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Thermal Processing Strategies Enhance the Safety and Quality of Whole-Grain Pearl Millet Flour

Estratégias de Processamento Térmico Melhoram a Segurança e a Qualidade de Farinha de Milheto Integral

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Abstract

Pearl millet grains harvested in the field undergo physical cleaning to remove extraneous materials and impurities; however, these processes do not avoid microbiological contamination. This study aimed to evaluate the effects of cooking, extrusion, and roasting of whole-grain pearl millet flours on the reduction of microbial contamination, color and water absorption capacity. Thermal processes significantly reduced contamination across all treated samples, with raw flour showing the highest contamination (up to 6.19 log_{10} for enterobacteria). In contrast, extruded, roasted, and pre-cooked flours had lower contamination levels (ranging from 1.00 to 2.1 log_{10}). No *Salmonella* was detected, and *E. coli* and *Bacillus cereus* were within safe limits. Pre-cooking led to significant darkening, while extrusion preserved brightness. Furthermore, the water absorption index was notably higher (4.12 g/g) in extruded flour, suggesting improved functionality for hydration-demanding applications. In contrast, roasted flour exhibited the lowest water absorption capacity (2.08 g/g), indicating application in products with dry texture. The thermal processing improves the microbiological safety of whole-grain pearl millet flour, making it a safe option for the cereal processing industry.

Keywords: Food Contamination. Food Industry. Food Safety. Technological Properties. Whole-Grain Cereal.

Resumo

Os grãos de milheto coletados no campo passam por limpeza física para remover materiais estranhos e impurezas; entretanto, esses processos não evitam a contaminação microbiológica. Este estudo objetivou avaliar os efeitos do cozimento, extrusão e torrefação de farinha de milheto integral na redução da contaminação microbiana, na cor e na capacidade de absorção de água. Os processos térmicos reduziram significativamente a contaminação em todas as amostras tratadas, com a farinha crua apresentando maior contaminação (até 6,19 log_{10} para enterobactérias). Em contraste, as farinhas extrudadas, tostadas e pré-cozidas tiveram níveis de contaminação menores (variando de 1,00 a 2,1 log_{10}). *Salmonella* não foi detectada e *E. coli* e *Bacillus cereus* estavam dentro dos limites seguros. O pré-cozimento levou a um escurecimento significativo, enquanto a extrusão preservou a luminosidade. Além disso, o índice de absorção de água foi mais alto (4,12 g/g) na farinha extrudada, sugerindo uma funcionalidade melhorada para aplicações que demandam hidratação. Em contraste, a farinha tostada exibiu a menor capacidade de absorção de água (2,08 g/g), indicando aplicações em produtos com textura seca. O processamento térmico melhora a segurança microbiológica da farinha de milheto integral, tornando-a uma opção segura para a indústria de processamento de cereais.

Palavras-chave: Cereal Integral. Contaminação de Alimentos. Indústria de Alimentos. Propriedades Tecnológicas. Segurança de Alimentos.

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1 Introduction

Whole-grain pearl millet flour has seen limited exploration in the Brazilian food industry, yet it has the potential for developing various products like bakery goods, meat products, and dairy products. The processing of millet grains starts with separating the grains from the panicle, followed by cleaning (removing dirt, stones, and sand), polishing, and the desired processing method (Dias-Martins *et al*., 2024). Simply grinding the grains results in raw flour that has not undergone any other process.

In roasting, grains are exposed to dry heat at a high temperature (above 100 ºC) in an oven for a processing time that varies according to the purpose and raw material. This treatment modifies the color by enhancing redness and yellowness, produces aroma compounds, decreases antinutritional factors, inactivates enzymes, such as lipases responsible for hydrolytic rancidity, and lowers microbial contamination, thereby extending the product's shelf (Li *et al*., 2023; Sruthi *et al*., 2021).

Millet grain cooking can be performed by various methods such as boiling/cooking (conventional), extrusion, ohmic heating, and microwave ovens. Pre-cooking by conventional methods involves immersing the grains in boiling water for about 35 min until the starch is fully gelatinized, followed by dehydration and grinding to produce pre-cooked millet flour. This process enhances technological, microbiological, and nutritional properties, increasing water solubility, and water and oil absorption capacities, contributing to functional, safety, and sensory properties of products (Andrade *et al.,* 2021; Dias-Martins *et al*,, 2021).

Extrusion cooking involves moistening the sample (mixing ingredients with water) and forcing it through a die under high temperature, pressure, and shear conditions. This process causes structural degradation and molecular changes, resulting in physicochemical and functional modifications of the extrudates, including increased water absorption capacity and changes in flour color (Dias-Martins *et al.*, 2024; Mosibo *et al*., 2020; Yadav; Dalbhagat; Mishra, 2022). Advantages of extrusion also include decreased antinutritional factors, such as condensed tannins, trypsin inhibitors, and phytic acid, extended shelf life due to reduced microbial contamination, inhibition of lipolytic enzymes with consequent reduced lipid oxidation, increased bioavailability of phenolic acids and minerals and improved protein digestibility (Pathare *et al*., 2024; Yi *et al.*, 2022).

Each type of thermal treatment applied to millet grains can help reduce microbiological contamination. Grains harvested in the field undergo physical cleaning processes, such as sieving and blowing with compressed air, to remove extraneous materials and physical impurities. However, molds and yeasts naturally present in these cereal grains can cause product deterioration, or, under ideal conditions, filamentous fungi can develop and produce mycotoxins (Freitas-Silva *et al.,* 2021). Pathogenic bacteria like *Escherichia coli*, *Bacillus cereus*, and *Salmonella* can also be present in raw flour. Therefore, it is important not only to evaluate the microbiological quality of whole-grain flours but also to study traditional processing methods that can reduce this contamination (Naraharasetti *et al.*, 2025). Furthermore, thermal treatments can alter some physical characteristics of flours and influence the development of new food products.

The aim of this study was to produce whole-grain pearl millet flours through thermal processing methods - cooking, extrusion, and roasting - and to assess the effects of these thermal processes on reducing microbiological contamination, color and water absorption capacity.

2 Material and Methods

2.1 Processing of grains and production of whole-grain pearl millet flours

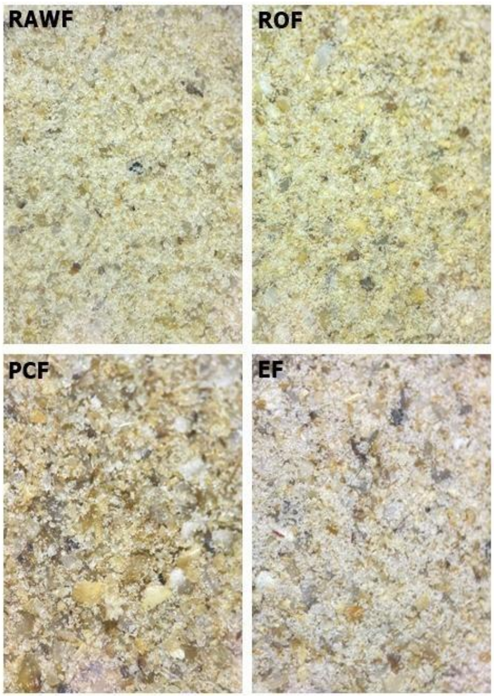
Pearl millet (*Pennisetum glaucum*) ADRG 9070 grains were provided by Atto Sementes® (ATTO Sementes, Rondonópolis, Brazil). The whole grains were pre-cleaned by manual sieving using a stainless-steel sieve with a 2000 µm mesh opening to remove coarse extraneous materials.

To obtain the whole-grain pearl millet flour (Figure 1 and 2), four types of processing were used to produce: raw whole-grain pearl millet flour (RAWF), extruded flour (EF), roasted flour (ROF), and pre-cooked flour (PCF).

Figure 1 – Whole-grain pearl millet flours processed using different thermal methods

RAWF: raw whole-grain pearl millet flour, ROF: roasted flour, PCF: pre-cooked flour, EF: extruded flour.
Source: the authors.

Figure 2 – Whole-grain pearl millet flours subjected to different types of processing viewed under a Zeiss Stemi 2000-C stereo microscope (20x magnification)



RAWF: raw whole-grain pearl millet flour, ROF: roasted flour, PCF: pre-cooked flour, EF: extruded flour.
Source: the authors.

To produce RAWF, pearl millet grains were milled using a domestic mill (RI7761, Philips Walita, São Paulo, Brazil), sieved through a stainless-steel sieve with a 600 µm mesh opening, and packed in polyethylene plastic bags at 23 °C ± 2 °C until analysis. For ROF, grains were exposed to dry heat (150 ºC ± 2 °C for 15 min) in a turbo oven (FTE 120, G. Paniz, São Paulo, Brazil), ground using a knife grinder MA 680 (Marconi, São Paulo, Brazil), remilled in a domestic mill (RI7761, Philips Walita, São Paulo, Brazil), sieved through a 600 µm mesh, and stored as described above. For PCF, grains were boiled (1:2 grain-to-water ratio) for 35 min, dehydrated at 45 °C in an electric oven FTE 120 (G. Paniz, São Paulo, Brazil) for 22 h, ground with a knife grinder MA 680 (Marconi, São Paulo, Brazil), remilled in a domestic mill (RI7761, Philips Walita, São Paulo, Brazil), sieved through a 600 µm mesh, and stored under the same conditions.

In the production of EF, grains were cleaned using an air-screen cleaner Clipper Office Tester 400/B (A.T. Ferrell Co., Bluffton, USA) equipped with 7.5 mm and 8 mm sieves, and then ground with a knife and hammer mill – 7.5 hp (TREU 95-018-B, Rio de Janeiro, Brazil) equipped with a 1 mm stainless steel sieve. The resulting flour was fed into the extruder at a rate of 10 kg/h. Extrusion was performed according to the procedure described by Costa *et al*. (2024). The extrudates (Figure 3) were dried in a forced-air oven (Hauber Macanuda, Santa Catarina, Brazil) at 60 ± 2 ºC for 24 h. After processing, the extrudates were ground with a knife and hammer mill 95-018-B 7.5 hp (TREU, Rio de Janeiro, Brazil) equipped with a 1.25 mm stainless steel sieve. The resulting flour was also stored in polyethylene bags at a temperature of 23 °C ± 2 °C until analysis.

Figure 3 – Extrudate obtained from ADRG 9070 pearl millet grains

Source: the authors.

2.2 Microbiological analyses

The microbiological analyses were performed in triplicate. To prepare the dilutions, 25 g of each sample was homogenized with 225 mL of peptone saline solution, resulting in a 10^{-1} dilution. From this, serial dilutions were made by transferring 1 mL of the sample to a tube containing 9 mL of 0,85% peptone saline solution This process was repeated until the appropriate dilution of 10^{-5} was obtained, when necessary.

After the dilution process, the following analyses were conducted: aerobic mesophiles count (CFU/g), total coliforms (CFU/g), *Escherichia coli*/g, presumptive *Bacillus cereus*/g, molds and yeasts/g, and *Salmonella* test, according to the procedures described in the Bacteriological Analytical Manual (BAM/FDA) (Food and Drug Administration, 1998).

2.3 Color

The color of the flours was evaluated using a portable colorimeter (Minolta Chroma Meter CR-210) based on reflectance measurements in the L*, a*, and b* color space (Costa *et al.*, 2024). Measurements were taken at three different points on the sample contained in the cuvette, with each sample analyzed with four replicates.

2.4 Water absorption index

The water absorption index (WAI) of the flours was determined in triplicate, following the AACC International Method 56-20.01 (AACC, 2010). For each test, 2.0 g of sample was placed in centrifuge tubes, followed by the addition of 40 mL of distilled water. The mixture was allowed to rest for 10 min, with stirring at 5 and 10 min. After this period, the samples were centrifuged at 1000 g for 15 min. The supernatant was then carefully decanted, and the sediment was weighed. The WAI was calculated as the ratio of the sediment weight after centrifugation to the initial sample weight.

2.5 Statistical analysis of data

The results of the microbiological analysis (expressed in $\log_{10}$) were subjected to a normality test (Shapiro-Wilk test). Since the data did not follow a normal distribution, a non-parametric approach was adopted. The medians of the results were analyzed using the Kruskal-Wallis test at a significance level of 5% ($p \leq 0.05$). For the objective color and water absorption index results, data followed a normal distribution, and analysis of variance (ANOVA) was performed, followed by Tukey's multiple comparison test, also at a 5% significance level ($p \leq 0.05$). All statistical analyses were carried out using the InfoStat software, version 2020.

3 Results and Discussion

3.1 Microbiological analyses

All samples showed absence of *Salmonella*, and the counts of *E. coli* and presumptive *B. cereus* were <1.0 x 10^1 CFU/g and <1.0 x 10^2 CFU/g. The results of the other microbiological analyses are presented below (Table 1).

Table 1 – Median results of microbiological analyses of raw, extruded, pre-cooked, and roasted pearl millet flours

Samples	Microbiological Counts Expressed in Log_{10}* CFU/g			
	Enterobacteria	Total coliforms	Mesophiles	Molds and Yeasts
RAWF	6.19[a]	5.69[a]	6.04[a]	4.4[a]
EF	1.85[b]	1.05[b]	1.39[b]	2.00[b]
PCF	1.00[b]	1.00[b]	1.48[b]	2.00[b]
ROF	1.00[b]	1.00[b]	2.1[b]	2.00[b]

Medians ± standard deviation with the same letters in the column do not differ statistically from each other by the Kruskal-Wallis test (p>0.05). *No presence of *Salmonella* was found, and the results for *E. coli* and presumptive *B. cereus* were lower than 1.0 x 10^1 CFU/g and 1.0 x 10^2 CFU/g, respectively. RAWF: raw flour; EF: extruded flour; PCF: pre-cooked flour; ROF: roasted flour.
Source: research data.

According to Normative Instruction No. 161 (Brasil, 2022), only the analyses of *Salmonella*/25 g, *E. coli*/g, and *Bacillus* cereus/g are required, and all samples were below the limits established by Brazilian legislation, indicating compliance with microbiological safety standards.

Although only the evaluation of these microorganisms (*Salmonella*, *E. coli* and *B. cereus*) is required by legislation, other important microorganisms in food safety were also studied in the present research. Sabillón *et al.* (2021) emphasize that microbial contamination of cereal flour primarily originates from the surface of the grain and the milling equipment. This contamination can persist due to a lack of microbial-reduction steps in the milling process, leading to the presence of pathogenic bacteria and fungi in the final product. For thermally processed millet flour, understanding these contamination points is crucial.

For total enterobacteria, total coliforms, mesophiles, and molds and yeasts, the raw flour showed the highest contamination (6.19, 5.69, 6.04 and 4.4 log_{10} CFU/g), significantly differing from the others ($p < 0.05$). For enterobacteria count, there was approximately a 4 log reduction for EF and 5 log reduction for PCF and ROF. However, there was no significant difference ($p > 0.05$) between the heat-treated samples for this group of bacteria. Total coliforms and mesophiles counts reduced close to 4 log for all samples, while molds and yeasts count decreased 2 log for all samples.

Enterobacteria and total coliforms are indicators of overall sanitation, manufacturing processes, and hygiene conditions, while mesophiles represent a broad category of bacteria that can include spoilage organisms or pathogens (Mladenovic *et al.*, 2021; Sabillón; Bianchini, 2016). The presence of molds and yeast in flour can impact product quality and shelf life, and lower counts suggested that the thermal treatments applied significantly influenced their levels. The reduction in these counts in extruded, pre-cooked, and roasted flour samples suggests that these processing methods effectively mitigate microbiological contamination, enhancing the safety and quality of the final products.

These findings are also consistent with previous studies demonstrating the effectiveness of thermal processing in reducing microbial contamination in cereal flours. Boyd *et al*. (2017) reported significant reductions in the microbial count of mesophiles (3.5 log CFU/g), mold and yeast (1.0 log CFU/g), and coliforms (0.2 log CFU/g), after roasting (110 ºC/90 min in a convection oven) whole-grain barley. *E. coli* was not detected in the whole-grain barley.

Similarly, Choudhary *et al.* (2025) observed a significant reduction in the microbial count for coliforms (approximately 2.0 log CFU/g), and for aerobic mesophiles and for yeast (3.0 log log CFU/g), after open steaming process (120 – 190 ºC/20 min) of pearl millet. For blanched sample (boiling water/30 s), they observed 4.0 log reduction for coliforms, and 3.0 log reduction for aerobic mesophiles and for yeast.

Correia *et al.* (2020) characterized microbiological quality of extruded sorghum flour of the BRS 305 genotype. All parameters evaluated (total coliforms and thermotolerant coliforms, mold and yeast count, *Bacillus cereus* and *Salmonella* spp.) were within the recommended standards by Brazilian legislation, similar to the results found in this study. Extrusion processing enhances the microbiological stability of flours by combining high temperature, moisture, and mechanical shear, which collectively lead to the inactivation of microorganisms, including cells, enzymes, spores, and toxins. The thermal conditions applied reduce the survival of contaminating microbes, while the resulting low water activity of extruded flours creates an unfavorable environment for microbial growth, especially bacteria, thereby enhancing product safety and shelf-life stability (Nwadi & Okonkwo, 2026).

Therefore, the reductions observed in the present study align with these reports and confirm that thermal treatments significantly improve the microbiological quality of pearl millet flours.

3.2 Objective color evaluation

The RAWF, EF, and ROF samples showed similar L* values ($p > 0.05$), suggesting that extrusion and roasting processes did not significantly alter the brightness of the flour compared to raw flour. However, the PCF sample showed a significant reduction ($p < 0.05$) in brightness (62.26), indicating that pre-cooking promotes flour darkening (Table 2). PCF and ROF showed more intense red and yellow hues, resulting in a brownish coloration. Pre-cooking and roasting may enhance the development of brown-colored compounds due to the thermal degradation of flour components, such as sugars and proteins, as well as increased intensity of Maillard reactions or caramelization (Oracz; Nebesny, 2019; Sruthi *et al*., 2021).

Table 2 – Color parameters of raw, extruded, pre-cooked, and roasted pearl millet flours

Samples	L*	a*	b*
RAWF	71.74 ± 0.90[a]	1.13 ± 0.18[b]	17.80 ± 0.55[c]
EF	72.61 ± 1.26[a]	0.76 ± 0.05[c]	19.15 ± 0.32[b]
PCF	62.26 ± 1.06[b]	1.59 ± 0.17[a]	21.62 ± 0.87[a]
ROF	69.51 ± 3.44[a]	1.50 ± 0.26[a]	21.62 ± 0.13[a]

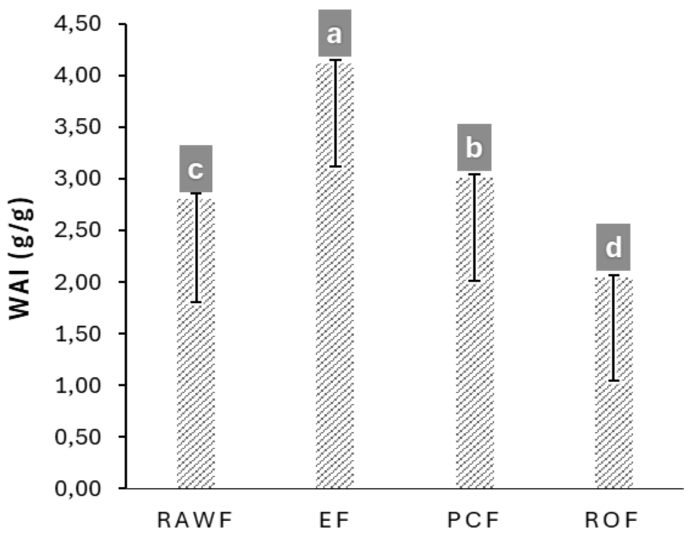
Means ± standard deviation with the same letters in the column do not differ statistically according to Tukey's test ($p > 0.05$). RAWF: raw flour; EF: extruded flour; PCF: pre-cooked flour; ROF: roasted flour.
Source: research data.

EF exhibited the lowest a* value (0,76), indicating that it is less reddish than the others. This lower a* value may be attributed to the shorter cooking time involved in the extrusion process. Additionally, extrusion increased the b* value compared to raw flour, although to a lesser extent than in the pre-cooked and roasted samples, suggesting that this process also contributes to the formation of yellowish compounds.

3.3 Water absorption index

The different treatments significantly impacted the water absorption capacity of the flours. The extruded flour exhibited the highest WAI ($p < 0.05$), likely due to the increased amount of pre-gelatinized starch formed during the extrusion process (Figure 4).

Figure 4 – Water absorption index of raw, extruded, precooked, and roasted pearl millet flours



Means with the same letters do not differ statistically according to Tukey's test ($p > 0.05$). WAI: water absorption index; RAWF: raw flour; EF: extruded flour; PCF: pre-cooked flour; ROF: roasted

flour.
Source: research data.

The different treatments significantly impacted the water absorption capacity of the flours. The extruded flour exhibited the highest WAI ($p < 0.05$), likely due to the increased amount of pre-gelatinized starch formed during the extrusion process. This process involves high temperatures and pressure, which modify the structure of proteins and starches, denaturing them and breaking intermolecular bonds. As a result, the interaction of molecules with water is facilitated, leading to greater absorption capacity (Mosibo *et al*., 2020). These results agree with Dias-Martins *et al.* (2024). who also found a higher WAI for pearl millet extruded flour (4.12 g/g) when compared to raw flour (1.96 g/g).

Conversely, the roasted flour showed the lowest WAI ($p < 0.05$), likely due to the accelerated movement of starch chains during the process, which forms intermolecular hydrogen bonds and limits the availability of hydroxyl groups to interact with water. Additionally, roasting reduces the capacity for starch degradation into smaller molecules through dextrinization or gelatinization (Li *et al*., 2023).

The pre-cooked flour presented a higher WAI than raw flour but lower than that of extruded flour. Pre-cooking alters the structure of starch and proteins, enhancing their water absorption capacity, although less intensively than extrusion, as pre-cooking involves lower pressure (Andrade *et al.*, 2021; Dias-Martins *et al*., 2019). Raw flour, in contrast, retains a less modified starchy structure, resulting in a low water absorption capacity due to the presence of native starch (Yadav; Dalbhagat; Mishra, 2022).

The variation in water absorption values among the pearl millet flours can significantly influence their application in food products, as this parameter affects the texture, viscosity, and moisture retention capacity of the products (Godswill; Somtochukwu; Kate, 2019). The choice of flour depends on the desired characteristics of the final product. Food products requiring moisture retention or higher hydration, such as bread, cakes, and pasta, would benefit from flours with higher water absorption capacity, as would sauces that do not require heating (Cayres *et al*., 2021; Román; Reguilón; Gómez, 2018). Conversely, for products that require a drier or crispier texture, such as cookies and toast, flours with lower water absorption may be more suitable. Thus, the whole pearl millet flours studied could be utilized for the development of a variety of food products.

These findings underscore the potential of whole-grain pearl millet flour as a viable option for the cereal processing industry, supporting its use in the development of safe and nutritious food products. Nonetheless, for future studies, parameters such as proximate composition, starch and protein modifications, bioactive compounds content, could provide a more complete understanding of the effects of the processing methods to help food application of the pearl millet flours.

4 Conclusion

Cooking, extrusion, and roasting proved to be effective strategies for producing whole-grain pearl millet flours with improved microbiological quality, confirming the potential of thermal processing to enhance food safety. In addition to reducing microbial contamination, the treatments promoted distinct modifications in color and water absorption capacity, demonstrating that each process influences both safety and technological performance. Extrusion and pre-cooking enhanced hydration-related functionality, while roasting contributed to characteristic color changes and lower water absorption behavior.

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