01 ^f | 0.97 ± 0.01 ^{cd} | 0.29 ± 0.02 ^b | 18.60 ± 0.30 ^{cd} | 0.47 ± 0.02 ^{de} | 0.84 ± 0.03 ^{ab} | 8.74 ± 0.26 ^{ab} | 7.32 ± 0.46 ^{ab} |
| CS 30 | 30.72 ± 0.35 ^{abcd} | 0.94 ± 0.02 ^{ab} | 12.48 ± 0.19 ^{abc} | 1.38 ± 0.29 ^c | 0.74 ± 0.15 ^f | 1.00 ± 0.02 ^d | 0.26 ± 0.14 ^b | 21.09 ± 0.69 ^d | 0.46 ± 0.04 ^{de} | 0.75 ± 0.05 ^a | 9.75 ± 1.11 ^{ab} | 7.30 ± 0.37 ^{ab} |
| RWF 80 | 29.63 ± 1.91 ^{bcd} | 0.91 ± 0.01 ^{cd} | 10.48 ± 0.04 ^{ab} | 2.61 ± 0.01 ^f | 0.38 ± 0.00 ^{ab} | 0.70 ± 0.02 ^{ab} | 0.45 ± 0.01 ^{ab} | 8.82 ± 1.85 ^{ab} | 0.53 ± 0.02 ^{bc} | 0.71 ± 0.01 ^a | 4.70 ± 1.15 ^a | 3.34 ± 0.86 ^a |
| GS 10 | 27.91 ± 0.99 ^{cd} | 0.93 ± 0.05 ^{bcd} | 10.33 ± 1.16 ^{ab} | 2.07 ± 0.00 ^{bd} | 0.48 ± 0.00 ^{bcd} | 0.75 ± 0.22 ^{ab} | 0.32 ± 0.20 ^b | 10.66 ± 0.70 ^{bcd} | 0.45 ± 0.00 ^e | 1.12 ± 0.08 ^c | 4.74 ± 0.26 ^a | 5.34 ± 0.68 ^{ab} |
| GS 20 | 26.37 ± 3.01 ^{de} | 0.92 ± 0.01 ^{bcd} | 10.98 ± 0.97 ^{abc} | 1.56 ± 0.04 ^c | 0.64 ± 0.02 ^{ef} | 0.95 ± 0.05 ^{cd} | 0.32 ± 0.02 ^b | 17.95 ± 0.07 ^{cd} | 0.39 ± 0.03 ^f | 0.70 ± 0.10 ^a | 7.00 ± 0.43 ^{ab} | 4.95 ± 0.98 ^a |
| GS 30 | 25.3 ± 0.78 ^c | 0.9 ± 0.04 ^d | 13.52 ± 3.01 ^{ab} | 1.54 ± 0.01 ^c | 0.65 ± 0.00 ^f | 0.94 ± 0.02 ^{cd} | 0.31 ± 0.01 ^b | 20.23 ± 0.52 ^d | 0.34 ± 0.01 ^f | 0.41 ± 0.01 ^d | 6.93 ± 0.08 ^{ab} | 2.86 ± 0.04 ^a |
| GS 40 | 28.12 ± 0.12 ^{cde} | 0.95 ± 0.01 ^{ab} | 12.47 ± 2.25 ^{abc} | 2.63 ± 0.01 ^f | 0.38 ± 0.00 ^{ab} | 0.69 ± 0.01 ^{ab} | 0.45 ± 0.01 ^{ab} | 19.20 ± 2.39 ^{cd} | 0.66 ± 0.01 ^a | 0.83 ± 0.11 ^{ab} | 12.64 ± 1.82 ^b | 10.41 ± 0.17 ^b |
| RWF 80 | 29.59 ± 0.18 ^{bcd} | 0.94 ± 0.03 ^{abc} | 14.33 ± 1.24 ^c | 2.31 ± 0.10 ^c | 0.43 ± 0.02 ^{bc} | 0.71 ± 0.05 ^{ab} | 0.39 ± 0.02 ^{ab} | 32.94 ± 0.69 ^e | 0.62 ± 0.02 ^a | 0.84 ± 0.01 ^{ab} | 20.51 ± 1.22 ^c | 17.24 ± 0.75 ^c |
| RS 10 | 29.24 ± 0.01 ^{bcd} | 0.94 ± 0.02 ^{ab} | 12.38 ± 0.49 ^{abc} | 2.14 ± 0.01 ^{bc} | 0.47 ± 0.00 ^{bcd} | 0.72 ± 0.01 ^{ab} | 0.36 ± 0.01 ^{ab} | 41.95 ± 3.95 ^f | 0.56 ± 0.01 ^b | 0.78 ± 0.02 ^a | 23.52 ± 1.98 ^c | 18.33 ± 1.07 ^c |
| RS 20 | | | | | | | | | | | | |
| RS 30 | | | | | | | | | | | | |

Table 2 (continued)

| Sample (%) | Physical properties | | | | | Textural properties | | | | | | |
|------------|-----------------------------|--------------------------|----------------------------|---------------------------|---------------------------|----------------------------|--------------------------|----------------------------|---------------------------|---------------------------|---------------------------|---------------------------|
| | MC | a _w | BL | SV | BD | AD | P | Hardness (N) | Cohesiveness | Springiness | Gumminess (N) | Chewiness (N) |
| RWF | 31.93 ± 0.43 ^{abc} | 0.96 ± 0.01 ^a | 10.72 ± 0.98 ^{ab} | 1.89 ± 0.10 ^{de} | 0.53 ± 0.03 ^{cd} | 0.79 ± 0.15 ^{abc} | 0.32 ± 0.16 ^b | 50.31 ± 11.90 ^g | 0.51 ± 0.04 ^{cd} | 0.71 ± 0.07 ^{ab} | 25.97 ± 8.02 ^c | 18.81 ± 7.60 ^c |
| 50 | | | | | | | | | | | | |
| RS | | | | | | | | | | | | |
| 40 | | | | | | | | | | | | |

RWF refine wheat flour, CS control sorghum flour, GS germinated sorghum flour, RS roasted sorghum flour, MC moisture content (wet basis), a_w water activity, BS baking loss (%), SV specific volume (cm³/g), BD bulk density (g/cm³), AP apparent density (g/cm³), P porosity (%)

#Mean ± standard deviation of three replications. The value with different superscript a, b, c, d, e, f in a row is significantly (*p* < 0.05) different

under the category of weak-medium flours and thus needs proper mixing with strong/very strong flours.

It was observed that the RWF and RWF50CS40 had a degree of softening below 80 Farinograph units (FU) which showed their strong nature, whereas RWF60CS30 had a value 82.8 FU, demonstrating weak-medium nature. On the other hand, all other composite flours had a degree of softening beyond 80 FU, demonstrating their weak nature. The higher degree of softening in composite flour might be due to the dilution of gluten content responsible for cross-linking and networking in RWF (Mekhael 2005). The increase in the degree of softening was also reported by El-Taib et al. (2018) with the addition of barley flour. The results of the present study are in accordance with other published literature. Abdelghafor et al. (2013) reported a decrease in the stability time, DDT, water absorption, and farinograph quality number upon the addition of sorghum flour to winter wheat. Similar results have been reported for sorghum addition in bread by Kulamarva et al. (2009). Contrarily, Jafari et al. (2017) reported an increase in water absorption and DDT and a decrease in stability time for extruded sorghum-wheat composite dough.

Bread properties

Water activity and moisture content

Bread moisture is an important parameter as it aids in keeping the product moistened and lubricated while regulating the crumb-firming process. Higher moisture content signifies (a) higher gelatinization of starch and (b) early crust formation, which prevents moisture from evaporation (Ibrahim et al. 2020). As observed from farinograph and pasting properties above, the concentration of gelatinized starch differed in the derived composite flours. Also, the crust color differed (darkening shows crusts’ exposure to heat for longer duration and early formation) with the type of sorghum flour used (processed/un-processed) and the amount of sorghum flour used for the development of composites bread. Hence the moisture content of the developed breads varied, with RWF bread possessing the highest value (34.83) while reduction occurred from 34.71 to 30.72 and 29.63 to 25.3 when the concentration of CS and GS sorghum increased from 10 to 40% in composite flours (Table 2). On the other hand, RS-based composite bread possessed less moisture content than RWF bread, but as the concentration of sorghum flour increased, the moisture content also increased.

The crispiness of food products is especially affected by the water activity of the product, whereby increased moisture content leads to the loss of crispiness. It is also defined as the brittleness and hardness of a food product. It was observed that the water activity of RWF, CS, and RS-based composite bread were at par with each other

and did not differ significantly, while the reduction was reported in GS-based composite bread. This reduction may be attributed to the change in the protein-carbohydrate interactions as observed through the pasting properties of flour. It may thus be postulated that the crispiness of these breads will increase. Khating et al. (2014) reported a reduction in moisture content from 37.80 to 36.10% of sorghum and wheat composite bread with an increase in the concentration of sorghum from 0 to 25%. Angioloni and Collar (2012) reported a moisture content in the range of 33.2–33.6 g for the bread prepared by the replacement (40%) of wheat flour with control and high hydrostatic pressure treated sorghum flour. They reported that the reduction in moisture content with an increase in sorghum concentration might be due to the increase in solid matter.

Baking loss and specific volume

Baking loss refers to a kind of technological loss whereby bread mass decreases during the baking time (Rakcejeva et al. 2011). Baking loss is a function of initial baking time, water evaporation from bread, water retention through glucans, water activity, presence of additives, and wheat fiber (Rühmkorf et al. 2012). As observed from Table 2, the significant variation over RWF for baking loss occurred in RWF70RS20. It was observed that the incorporation of 10–30% of CS and GS in RWF led to a reduction in baking loss, while an increase in RWF occurred when 40% supplementation was done. On the other hand, the incorporation of RS at 10–30% enhanced the baking loss, while a 40% reduction over RWF occurred. The trend of baking loss may be connected well with the recorded moisture content for the composite bread flours, as more evaporation and water activity favor high baking losses (Rakcejeva et al. 2011). Water movement from the bread center to the surface got restricted due to the formation of hard texture during the initial baking process and also favors a reduction in baking loss. The other reason for high baking losses is the decrease in wheat fiber content in composite flours.

Specific volume is another technical indicator of bread that has an immense effect on consumers' choice, the cut point being set as 3.5–5.5 cm³/g for wheat bread by industries. It is related to water retained in the gluten network and the gas retention capacity of gluten strands during fermentation and dough proofing. Denser dough with low specific volume is less preferable by consumers owing to unpleasant flavor/taste, difficulty in chewing, and higher moisture content. The specific volume of bread reduces when amylose content is low in flour, small bubbles are formed, and the mass fails to hold the gas/air bubbles during

cooking—'limiting consistency' (Monteiro et al. 2021). It is also stated that additives, proteins, enzymes, and hydrocolloids improve specific volume in gluten-free bread by making a few large holes in the crumb. However, in the present study, reduction was observed in a specific volume of composite breads over RWF, which might be due to the lack of gluten in sorghum flour, limiting consistency, and absence of any additives/hydrocolloids. The reduction increased when the concentration of CS, GS, and RS flour increased in the composite flours. A 0.43–0.68, 0.48–0.82, and 0.6–0.83fold changes were noted in specific volumes of CS, GS, and RS-based composite flours, respectively.

Apparent density, bulk density, and porosity

The porosity of bread is another technological parameter. It was observed that RWF bread had the highest porosity (0.53) which was reduced upon the addition of sorghum flour for composite bread development (Table 2). Moreover, the reduction was continuous upon an increase in the sorghum flour concentration in composite bread, amounting to 50.94%, 41.51%, and 39.62% in CS, GS, and RS-based bread, respectively. The highest porosity in RWF is attributed to its gluten-favored better gas- and water vapor-retention capacity, favoring the formation of larger pores. On the other hand, composite bread had reduced gluten content as sorghum flour lacks gluten. Because of this, there was a reduction in bubble holding capacity, which was followed by the collapse of dough boundary pores and thus reduced the porosity.

The bulk density of the bread is an important parameter that also affects the packaging design of the product. The lowest bulk density was observed for the RWF bread, while the continuous increase was recorded in all composite bread as the addition of sorghum changed from 10 to 40%. This increase was also related to the reduction in the specific volume and porosity of bread, as they are negatively correlated (Arufe et al. 2018). Contrary to bulk density, an increasing trend was observed in apparent density.

Color

Color parameters of bread developed with different treatments and varying compositions of sorghum flour are depicted in Table 3. It was observed that the lightness of the RWF bread (crust and crumb) was the highest, while the addition of sorghum flour (processed or unprocessed) reduced the lightness. Crust lightness was reduced by 0.75–0.9, 0.69–0.82, and 0.84–0.92 fold, respectively, when CS, GS, and RS composite flours were used. Similarly, crumb lightness was reduced by 0.87–0.97, 0.65–0.88, and 0.75–0.94 fold, respectively. With respect to crust a*

Table 3 Effect of addition of sorghum flour on color of composite bread

| Conditions | Compositions (%) | | | ΔE | BI | In vitro digestibility | | Mesophilic bacteria/aerobic bacteria (log cfu/g) | | | |
|--------------|---------------------------|-----------------------------|--|-----------------------------|-----------------------------|---------------------------|---------------------------|--|----------|-------------|--|
| | L* | a* | b* | | | Protein digestibility (%) | Starch digestibility (%) | Storage time (days) | | | |
| | | | | | | | | 0th day | 2nd days | 4th day | |
| <i>Crust</i> | | | | | | | | | | | |
| Control | 46.24 ± 2.28 ^a | 10.07 ± 0.97 ^{abc} | 17.53 ± 0.91 ^{ab} | – | – | 75.85 ± 0.07 ^a | 61.69 ± 0.01 ^a | 0.14 | 1.18 | Mold growth | |
| Raw | RWF 80CS10 | 45.05 ± 2.27 ^{ab} | 11.41 ± 0.86 ^{ab} | 19.1 ± 1.16 ^a | 2.55 ± 0.97 ^{ab} | 7.07 ± 0.26 ^a | 53.13 ± 0.05 ^b | Nil | 2 | | |
| | RWF70CS20 | 44.76 ± 2.55 ^{ab} | 10.45 ± 1.65 ^{abc} | 19.08 ± 0.66 ^a | 4.4 ± 2.01 ^{abc} | 6.99 ± 0.51 ^a | 55.54 ± 0.01 ^c | 0.4 | 1.85 | | |
| | RWF60CS30 | 40.82 ± 2.48 ^{cd} | 6.92 ± 0.29 ^d | 15.66 ± 1.84 ^{cd} | 6.64 ± 1.34 ^{bcd} | 5.84 ± 0.37 ^{bc} | 81.02 ± 0.73 ^c | 64.83 ± 0.04 ^d | 0.82 | 1.93 | |
| | RWF50CS40 | 35 ± 2.48 ^{ef} | 6.72 ± 1.5 ^d | 13.48 ± 0.98 ^e | 12.55 ± 1.78 ^{efg} | 6.02 ± 0.21 ^{cd} | 81.72 ± 0.06 ^d | 67.84 ± 0.05 ^e | 0.74 | 1.52 | |
| | RWF 80GS10 | 32.06 ± 1.28 ^f | 10.75 ± 0.21 ^{a^{bc}} | 10.31 ± 0.31 ^f | 15.95 ± 2.71 ^g | 6.09 ± 0.3 ^{cd} | 81.97 ± 0.02 ^d | 58.98 ± 0.04 ^f | 0.361 | 2.36 | |
| Germinated | RWF70GS20 | 33.71 ± 1.52 ^f | 10.71 ± 0.28 ^{abc} | 10.84 ± 1.24 ^f | 14.23 ± 4.37 ^{fg} | 5.96 ± 0.25 ^{cd} | 58.44 ± 0.04 ^g | 0.32 | 2.32 | | |
| | RWF60GS30 | 37.52 ± 1.85 ^{de} | 10.21 ± 1.33 ^{abc} | 13.35 ± 1.54 ^e | 9.84 ± 3 ^{def} | 6.13 ± 0.51 ^{cd} | 63.97 ± 0.02 ^h | 0.27 | 2.39 | | |
| | RWF50GS40 | 38.35 ± 1.55 ^d | 9.95 ± 0.9 ^{a^{bc}} | 13.98 ± 0.82 ^{de} | 8.76 ± 3.54 ^{cde} | 6.19 ± 0.14 ^{cd} | 83.91 ± 0.04 ^e | 0.43 | 2.43 | | |
| | RWF 80RS10 | 42.8 ± 0.96 ^{bc} | 11.77 ± 0.89 ^a | 16.81 ± 0.87 ^{bc} | 4.02 ± 2.24 ^{abc} | 6.73 ± 0.3 ^{ae} | 73.23 ± 0.04 ^f | 57.25 ± 0.01 ^j | 0.16 | 2.16 | |
| | RWF70RS20 | 42.58 ± 0.74 ^{bc} | 9.68 ± 1.07 ^{bc} | 16.78 ± 0.42 ^{bc} | 3.94 ± 2.95 ^{abc} | 6.42 ± 0.32 ^{de} | 71.94 ± 0.07 ^g | 59.21 ± 0.08 ^k | 0.88 | 1.78 | |
| Roasted | RWF60RS30 | 39.94 ± 1.49 ^{cd} | 9.02 ± 0.28 ^c | 14.2 ± 0.84 ^{de} | 7.28 ± 4 ^{bcd} | 5.82 ± 0.1 ^{bc} | 72.94 ± 0.05 ^f | 0.39 | 1.4 | | |
| | RWF50RS40 | 38.85 ± 0.77 ^d | 7.34 ± 0.36 ^d | 13.36 ± 0.46 ^e | 8.98 ± 2.86 ^{cde} | 5.39 ± 0.11 ^b | 70.9 ± 0.016 ^h | NIL | 1.23 | | |
| | Control | 66.52 ± 1.81 ^a | -0.14 ± 0.04 ^a | 12.9 ± 0.42 ^a | – | – | 75.85 ± 0.07 ^a | 61.69 ± 0.01 ^a | 0.14 | 1.18 | |
| | Raw | RWF 80CS10 | 64.81 ± 0.21 ^{ab} | 0.59 ± 0.07 ^b | 14.19 ± 0.41 ^{bc} | 2.5 ± 1.27 ^{ab} | 2.43 ± 0.08 ^a | 79.92 ± 0.05 ^b | Nil | 2 | |
| | | RWF70CS20 | 62.01 ± 0.44 ^c | 0.84 ± 0.06 ^b | 14.44 ± 0.19 ^{bc} | 4.92 ± 2.11 ^b | 2.63 ± 0.06 ^{ab} | 80.89 ± 0.67 ^c | 0.4 | 1.85 | |
| RWF60CS30 | | 58.87 ± 0.49 ^d | 1.25 ± 0.16 ^c | 14.82 ± 0.16 ^{cd} | 8.03 ± 1.79 ^c | 2.92 ± 0.07 ^{cd} | 81.02 ± 0.73 ^c | 64.83 ± 0.04 ^d | 0.82 | 1.93 | |
| RWF50CS40 | | 58.1 ± 0.89 ^d | 1.56 ± 0.16 ^{cd} | 15.2 ± 0.52 ^d | 8.94 ± 1.08 ^c | 3.09 ± 0.11 ^{de} | 81.72 ± 0.06 ^d | 67.84 ± 0.05 ^e | 0.74 | 1.52 | |
| RWF 80GS10 | | 58.63 ± 1.2 ^d | 1.34 ± 0.07 ^{cd} | 14.16 ± 0.41 ^{bce} | 8.15 ± 1.62 ^c | 2.81 ± 0.06 ^{bc} | 81.97 ± 0.02 ^d | 58.98 ± 0.04 ^f | 0.361 | 2.36 | |
| Germinated | RWF70GS20 | 50.46 ± 1.75 ^e | 2.76 ± 0.21 ^{ef} | 14.37 ± 0.76 ^{bc} | 16.22 ± 0.11 ^d | 3.58 ± 0.11 ^f | 83.57 ± 0.01 ^e | 0.32 | 2.32 | | |
| | RWF60GS30 | 50.46 ± 0.42 ^e | 3.29 ± 0.43 ^g | 14.82 ± 0.4 ^{cd} | 16.55 ± 1.82 ^d | 3.8 ± 0.18 ^f | 83.91 ± 0.04 ^e | 0.27 | 2.39 | | |
| | RWF50GS40 | 43.35 ± 2.85 ^f | 4.12 ± 0.61 ^h | 13.49 ± 0.38 ^{ae} | 23.56 ± 2.33 ^e | 4.27 ± 0.32 ^g | 83.96 ± 0.03 ^e | 68.81 ± 0.03 ⁱ | 0.43 | 2.43 | |
| | RWF 80RS10 | 63.19 ± 0.67 ^{bc} | 1.74 ± 0.06 ^d | 14.65 ± 0.17 ^{bcd} | 4.36 ± 2.05 ^b | 2.72 ± 0.07 ^{bc} | 73.23 ± 0.04 ^f | 57.25 ± 0.01 ^j | 0.16 | 2.16 | |
| | RWF70RS20 | 57.28 ± 1.56 ^d | 2.62 ± 0.13 ^e | 14.76 ± 0.28 ^{cd} | 9.87 ± 2.74 ^c | 3.18 ± 0.11 ^e | 71.94 ± 0.07 ^g | 59.21 ± 0.08 ^k | 0.88 | 1.78 | |
| Roasted | RWF60RS30 | 52.68 ± 2.17 ^e | 3.13 ± 0.14 ^{fg} | 14.78 ± 0.07 ^{cd} | 14.35 ± 1.02 ^d | 3.58 ± 0.18 ^f | 72.94 ± 0.05 ^f | 0.39 | 1.4 | | |
| | RWF50RS40 | 50.46 ± 1.04 ^e | 3.27 ± 0.11 ^g | 13.96 ± 0.12 ^{bc} | 16.46 ± 2.77 ^d | 3.57 ± 0.09 ^f | 70.9 ± 0.016 ^h | NIL | 1.23 | | |

RFE refine wheat flour, CS Control sorghum flour, GS germinated sorghum flour, RS roasted sorghum flour

#Mean ± standard deviation of three replications. The value with different superscript a, b, c, d, e, f in a row is significantly ($p < 0.05$) different. (L* = Lightness, a* = Redness, b* = Yellowness, ΔE = Change in color, BI = Browning Index, RWF = Refined Wheat Flour, CS = Control Sorghum, GS = Germinated sorghum, RS = Roasted sorghum)

value, significant variation over RWF flour was reported for RWF60CS30, RWF50CS40, and RWF50RS40. Contrarily crumb a^* value differed significantly for all composite flours over RWF, with the highest a^* value reported for RWF50GS40. Significant variation in the crust b^* was observed for all the GS composite flours, RWF60CS30, RWF50CS40, RWF60RS30 and RWF50RS40 whereas crumb b^* value significantly varied for all the samples over RWF. Significant change in crust total color was thus noted for RWF50CS40 among CS samples while RWF80GS10 and RWF70GS20 differed significantly from the other two GS samples. Contrarily, not much significant variation occurred among RS composite samples. Overall highest variation in crust color occurred for CS composite flours, followed by GS and RS samples. In the CS group, the total color change of samples RWF60CS30 and RWF50CS40 did not vary significantly. While samples RWF70GS20/RWF60GS30, and RWF60RS30/RWF50RS40, did not differ significantly in their respective groups. Crust BI exhibited a decreasing pattern with CS and RS, while for GS, no significant variation occurred. Crumb BI increased with an increase in the concentration of sorghum, irrespective of treatment.

It may be stated that the polyphenols oxidation in the baking process, in general, develops brown color in the bread. However, the sorghum flour addition darkens the color of the bread, owing to its chemical composition. The higher changes in color parameters brought by RS flour might be attributed to roasting-related changes in flour, such as brown pigments/compound formation by Maillard reaction and sugar caramelization (Dube et al. 2020; Sharanagat et al. 2019). Whereas color changes in GS composite flours are related to the starch modification and protein content brought by the germination process. The darkening of sorghum composite breads was also reported by Marston et al. (2016). Mtelisi et al. (2020) stated that the reduction in brightness value due to the addition of sorghum flour above the 25% level is due to the higher availability of minerals.

Textural properties of bread

Bread texture is a very important parameter as it directly influences the consumer perceptions and commercial potential of bread. The oral processing of bread depends on the textural properties mainly as it determines the resistance to deformation under the application of a force. Various textural attributes like hardness, cohesiveness, springiness, gumminess, and chewiness of RWF and composite bread are presented in Table 2.

The RWF bread had the lowest hardness (7.60 N), while the hardness of bread crumb which is associated with peak force during first compression, was found significantly increased with an increase in the fraction of CS, GS, and RS in composite bread. However, replacement with CS

and GS flour produced lower hardness i.e., 13.34–21.09 N and 8.82–20.23 N, respectively, in bread as compared to RS (19.20–50.31 N). The increased hardness may be due to the fractional loss of starch integrity and weak gluten-starch network (Mtelisi et al. 2020). Higher hardness in RS-based bread could be due to reduced sorghum starch integrity upon heat treatment. Reduction in the structuring component (gluten) with an increase in CS, GS, and RS also led to a reduction in the cohesiveness and springiness values. This demonstrates that the composite bread required more time to recover in shape. The higher springiness of RWF and CS could also be due to the better interaction between gelatinized starch and gluten, which increases the dough elasticity and form a continuous sponge structure in the bread after heating (Amin et al. 2019). As a result of these parameters, the dependent parameters i.e. gumminess increased significantly. The gumminess of RWF replaced with 10% (4.70) and 20% (4.74) GS was comparable to RWF (4.90) bread sample.

Similarly, bread with GS showed comparable chewiness value due to limited starch in germinated flour which did not contribute much to structure by gelatinization and retrogradation. Though the germinated flour offered bulkiness to bread without improving structure, due to which the hardness of the bread increased and other properties either reduced or remained comparable to RWF. In addition, the higher α -amylase and protease activities in the germinated flour increased the transformation of wheat starch into dextrin and enhanced the retrogradation (Guardado-Félix et al. 2020). The increase in the hardness and reduction in springiness and cohesiveness of CS and RS can be attributed to the reduction in water absorption of the dough and the subsequent reduction in the moisture content of the crumb (Millar et al. 2019).

Moreover, these parameters are also strongly correlated with the volume of the bread; mostly, lower volume is associated with a more compact crumb and results in higher hardness and lower cohesiveness and springiness (Mikulec et al. 2019). Roasting significantly affects the starch and protein, reducing flour's water absorption capacity (Germishuys et al. 2020). However, unlike in the case of germinated flour, it also suppresses the α -amylase and protease activities, which results in better retrogradation in the case of composite flours. Therefore, bread made with RWF and RS showed very high hardness values as compared to CS and GS. In addition, due to the reduction in gluten, bread is unable to hold in the gases from fermentation, which causes a reduction elasticity of the bread (Pylar 1973). A similar result has been reported by Mikulec et al. (2019) for bread prepared with partially replaced RWF with untreated hemp flour. In addition, the present study's findings are also in line with the study of Millar et al. (2019) in which the wheat bread was supplemented with raw, germinated and toasted pea flour.

In-vitro protein and starch digestibility

The addition of sorghum flour to RWF for the development of composite bread had a pronounced effect on the breads' in-vitro protein and starch digestibility and is shown in Table 3. The protein digestibility of RWF bread was found to be 75.85%, which increased in CS and GS-based bread but decreased in RS-based bread. The increase in CS-based bread may be attributed to the interaction with RWF with the likely role of the particulate nature of flour having a greater surface area, thereby providing digestive enzymes greater accessibility to protein and starch. On the other hand, the activation of intrinsic amylases, proteases, phytases, and fiber-degrading enzymes upon germination resulted in increased nutrient digestibility (in-vitro protein digestibility) of GS-based bread. Roasting of sorghum suppressed protein digestibility of gluten proteins (owing to denaturation), which may be a reason for the observed reduction in protein digestibility. Besides this, protein aggregation and networking through intra- and inter-molecular S–S bonds and the development of β -sheet structure (during heat treatment) could render sorghum protein less digestible in RS-based bread (Duodu et al. 2002). Ouazib et al. (2016) also reported an increase in in-vitro protein digestibility of bread prepared with germinated chickpea. In another study, Angioloni and Collar (2012) also reported a reduction in protein digestibility with the addition of high hydrostatic pressure treated sorghum compared to wheat bread and untreated sorghum (40%) bread.

Starch digestibility of RWF-based bread was found to be 61.70% which changed to 53.13–67.84%, 57.25–68.81%, and 58.98–61.76%, respectively, with an increase in the concentrations of CS, GS, and RS flours in the composite bread. A decrease followed by an increase was observed in CS and GS-based samples over RWF. It was stated that sorghum starch, as such, does not have unusual structural and chemical characteristics which slow down its digestibility. Additionally, the sorghum protein matrix acts as a barrier to starch gelatinization as well as digestibility due to cross-linking between γ - and β -kafirins and matrix proteins. However, in GS-based samples, reduced digestibility at initial concentrations may be due to the combined effects of sorghum-polyphenols, sorghum starch–protein interactions related to the high proportion of di-sulfide cross-linkages in sorghum proteins (Yousif et al. 2012) and RWF starch. An increase observed at higher concentrations might be due to the increased levels of starch exposed to hydration and enzymatic action of sorghum and its interaction with RWF flour. The decrease in starch digestibility of RS-based bread may be attributed to the limiting activity of the *in-vitro* digestive enzymes owing to (a) impeded starch gelatinization, (b) impeded starch accessibility because of the development of

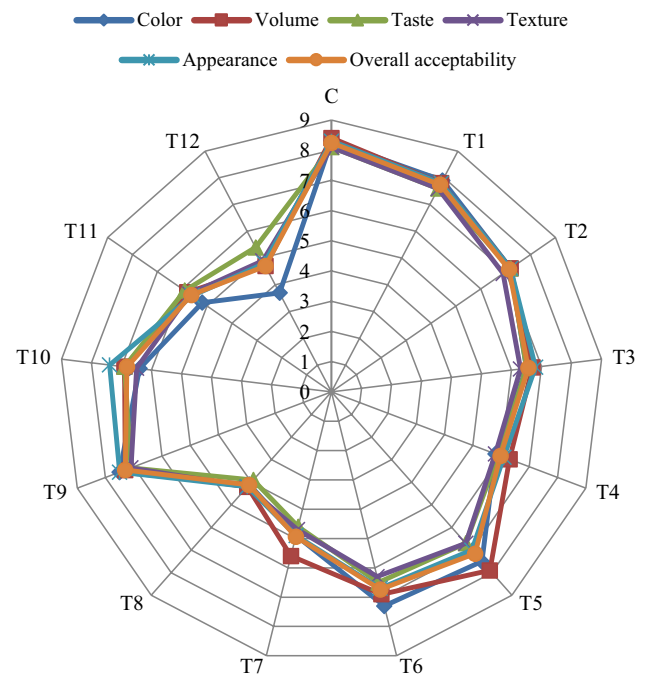


Fig. 2 Sensory analysis of composite bread (C: RWF (100%), T1: RWF 80%+CS 10%+RF 10%, T2: RWF 70%+CS 20%+RF 10%, T3: RWF 60%+CS 30%+RF 10%, T4: RWF 50%+CS 40%+RF 10%, T5: RWF 80%+GS 10%+RF 10%, T6: RWF 70%+GS 20%+RF 10%, T7: RWF 60%+GS 30%+RF 10%, T8: RWF 50%+GS 40%+RF 10%, T9: RWF 80%+RS 10%+RF 10%, T10: RWF 70%+RS 20%+RF 10%, T11: RWF 60%+RS 30%+RF 10%, T12: RWF 50%+RS 40%+RF 10%)

a low-digestible protein matrix, (c) sorghum polyphenolics based inhibition of enzymes/direct interaction with starch. Angioloni and Collar (2012) also reported a reduction followed by an increase in starch digestibility for the bread prepared with the combination of wheat and 40% untreated and high hydrostatic pressure treated sorghum, respectively, compared to wheat bread.

Microbiological analysis and shelf life analysis

Microbiological analysis of prepared bread samples with respect to storage time (at 27 °C) is presented in Table 3. The entire prepared samples showed very less microbiological count < 1 log CFU/g at the beginning of storage. An increase in storage time of up to 2 days increased the microbiological count up to 2.389 log CFU/mL (maximum). No mold and fungi were observed up to 3 days of storage. An increase in storage time led to an increase in microbiological count, and storage beyond 3 days led to the development of mold and fungi. This might be due to the high moisture content (31–35%) and water activity (95%), making bread susceptible to the growth of mold and fungi. In addition, the

absence of preservatives reduced the shelf-life of the bread. However, the prepared bread samples had less microbiological count as compared to the International Microbiological Standards Recommendation for dry and ready-to-eat foods i.e., 10^3 – 10^2 cfu/g for coliforms and $< 10^3$ cfu/g for total heterotrophic bacteria (Khanom et al. 2016).

Hence, it may be stated that the prepared bread samples are safe for consumption up to 2 days of storage, but further work needs to be performed on increasing the shelf-life of bread using additives, salts, and preservatives. Similar findings were reported by Deseta et al. (2021), where mold and fungi developed in preservative-free bread within 3–4 days of storage at 25 °C.

Sensory analysis

The sensory analysis of the sorghum bread was analyzed by the descriptive method of hedonic scale measurement (Ouazib et al. 2016). The sensory evaluation of RWF and different sorghum-RWF composite bread was performed after 1 day of baking. The collected mean scores for each sample are shown through the radar chart in Fig. 2. From the radar chart, it was observed that as the level of treated flours of sorghum increased in the blends, the sensory score of color, volume, taste, texture, appearance, and overall acceptability of bread decreased. Rice flour did not show an effect on sorghum bread as it had a constant composition in every formulation.

There was an uneven formation of crust in composite breads, especially at high replacement levels (40%). Thus the acceptance score decreased for composite bread. Ognean (2015) reported that the shape of the bread was symmetric and normal when sorghum addition was up to 10% and 20%, while a further increases to 30% and 40% of sorghum flour made bread more flat, which also comply with observations of the present study.

The taste score of the sorghum bread decreased significantly with high replacement levels of different treated sorghum flours. An increase in bitterness was observed in sorghum bread at high levels due to the increased composition of tannin in bread formulation (Abdelghafor et al. 2011). The impact was more in GS and RS-based bread (40% addition) as germination may develop off-flavors of grain while roasting followed by baking may develop other compounds and a burnt flavor. The results of the present study are in agreement with reports of Chavan et al. (2014), in which increased levels of sorghum flour (30%, 40%, and 50%) in blends of cookies and bread increased the bitterness in the final product.

The score of color attributes of bread followed the pattern as control bread > CS bread > GS bread > RS bread with varied in replacement levels from 10 to 40%. The crust and

crumb of RS bread were darker in color compared to the other sorghum-based bread due to the inherent browning of sorghum flour developed upon roasting i.e., high-temperature treatment caused the formation of brown pigments through Maillard reaction in flour (Ranganathan et al. 2014).

The texture is one of the main sensory attributes which affect the quality of bakery products. The increased levels of the sorghum flours in blends gave a harder texture at the outer side of the crust due to enhanced bread density and low volume. The high reduction of the mean score was also observed for volume at 30% and 40% replacement levels of GS and RS flours in bread. This was due to the high water binding capacity of these flours, which tend to evolve or release less amount of gas during baking and gives lower volume to bread. Even though the dense structure of the crust layer was formed on the top surface of the GS-based bread, these bread were found to be stickier as compared to CS and RS-based bread. This was because of the high water-holding capacity of GS flour.

Additionally, the evolution of gases during baking made GS-based breads more crumbly in nature (Fig. 3), making these breads unacceptable for consumers due to their non-effective/appealing appearance. Overall, the high concentration sorghum-based breads were harder and darker than RWF breads. Still, the present study reports that the bread prepared with 10% CS possessed the best texture, good taste, and adequate appearance at par to control sample, and the same is verified by high means scores for various sensory attributes in CS-based bread. With respect to GS and RS-based bread, 10–20%, and 10–30% incorporation may be done; however, taste enhancements may add benefits for customers.

Conclusion

In the present study, the sorghum flour derived from two traditional pretreatment i.e. germination and roasting, were utilized for bread development and mitigating the existing knowledge gaps in published literature for the benefit of researchers. Though most of the properties and their trends in composite bread were common irrespective of the processing performed on sorghum grains, prominent variability was observed for dynamic rheology, bread moisture content, baking loss, and in-vitro digestibility (protein and starch). Also, gluten reduction brought by sorghum flour upon replacement in RWF was the main reason for crust hardness and darker color of bread, still the processing or pre-treatments held their own impact with visibly prominent effect on chewiness, stickiness, bulkiness, and digestibility. Microbiological analysis

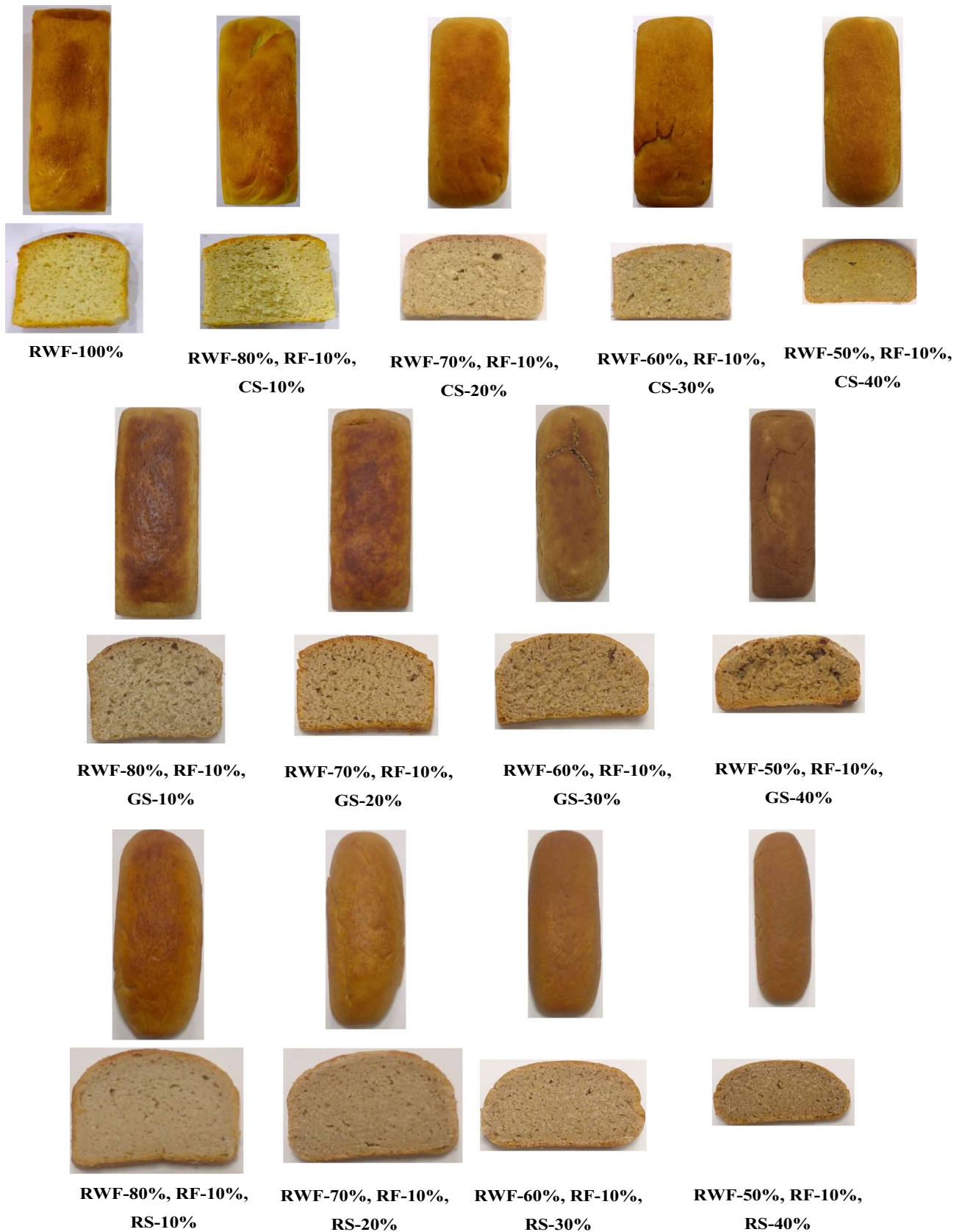


Fig. 3 Effect of addition of sorghum flour on color of composite bread

showed that the preservative-free breads can be stored up to 3 days but still future research is needed on extending shelf life through preservatives for commercial purposes. Similarly, replacement up to 30% RS, 20% CS, and 20% GS was found to have the most acceptable sensory scores. Though an advantage of incorporating sorghum in bread is justified, especially for concerns related to gluten intolerance and celiac disease, further work is warranted in the field to improve the bread quality through compositional changes, mixing of additives/preservatives, and moving towards higher replacement levels (such as beyond 40%) with acceptable taste, flavor and other sensory attributes.

Acknowledgements All the authors are thankful to the National Institute of Food Technology Entrepreneurship and Management, Kundli, Haryana, India, for infrastructural and financial support to conduct the present study.

Author contributions VSS—experiment, methodology, writing- original draft preparation. PKN—conceptualization, validation, supervision, project administration, writing- reviewing and editing. LS—conceptualization, validation, writing- reviewing and editing. SM—experiment, methodology, instrument support. AK—conceptualization, experiment, methodology, reviewing and editing.

Funding No special funding has been received for the present work.

Data availability All the data used in present study have been provided in the manuscript and supplementary material.

Code availability Not required.

Declarations

Ethics approval The present study do not require ethical approval.

Consent to participate Formal consent has been taken from the participant before the sensory analysis.

Consent for publication All the images and data presented in manuscript have been generated in the study. For the supporting research, the sources have been cited in the reference section.

Conflict of interest Authors do not have any known conflict of interest.

References

- Abdelghafor RF, Mustafa AI, Ibrahim AMH, Krishnan PG (2011) Quality of bread from composite flour of sorghum and hard white winter wheat. *Adv J Food Sci Technol* 3(1):9–15
- Abdelghafor RF, Mustafa AI, Ibrahim AM, Chen YR, Krishnan PG (2013) Effects of sorghum flour addition on chemical and rheological properties of hard white winter wheat. *Adv J Food Sci Technol* 5(11):1407–1412
- Almaiman SA, Albadr NA, Alsulaim S, Alhuthayli HF, Osman MA, Hassan AB (2021) Effects of microwave heat treatment on fungal growth, functional properties, total phenolic content, and antioxidant activity of sorghum (*Sorghum bicolor* L.) grain. *Food Chem* 348:128979
- American Association of Cereal Chemists (AACC) (2000) Approved methods of the AACC, 11th ed. Method 02-52, approved April 1961, revised October 1976 and October 1981; Method 44-15A, approved October 1975, Method 55-30.01, Method 02-52, revised October 1981 Method 55-30.01. The Association: St. Paul, MN
- Amin MNG, Hasan MN, Pralebda SA, Pramono H, Saputra E, Subekti S, Alamsjah MA (2019) Texture profile of the bread produced from composite flour Bruguiera gymnorrhiza flour (BGF) and wheat flour. In: IOP conference series: earth and environmental science, vol 236, no 1. IOP Publishing, p 012109
- Angioloni A, Collar C (2012) Effects of pressure treatment of hydrated oat, finger millet and sorghum flours on the quality and nutritional properties of composite wheat breads. *J Cereal Sci* 56(3):713–719
- Arufe S, Della Valle G, Chiron H, Chenlo F, Sineiro J, Moreira R (2018) Effect of brown seaweed powder on physical and textural properties of wheat bread. *Eur Food Res Technol* 244(1):1–10
- Campos-Vega R, Loarca-Piña G, Oomah BD (2010) Minor components of pulses and their potential impact on human health. *Food Res Int* 43(2):461–482
- Chavan et al (2014) Effect of seed priming on field performance and seed yield of soybean [*Glycine Max* (L.) Merrill] varieties. *Bioscan* 9(1):111–114
- Collar C, Jiménez T, Conte P, Fadda C (2014) Impact of ancient cereals, pseudocereals and legumes on starch hydrolysis and antiradiative activity of technologically viable blended breads. *Carbohydr Polym* 113:149–158
- Cornejo F, Caceres PJ, Martínez-Villaluenga C, Rosell CM, Frias J (2015) Effects of germination on the nutritive value and bioactive compounds of brown rice breads. *Food Chem* 173:298–304
- Coronel EB, Guiotto EN, Aspiroz MC, Tomás MC, Nolasco SM, Capitani MI (2021) Development of gluten-free premixes with buckwheat and chia flours: application in a bread product. *LWT* 141:110916
- Deseta ML, Sponton OE, Erben M, Osella CA, Frisón LN, Fenoglio C et al (2021) Nanocomplexes based on egg white protein nanoparticles and bioactive compounds as antifungal edible coatings to extend bread shelf life. *Food Res Int* 148:110597
- Dube NM, Xu F, Zhao R (2020) The efficacy of sorghum flour addition on dough rheological properties and bread quality: A short review. *Grain Oil Sci Technol* 3(4):164–171
- Dhua S, Kheto A, Sharanagat VS, Singh L, Kumar K, Nema PK (2021) Quality characteristics of sand, pan and microwave roasted pigmented wheat (*Triticumaestivum*). *Food Chem* 365:130372
- Duodu KG, Nunes A, Delgadillo I, Parker ML, Mills ENC, Belton PS, Taylor JRN (2002) Effect of grain structure and cooking on sorghum and maize in vitro protein digestibility. *J Cereal Sci* 35(2):161–174
- El-Taib HI, Rizk IRSA, Yousif EI, Hassan AA (2018) Effect of barley flour on wheat bread quality. *Arab Univ J Agric Sci* 26(Special issue (2A)):1109–1119
- Gebre E, Workiyie A, Haile K (2021) Determinants of sorghum crop commercialization the case of Southwest Ethiopia. *Heliyon* 7(7):e07453
- Germishuys Z, Delcour JA, Deleu LJ, Manley M (2020) Characterization of white flour produced from roasted wheats differing in hardness and protein content. *Cereal Chem* 97(2):339–348
- Guardado-Félix D, Lazo-Vélez MA, Pérez-Carrillo E, Panata-Saquicili DE, Serna-Saldívar SO (2020) Effect of partial replacement of wheat flour with sprouted chickpea flours with or without selenium on physicochemical, sensory, antioxidant and protein quality of yeast-leavened breads. *LWT* 129:109517
- Ibrahim UK, Rahman NAA, Suzihaque MUH, Hashib SA, Aziz RAA (2020) Effect of baking conditions on the physical properties of bread incorporated with green coffee beans (GCB). In: IOP

- conference series: materials science and engineering, vol 736, no 6. IOP Publishing, p 062019
- ICRISAT (2018) Millets and sorghum: forgotten foods for the future. ICRISAT Happenings Newsletter. www.icrisat.org/latest-happenings/
- IS 5402 (2012) Microbiology of food and animal feeding stuffs—horizontal method for the enumeration of micro-organisms—colony-count technique at 30°C [FAD 15: Food Hygiene, Safety Management, and Other Systems]
- Jafari M, Koocheki A, Milani E (2017) Effect of extrusion cooking of sorghum flour on rheology, morphology and heating rate of sorghum–wheat composite dough. *J Cereal Sci* 77:49–57
- Joghalli P, Singh L, Sharanagat VS (2017) Effect of microwave roasting parameters on functional and antioxidant properties of chickpea (*Cicer arietinum*). *LWT Food Sci Technol* 79:223–233
- Khanom A, Shammi T, Kabir MS (2016) Determination of microbiological quality of packed and unpacked bread. *Stamford J Microbiol* 6(1):24–29
- Khating KP, Kenghe RN, Yenge GB, Ingale VM, Shelar SD (2014) Effect of incorporation of sorghum flour on wheat composite bread. *J Progress Agric* 5(1):93–98
- Kulammarva AG, Sosle VR, Raghavan GV (2009) Nutritional and rheological properties of sorghum. *Int J Food Prop* 12(1):55–69
- Kumar Y, Sharanagat VS, Singh L, Mani S (2020) Effect of germination and roasting on the proximate composition, total phenolics, and functional properties of black chickpea (*Cicer arietinum*). *Legume Sci* 2(1):e20
- Marchini M, Carini E, Cataldi N, Boukid F, Blandino M, Ganino T et al (2021) The use of red lentil flour in bakery products: How do particle size and substitution level affect rheological properties of wheat bread dough? *Lwt* 136:110299
- Marston K, Khouryieh H, Aramouni F (2016) Effect of heat treatment of sorghum flour on the functional properties of gluten-free bread and cake. *LWT Food Sci Technol* 65:637–644
- Mekhael EK (2005) Biochemical and Technological Studies on Balaady Bread made from Durum wheat mixed with some cereals and cereal by products. Doctoral dissertation, M. Sc. Thesis, Cairo Univ., Egypt, pp 65–67
- Mikulec A, Kowalski S, Sabat R, Skoczylas Ł, Tabaszewska M, Wywrocka-Gurgul A (2019) Hemp flour as a valuable component for enriching physicochemical and antioxidant properties of wheat bread. *LWT* 102:164–172
- Millar KA, Barry-Ryan C, Burke R, McCarthy S, Gallagher E (2019) Dough properties and baking characteristics of white bread, as affected by addition of raw, germinated and toasted pea flour. *Innov Food Sci Emerg Technol* 56:102189
- Mondor M, Guévremont E, Villeneuve S (2014) Processing, characterization and bread-making potential of malted yellow peas. *Food Biosci* 7:11–18
- Monteiro JS, Farage P, Zandonadi RP, Botelho R, de Oliveira LDL, Raposo A, Shakeel F, Alshehri S, Mahdi WA, Araújo W (2021) A systematic review on gluten-free bread formulations using specific volume as a quality indicator. *Foods* 10(3):614
- Nasir S, Allai FM, Gani M, Ganaie S, Gul K, Jabeen A, Majeed D (2020) Physical, textural, rheological, and sensory characteristics of amaranth-based wheat flour bread. *Int J Food Sci*
- Nirmaan AMC, Rohitha Prasantha BD, Peiris BL (2020) Comparison of microwave drying and oven-drying techniques for moisture determination of three paddy (*Oryza sativa* L.) varieties. *Chem Biol Technol Agric* 7:1–7
- Ognean CF (2015) Technological and sensorial effects of sorghum addition at wheat bread. *Agric Food*
- Ogunsakin OA, Banwo K, Ogunremi OR, Sanni AI (2015) Microbiological and physicochemical properties of sourdough bread from sorghum flour. *Int Food Res J* 22(6)
- Olojede AO, Sanni AI, Banwo K (2020) Effect of legume addition on the physicochemical and sensorial attributes of sorghum-based sourdough bread. *LWT* 118:108769
- Ouazib M, Dura A, Zaidi F, Rosell CM (2016) Effect of partial substitution of wheat flour by processed (germinated, toasted, cooked) chickpea on bread quality
- Phattanakulkaewmorie N, Paseephol T, Moongngarm A (2011) Chemical compositions and physico-chemical properties of malted sorghum flour and characteristics of gluten free bread. *World Acad Sci Eng Technol* 5(7):532–538
- Pylar EJ (1973) Baking science and technology. Seibel Pub Com 358–364
- Rakcejava T, Galoburda R, Cude L, Strautniece E (2011) Use of dried pumpkins in wheat bread production. *Procedia Food Sci* 1:441–447
- Ranganathan V, Nunjundiah IT, Bhattacharya S (2014) Effect of roasting on rheological and functional properties of sorghum flour. *Food Sci Technol Int* 20(8):579–589
- Rashwan AK, Yones HA, Karim N, Taha EM, Chen W (2021) Potential processing technologies for developing sorghum-based food products: an update and comprehensive review. *Trends Food Sci Technol* 110:168–182
- Raju J, Narasimha J, Raghunadan T, Reddy R (2021) Effect of complete diets containing different dual-purpose sorghum stovers on nutrient utilization and growth performance in sheep. *Small Rumin Res* 201:106413
- Rahman MS, Al-Zakwani I, Guizani N (2005) Pore formation in apple during air-drying as a function of temperature: porosity and pore-size distribution. *J Sci Food Agric* 85(6):979–989
- Rühmkorf C, Rüksam H, Becker T, Bork C, Voiges K, Mischnick P et al (2012) Effect of structurally different microbial homoexopolysaccharides on the quality of gluten-free bread. *Eur Food Res Technol* 235(1):139–146
- Sharanagat VS, Suhag R, Anand P, Deswal G, Kumar R, Chaudhary A et al (2019) Physico-functional, thermo-pasting and antioxidant properties of microwave roasted sorghum [*Sorghum bicolor* (L.) Moench]. *J Cereal Sci* 85:111–119
- Sharma B, Gujral HS (2019) Modulation in quality attributes of dough and starch digestibility of unleavened flat bread on replacing wheat flour with different minor millet flours. *Int J Biol Macromol* 141:117–124
- Suhag R, Dhiman A, Deswal G, Thakur D, Sharanagat VS, Kumar K, Kumar V (2021) Microwave processing: a way to reduce the anti-nutritional factors (ANFs) in food grains. *LWT* 15:111960
- Torbica A, Belović M, Popović L, Čakarević J (2021) Heat and hydrothermal treatments of non-wheat flours. *Food Chem* 334:127523
- Yousif A, Nhepera D, Johnson S (2012) Influence of sorghum flour addition on flat bread in vitro starch digestibility, antioxidant capacity and consumer acceptability. *Food Chem* 134(2):880–887

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Springer Nature or its licensor (e.g. a society or other partner) holds exclusive rights to this article under a publishing agreement with the author(s) or other rightsholder(s); author self-archiving of the accepted manuscript version of this article is solely governed by the terms of such publishing agreement and applicable law.