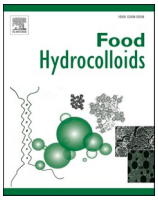
Food Hydrocolloids 159 (2025) 110680

Contents lists available at ScienceDirect

Food Hydrocolloids

journal homepage: www.elsevier.com/locate/foodhyd



The role of microwave absorption capacity and water mobility in the microwave treatment of grain vs. flour: Impact on the treated flour characteristics

Ainhoa Vicente [a,b], Marina Villanueva [a,b], Jose María Muñoz [b,c], Pedro A. Caballero [a,b], Felicidad Ronda [a,b,*]

[a] Department of Agriculture and Forestry Engineering, Food Technology, College of Agricultural and Forestry Engineering, University of Valladolid, Av. Madrid, 44, 34004, Palencia, Spain
[b] Research Institute on Bioeconomy - BioEcoUVa, PROCEREALtech Group, University of Valladolid, Spain
[c] Department of Electricity and Electronics, University of Valladolid, 47011, Valladolid, Spain

ARTICLE INFO

Keywords:
Microstructure
Thermal properties
Techno-functional properties
Amaranth
Buckwheat
Quinoa
Sorghum
Thermal treatment
Physical modification

ABSTRACT

Four alternative crops (amaranth, buckwheat, quinoa, and sorghum) were subjected to microwave-assisted heat-moisture treatment (MWT) under identical conditions (100 °C for 30 min at 25% moisture content) in two different forms (grain and flour). The effects of MWT on the microstructure, thermal properties, and techno-functional properties of the resulting flours were evaluated. The microwave absorption capacity and water mobility in flours and grains during heating were also determined to explain the different impacts of MWT depending on the matrix and its form. The results revealed that the microwave absorption capacity was comparable for all the samples, with water being the primary microwave-absorbing component. However, a different mobility and distribution of water during heating were observed for grains and flour and among matrices, leading to variations in the properties measured. The matrix, the form of treatment, and their interaction significantly influenced most of the analysed properties. MWT resulted in disruption of the native structure with partial fusion and loss of integrity of constituents, reduction in gelatinisation enthalpy, increase in the extent of amylopectin retrogradation, increase in water absorption capacity, and decrease in emulsifying capacity. The water absorption index and water solubility index showed increments or decrements depending on the matrix and the form of treatment. Moreover, the MWT of grain proved effective in limiting the colour change observed in the MWT of flour while enhancing the water absorption capacity. These findings highlight the importance of controlling not only the MWT conditions, but also the form of the matrix, as this influences the mobility of water and the final flour properties.

1. Introduction

In recent years, there has been a growing interest in using alternative grains as ingredients in cereal-based foods, particularly gluten-free products. Certain minority pseudocereals, such as amaranth, buckwheat, and quinoa, as well as cereals, such as sorghum, have gained attention because of their high intrinsic nutritional value (Khoddami et al., 2023; Martínez-Villaluenga et al., 2020). Amaranth and quinoa are protein-rich with a balanced amino acid profile and high bioavailability, high in dietary fibre, and contain a lipid fraction that is rich in unsaturated fatty acids and phytosterols (Alvarez-Jubete et al., 2010). Buckwheat is high in polyphenols and fagopyritols (Alvarez-Jubete et al., 2010). Sorghum is of interest because of its high dietary fibre and phenolic compound contents, as well as its lower glycemic index compared to other cereals (Khoddami et al., 2023). However, the use of these alternative flours in their natural form presents limitations in terms of functionality and sensory qualities when applied in industrial food production (Khoddami et al., 2023; Zhao et al., 2024).

Physical modification has been widely used to modify the techno-functional properties of flours and starches to adapt them to the

* Corresponding author. Department of Agriculture and Forestry Engineering, Food Technology, College of Agricultural and Forestry Engineering, University of Valladolid, Av. Madrid, 44, 34004, Palencia, Spain.
E-mail address: mfronda@uva.es (F. Ronda).

https://doi.org/10.1016/j.foodhyd.2024.110680
Received 5 July 2024; Received in revised form 22 August 2024; Accepted 24 September 2024
Available online 24 September 2024

requirements of the food industry (Schafranski et al., 2021; Zhao et al., 2024). Heat-moisture treatment (HMT) is a common physical modification method for starch/flour. It involves subjecting starch/flour with limited moisture (10–30%) to temperatures above the glass transition and below gelatinisation (90–120 °C) for a certain period of time (Schafranski et al., 2021). Microwave radiation (MW) has been proposed as an alternative technology to perform HMT, because it is a fast process that produces volumetric heating and is environmentally friendly compared to conventional heating. MW can modify starch and protein structures, achieving different results depending on the setting of control parameters (temperature, moisture, time, power, etc.) and matrix characteristics (Rao et al., 2023; Tao et al., 2020; Zhao et al., 2024). Microwaves are electromagnetic waves in the frequency range of 1 GHz–300 GHz, with 2.45 GHz being the most common frequency for the food industry and commercial microwaves (Villanueva et al., 2018). The MW electric field is the main agent responsible for the heating. The interaction between this field and the polar molecules of the material through their rotational relaxation processes, results in the conversion of electromagnetic energy into heat (Tao et al., 2020). The dielectric properties of materials reveal their interaction with electric fields, which is crucial in microwave treatment (MWT) as it influences the heating characteristics of the material (Sosa-Morales et al., 2010; Villanueva et al., 2018). The dielectric properties of food depend greatly on its moisture, salt, and mineral content, as food organic constituents can be considered transparent to MW compared to aqueous ionic fluids or water (Sosa-Morales et al., 2010). However, the dielectric properties of a material are influenced not only by the water and salt content but also by the way they are bonded or restricted by other food components, as well as the density and porosity of the material (Sosa-Morales et al., 2010; Tao et al., 2020). Therefore, studying these properties is important for designing and evaluating the effects of MWT on different food matrices.

MWT has primarily been applied to modify starches, but in recent years it has also been evaluated in more complex systems such as flours and grains. Buckwheat grain (Vicente et al., 2023a), maize grain (An et al., 2023), quinoa flour (Cao et al., 2022), quinoa grain (Vicente et al., 2023b), rice flour (Solaesa, Villanueva, Muñoz, & Ronda, 2021; Solaesa, Villanueva, Vela, & Ronda, 2022; Villanueva et al., 2018), millet flour (Rao et al., 2023), millet grain (Zhi et al., 2022), and tef flour (Calix-Rivera et al., 2023) have all been modified using MWT. These studies have shown that MWT can successfully modify the microstructure, thermal properties, and techno-functional properties of the resulting flours, with the specific modifications varying depending on the matrix and MWT conditions, which are not comparable between the different studies found in the literature. Conducting MWT on grains offers several industrial benefits over performing it on flours, including simplified treatment processes, improved homogeneity, and elimination of risks associated with handling powdery systems, e.g., explosive atmospheres (Vicente et al., 2023a). Concerning the MWT effect, the particle size and intact plant structure of grains may affect the kinetics of heat transfer and water mobility during the MWT, in contrast to flours, potentially leading to different dielectric properties. Additionally, differences in water mobility and binding capacity in different grain structures, as well as the distribution and interaction of major biopolymers, may lead to different modifications even under the same MWT conditions. However, to the best of our knowledge, no comparative study has been performed on MWT comparing flour and grain treatment.

Therefore, the objective of this study was to determine the effect of performing MWT on grain versus flour under the same conditions (moisture content, temperature, and time) for four different matrices (amaranth, buckwheat, quinoa, and sorghum) on the microstructure, thermal, and techno-functional properties of the resulting flours. This comprehensive study attempts to understand and explain the different impacts of MWT on the resulting treated flour depending on its source and form during treatment (grain or flour). To achieve this goal, the absorption capacity and water behaviour during heating of the treated matrices were evaluated. The findings will facilitate the industrial-scale application of this technology.

2. Materials and methods

2.1. Raw materials

Amaranth grain (*Amaranthus* spp.), purchased on the local market, had a proximate composition (g/100 g grain dry matter) of 14.4 ± 0.2% protein, 6.4 ± 0.1% fat, 2.5 ± 0.2% ash, 7.7 ± 1.6% dietary fibre, and 54.4 ± 0.2% starch (5.9 ± 0.6% amylose). Dehulled buckwheat (*Fagopyrum esculentum* Moench) grains of Kora variety were obtained from the Grupa Producentów Ekologicznych Dolina Gryki Sp ZOO (Miedzylesie, Poland), and had a proximate composition (g/100 g grain dry matter) of 14.5 ± 0.1% protein, 2.9 ± 0.1% fat, 1.9 ± 0.2% ash, 3.5 ± 0.9% dietary fibre, and 70.8 ± 3.4% starch (23.6 ± 0.2% amylose). Quinoa (*Chenopodium quinoa* Willd.) grains of the Vegarosa variety were provided by Herba Ricemills (Sevilla, Spain), and the proximate composition (g/100 g grain dry matter) was 13.3 ± 0.9% protein, 2.5 ± 0.1% fat, 2.2 ± 0.2% ash, 4.9 ± 1.2% dietary fibre, and 53.7 ± 2.1% starch (10.2 ± 0.1% amylose). Sorghum (*Sorghum bicolor* L. Moench) grains were provided by Salutef (Palencia, Spain) and had a proximal composition (g/100 g grain dry matter) of 7.5 ± 0.5% protein, 3.1 ± 0.1% fat, 1.6 ± 0.2% ash, 7.1 ± 1.4% dietary fibre, 73.1 ± 0.9% starch (24.5 ± 0.6% amylose). Protein, fat, and ash contents were determined using the AACC official methods 46–19.01, 30–10.01, and 08–01.01, respectively (AACC, 2010). Dietary fibre content was determined using the AOAC official method 991.43 (AOAC, 2023). The total starch content was determined with the method described by Englyst et al. (2006), and the amylose content was measured using the Concavalin A method (Gibson et al., 1997). The grains were ground in a hammer mill (LM 3100, Perten Instruments, Stockholm, Sweden) with a 500 μm mesh to obtain the untreated-native flours.

2.2. Microwave absorption capacity

The microwave absorption capacity of the untreated/native grains and flours of amaranth, buckwheat, quinoa, and sorghum, was determined using a Keysight E5071C network analyser (Agilent, CA, USA) following the method described by Villanueva et al. (2018) with slight modifications. The two ports of the network analyser were connected via coaxial lines and attenuators (20 dB) to two coaxial waveguide transitions (SMA to WR34) aligned and faced each other at a constant distance of 11 mm. Foam absorbers were added to eliminate resonant modes of the sample holder. The measurements were performed on samples at 25% MC (moisturized as described in 2.4 section) and at 2% moisture content (MC) (freeze-dried with a LyoQuest 55Plus (Telstar, Terrassa, Spain)). Samples at the desired MC were weighted and homogenously distributed in Petri dishes. The scattering coefficient S_{12} was measured in duplicate at 2.45 GHz. The equipment was calibrated with an empty Petri dish for 0 dB and 0 rad of phase angle. Microwave absorption was quantified in terms of attenuation and phase change using the following equations:

$$\alpha = 20 \cdot \log \frac{S_{12}(filled)}{S_{12}(empty)} \quad (1)$$

$$\Theta = arg \frac{S_{12}(filled)}{S_{12}(empty)} \quad (2)$$

where α is the attenuation, Θ is the phase shift, S_{12}(filled) is the scattering coefficient when the sample container is filled with the sample, and S_{12}(empty) when it is empty.

To apply the Lambert-Beer law, the measurement was normalised to the sample weight, since the optical path cannot be completely filled

with the sample and includes the air trapped between the particles. The attenuation was therefore expressed in dB/kg and the phase shift in rad/kg.

2.3. Water behaviour during the heating process: drying curves

The drying curves for the native flours and grains were obtained using a thermobalance MB120 (Ohaus, USA). A sample of 5.00 ± 0.01 g was weighted into an aluminium pan and then heated at 100 °C for 60 min. The weight was automatically recorded every 5 s with an accuracy of 0.001 g. The moisture ratio (MR) was determined using the experimental data obtained at each time interval, as reported by Sadaka (2022), using the following equation:

$$MR = (M - M_e) / (M_0 - M_e) \quad (3)$$

where M is the moisture content at time t, M_0 is the initial moisture content, and M_e is the equilibrium moisture content, which was neglected as commonly reported by previous authors utilizing this model (Togrul & Pehlivan, 2002). All moisture contents are referred to dry basis (db).

The drying curves obtained from the experiments (MR vs time) were fitted to several drying kinetic models (Sadaka, 2022). The best-fit model was the Logarithmic model (Onwude et al., 2016):

$$MR = a \cdot \exp(-k \cdot t) + b \quad (4)$$

where a and b are empirical constants (dimensionless), k is the drying constant (min^{-1}), and t is the heating time (min).

2.4. Microwave treatment

Native grains and flours of amaranth, buckwheat, quinoa, and sorghum were moisturized to obtain 25% MC by adding distilled water while mixing (Vicente et al., 2023a). MC was determined according to the AACC official method 44–15.02 (AACC, 2010). Microwave treatment was performed at 900 W and 2450 MHz using a customized microwave oven R342INW (SHARP, Sakai, Japan), with a computer-controlled system to regulate the microwave application pattern and time according to the programmed temperature. To perform the treatment, 120 ± 0.05 g of grain or flour were placed into a 250 mL hermetic and heat-resistant borosilicate glass container. The total treatment duration was 30 min. The first 5 min were dedicated to heating the sample from room temperature (22 ± 2 °C) to 100 °C. The temperature was then maintained at 100 ± 3 °C for the remaining 25 min. Each treatment was performed in triplicate. In addition, a temperature data logger (Pico VACQ, TMI-Orion, Castelnau-le-Lez, France) was placed inside the vessel in contact with the sample to record the temperature evolution during the treatment. The obtained flours and grains were dried at 35 °C to the natural moisture content of the original grains/flours (11–14%). The grains were then milled using a hammer mill (LM 3100, Perten Instruments, Stockholm, Sweden) with a 500 μm mesh (identical conditions to native grain milling). The treated flours were disaggregated using a stone mill Fidibus Medium (Komo, Hopfgarten, Austria). The flour samples were named with three letters, XX-Y, according to the treatment (XX) and the matrix (Y) as untreated/native (UN), treated in the form of grain (TG), or treated in the form of flour (TF), amaranth (A), buckwheat (B), quinoa (Q), or sorghum (S). This combination resulted in 12 samples.

2.5. Particle size distribution

The particle size distribution of the flours was studied using a laser diffraction particle size analyser (Mastersizer 2000, Malvern Instruments Ltd., Malvern, UK) with a particle refractive index of 1.53. The measurements were performed in triplicate. The results were expressed as D_{10}, D_{50} (median diameter), and D_{90}, representing the diameter at which 10%, 50%, and 90% of the particles have a smaller size, respectively. The arithmetic mean diameter of the flour was also determined. The size of the grains and their arithmetic mean diameter were determined by image analysis of grain images taken with a PowerShot SX410 IS camera (Canon, Japan) using the ImageJ software (National Institutes of Health, USA).

2.6. Scanning electron microscopy (SEM)

The morphological changes of the native and treated grains (grains cut in half with a scalpel) and treated flours were studied using a microscope (Quanta 200-F, FEI, Graz, Austria) equipped with an X-ray detector. The samples were mounted onto aluminium stubs using conductive carbon tape and coated with a 5 nm layer of gold using a sputter coater (SCD-05, Leica Microsystems, Wetzlar, Germany). The analysis was performed in low-vacuum mode, at an acceleration voltage of 2–5 keV, using a secondary electron detector. Micrographs were taken at various magnifications, and representative images were selected to illustrate microstructural changes.

2.7. Colour characteristics

Flour colour was measured in sextuplicate using a PCE-CSM5 colourimeter controlled by the CQCS3 software. The CIE L*a*b* and CIE L*C*h coordinates were determined with D65 standard illuminant and 10° standard observer. The colour difference (ΔE) between each treated sample and the corresponding untreated flour for the same matrix was calculated using the following equation:

$$\Delta E = \left[(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2\right]^{1/2} \quad (5)$$

2.8. Differential scanning calorimetry (DSC)

The gelatinisation and retrogradation transitions of the flour samples were determined in duplicate using a differential scanning calorimeter (DSC3, STARe-System, Mettler-Toledo, Switzerland). Approximately 6 mg of flour were weighed into 40 mL aluminium pans, and distilled water was added to obtain a water:flour ratio of 70:30. The samples were scanned from 0 to 120 °C at 5 °C/min using an empty pan as a reference. The gelatinised sample was subsequently stored at 4 °C for seven days before being rescanned to evaluate retrogradation. The enthalpy (ΔH), expressed as J/g of flour db, and the peak (T_p), onset (T_o), and endset (T_e) temperatures were determined for each of the detected peaks.

2.9. Techno-functional properties

The hydration properties of the flours in terms of water absorption capacity (WAC), oil absorption capacity (OAC), water absorption index (WAI), and water solubility index (WSI) were measured at 5% concentration, following the method described by Abebe et al. (2015) with modifications by Vicente et al. (2023a). WAC and OAC results were expressed as grams of water or oil retained per gram of flour db, WAI as grams of sediment per gram of flour db, and WSI as g of soluble solids per 100 g of flour db. Emulsifying activity (EA) and emulsion stability (ES) were determined as described by Kaushal et al. (2012) with modifications by Vicente et al. (2023a). EA and ES were expressed as percentages of the volume of emulsion formed and the emulsion volume after heating, respectively, in relation to the initial volume. Techno-functional properties were measured at least in triplicate.

2.10. Statical analysis

Statistical analysis was conducted using Statgraphics Centurion 19 software (Bitstream, Cambridge, MN, USA). To assess significant differences ($p < 0.05$) between samples, the least significant difference

(LSD) analysis of variance (ANOVA) was used. The results were presented as the mean values of the different replicates and their standard deviations. The experimental data for the drying curves were fitted using OriginPro 2023 (Northampton, MA, USA). The values of the adjusted coefficient of determination (adj-R^2) and Chi-Square (X^2) were evaluated to determine the best-fitting model (higher R^2 and lower X^2). Logarithm was employed to normalize the data and evaluate significant differences in the arithmetic mean diameter between flours and grains.

3. Results and discussion

3.1. Microwave absorption capacity and drying curves

The microwave absorption capacity of the untreated flours and grains was evaluated in terms of attenuation and phase shift at MC values of 25% and 2% (Table 1). The results showed no significant differences in the attenuation and phase shift at the same MC between the samples. However, the attenuation was approximately 30 times higher for 25% MC than for 2% MC, the latter being at the limit of detection of the equipment. The organic constituents can be considered dielectrically inert and transparent to MW compared to aqueous ionic fluids or water (Sosa-Morales et al., 2010). Our findings are consistent with those reported by Villanueva et al. (2018) for rice flour. These authors showed that water is the main responsible for microwave absorption, because the moisture content of the flour determined the attenuation and phase-shift, with the attenuation practically null in the absence of water. Additionally, our study found no significant differences in microwave absorption capacity between the matrices studied, regardless of whether they were analysed in the form of flour or grain. However, since the water in the product is responsible for the conversion of microwave energy into thermal energy, the distribution of water during treatment and its behaviour and mobility during heating should play a crucial role in the modification achieved after the MWT of grains in comparison to flours.

To evaluate the water mobility in the matrices studied during heating, samples at 25% MC were submitted to a drying process at 100 °C (MWT temperature). The drying curves obtained are presented in Fig. 1; Table 1 shows the drying parameters obtained after fitting the curves to a logarithmic model. The mean diameter of flour particles and grains is also included in Table 1, as the size of particles/grains affects the water loss kinetics. The logarithmic model is a semi-theoretical model based on Fick's second law of diffusion and has been widely applied to model the drying kinetics of foodstuffs (Onwude et al., 2016; Sadaka, 2022) when short-/medium-term drying processes are modelled. The model effectively described the evolution of the moisture ratio over time during the drying process of both grain and flour for the four matrices, presenting an adjusted coefficient of determination (Adj-R^2) > 0.989 for all samples. In addition, the "b" parameter obtained from the fitting was close to the experimental MR value at the longest drying time studied (1 h); while the addition of a+b was close to 1, as expected from the model, since MR at t = 0 must be equal to 1 (where $M = M_0$). This explains why when the parameter "a" decreased "b" increased, showing a strong negative correlation between them ($r = -0.984$, $p < 0.001$). The drying rate constant, k, was higher for flours than for grains, indicating faster drying of the flours, in the order of amaranth flour > sorghum flour > buckwheat flour = quinoa flour > amaranth grain > quinoa grain > buckwheat grain > sorghum grain. The drying rate was strongly correlated with the mean diameter of the grain/flour ($r = -0.975$, $p < 0.001$), which indicates smaller particle size led to faster drying. The lower "a" and higher "b" parameters for grains compared to flours also indicate the greater difficulty for grains in water loss during drying. It is noteworthy that this model can accurately describe the drying kinetics for the drying

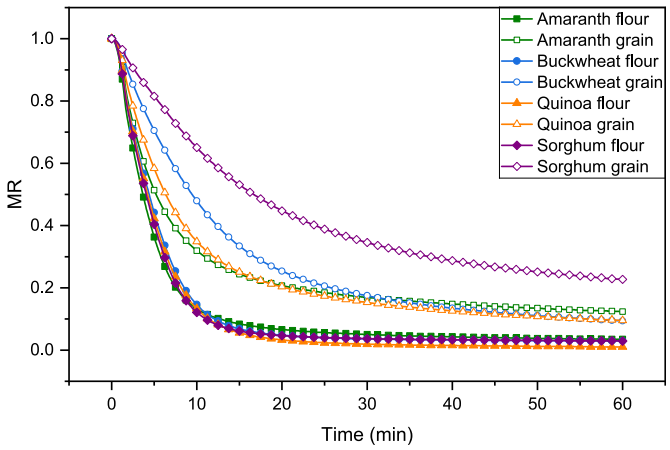


Fig. 1. Drying curves at 100 °C showing the evolution of the moisture ratio (MR) with time for native flours and grains with an initial moisture content of 25%.

Table 1
Microwave absorption capacity and drying kinetic constants of native flours and grains.

	Attenuation (dB/kg)		Phase shift (rad/kg)		Drying curves fitting parameters					Diameter (mm)
Sample	25% MC	2% MC	25% MC	2% MC	a	k (min^{-1})	b	X^2	Adj-R^2	
Amaranth flour	26.9 ± 3.0 a	0.8 ± 0.4 a	17.8 ± 1.1 a	7.8 ± 0.1 a	1.061 ± 0.001 e	0.243 ± 0.005 g	0.044 ± 0.001 c	0.0001	0.9962	0.172 ± 0.003 c
Amaranth grain	28.3 ± 0.1 a	2.4 ± 0.2 a	16.2 ± 0.1 a	7.8 ± 0.2 a	0.876 ± 0.008 b	0.168 ± 0.007 d	0.145 ± 0.005 f	0.0003	0.9894	12.2 ± 0.8 d
Buckwheat flour	31.3 ± 2.4 a	1.1 ± 0.7 a	17.9 ± 0.4 a	7.5 ± 0.1 a	1.085 ± 0.007 f	0.207 ± 0.013 e	0.037 ± 0.006 bc	0.0002	0.9958	0.113 ± 0.002 a
Buckwheat grain	28.0 ± 0.3 a	1.8 ± 0.2 a	15.8 ± 0.2 a	7.5 ± 0.2 a	0.932 ± 0.005 d	0.091 ± 0.001 b	0.107 ± 0.003 d	0.0001	0.9989	37.5 ± 2.9 f
Quinoa flour	26.2 ± 0.7 a	1.6 ± 0.8 a	16.1 ± 0.8 a	7.8 ± 0.1 a	1.100 ± 0.009 g	0.205 ± 0.008 e	0.020 ± 0.007 a	0.0001	0.9965	0.149 ± 0.015 b
Quinoa grain	30.8 ± 0.7 a	2.3 ± 0.3 a	16.9 ± 0.1 a	7.8 ± 0.1 a	0.916 ± 0.007 c	0.131 ± 0.001 c	0.116 ± 0.005 e	0.0002	0.9950	19.3 ± 1.9 e
Sorghum flour	27.3 ± 1.5 a	1.0 ± 0.1 a	17.4 ± 0.4 a	7.4 ± 0.1 a	1.092 ± 0.008 fg	0.220 ± 0.005 f	0.035 ± 0.005 b	0.0002	0.9954	0.170 ± 0.011 c
Sorghum grain	28.6 ± 0.3 a	2.2 ± 0.2 a	16.8 ± 0.3 a	7.9 ± 0.1 a	0.799 ± 0.008 a	0.062 ± 0.001 a	0.221 ± 0.006 g	0.0001	0.9995	38.0 ± 2.1 f

MC: moisture content. The drying kinetic constants were obtained after the adjustment to the logarithmic model [$MR = a \cdot exp(-k \cdot t) + b$], where *MR* is the moisture ratio (refer to section 2.3), *k* the drying constant (min^{-1}), and *a* and *b* are empirical constants. Adj-R^2 is the adjusted coefficient of determination and X^2 is the Chi-Square of the fitting model. Data is expressed as mean ± standard deviation. Significant statistical differences ($p < 0.05$) are indicated by different letters for the same parameter. Diameter refers to the arithmetic mean of particles/grains considered spherical.

time employed (1 h). At 1 h, the first term of the equation, which includes the independent variable "t", becomes nearly zero and the MR value is practically equal to the "b" value. Consequently, the constant "b" represents the residual MR at the end of the studied drying period. Nevertheless, the elevated values of the parameter "b" for grains suggest that adjusting to a model with additional exponential terms and without an independent term may be advantageous for long-term drying processes, as at MR value should be near zero at infinite drying time. Onwude et al. (2016) reported that the temperature and thickness of drying material were the most influential factors affecting the drying kinetics of food products. The moisture distribution within the particles during MWT can significantly affect the modifications obtained, as moisture is a key parameter in determining the effect of HMT (Schafranski et al., 2021). This is particularly important in MWT, as water absorbs MW and produces the heating that the sample uses to increase its temperature. During the initial stages of MWT, water moves from the inner to the outer layers of flour particles/grains, depending greatly on particle size. This behaviour occurred rapidly in the case of flour. For example, in the drying models, flour particles were almost completely dry within the first 10 min. However, for grains, this process was much slower, and significant moisture content remained even after 30 min (see Fig. 1). MWTs are usually performed under non-hermetic conditions where the sample is under atmospheric pressure and is nearly completely dried after treatment (Villanueva et al., 2018). This is the case in many studies carried out at laboratory scale, and most of the treatments performed at industrial level. In our study, the sample was introduced into a hermetic container, and at the end of treatment, after cooling, the MC was not modified. Therefore, one could think it is not possible to extrapolate this model results to our microwave system directly; however, it can be concluded that differences in moisture content and distribution within the flour particles and grains will occur during treatment. This will probably lead to notable variations in the physical modification of flours compared with grains. The smaller flour particles will dry quickly internally and have their surface surrounded by the evaporated water, which will occupy a large headspace inside the container where the sample is being treated. So, the real/actual MC of the sample during the treatment when the sample reaches 100 °C must be below the initial MC, even if it is higher than could be expected under open/nor-hermetic conditions. The larger grain particles will dry more slowly, presenting more moisture inside the grain for a longer time.

3.2. Microwave treatment

Fig. 2 shows the sample temperature evolution recorded during MWT. The temperature of the sample increased from 22 ± 2 °C to 100 ± 3 °C in 5 min and was maintained at 100 ± 3 °C for the remaining 25 min. To achieve the desired temperature profile, the MW energy (at maximum power of 900 W) was applied intermittently for 3.6 ± 0.2 min over the 30 min treatment, requiring 1.5 ± 0.1 min to increase the temperature in the ramp (first 5 min) and then 2.1 ± 0.1 min to maintain the temperature in the plateau (remaining 25 min). The control system automatically adjusted the MW application to maintain the desired temperature. There were no significant differences between the samples in the total MW time needed to increase and maintain the sample temperature during MWT. This finding aligns with the results of MW absorption capacity, which identified water content as the sole factor significantly influencing MW absorption. The temperature curve and control system were different from those in our previous studies, in which a constant microwave cycle of seconds of exposure and seconds of rest was applied, and a longer microwave time of 3–5 min was required to reach the plateau temperature, compared to 1.5 min in this study (Solaesa et al., 2021; Villanueva et al., 2018). This approach is similar to traditional HMT, where a fixed treatment temperature and time are set in the oven, and the sample is treated under these conditions, progressively increasing its temperature until the objective is achieved (Schafranski et al., 2021). However, our new approach uses the advantage of MW technology, where heat is generated inside the sample, as water absorbs MW radiation and transforms it into heat, resulting in a faster temperature rise and more efficient energy use, allowing the total treatment time and energy to be reduced.

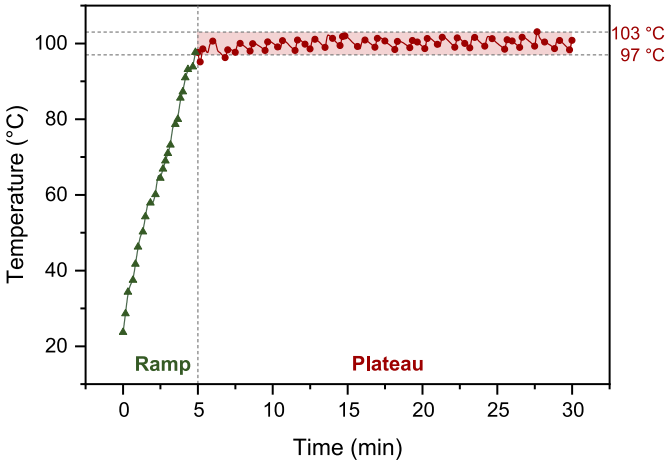


Fig. 2. Evolution of sample temperature during microwave treatment.

3.3. Particle size distribution

The particle size parameters of the flours obtained after MWT of flours and grains, measured by laser diffraction, are presented in Table 2. The particle size parameters obtained after treating and milling the samples were significantly affected by the matrix, treatment, and their interaction. Particle size differences should be taken into consideration when comparing treatment effects as they have an impact on the techno-functional and thermal properties of flour (An et al., 2023; Yang et al., 2017). The most remarkable effect of MWT was the increase in the particle size of the small fraction, as observed for the higher D_{10} of all treated samples; this effect was more intense for the flour treatment (see Supplementary Fig. 1). The largest differences in D_{10} were observed for buckwheat, with a 254% increase for TF-B and a 127% increase for TG-B compared to UN-B. The smallest differences were observed in sorghum, with increases of 41% and 20% compared to UN-S for TF-S and TG-S, respectively. The median diameter (D_{50}) and the diameter of the 90th percentile (D_{90}) of the treated buckwheat and quinoa samples showed a significant increase compared to the untreated flour. However, amaranth and sorghum showed no significant differences, with the exception of TF-A. Previous studies have reported increased particle size after MWT, with different impacts depending on the matrix and treatment conditions (An et al., 2023; Calix-Rivera et al., 2023; Vicente et al., 2023a). The authors associated the increase in particle size with modifications in the component structure resulting from MWT, such as starch gelatinisation and protein denaturation, which may induce aggregates, making milling more difficult and resulting in an increased particle size, particularly in obtaining the smallest particles.

3.4. Morphology

Fig. 3 shows SEM micrographs of the endosperm particles of the studied samples before milling obtained at a magnification of 6000 ×. Supplementary Fig. 2 displays micrographs of the same area at a wider range of magnifications (500 ×, 1000 × , 3000 × , and 6000 ×). The appearance and structure of the different matrices varied considerably. Amaranth presents small polygonal starch granules (0.5–2 μm) in a very compact matrix (Perez-Rea & Antezana-Gomez, 2018). Quinoa also has small polygonal granules (1–2 μm) that are presented individually or as spherically packed aggregates in the endosperm (Vicente et al., 2023b). Buckwheat starch granules vary in shape, being polygonal or spherical,

Table 2
Particle size and colour characteristics of flour samples obtained from untreated and microwave-treated flours and grains.

Sample	D_{10}	D_{50}	D_{90}	L*	C*	h	ΔE
UN-A	23 ± 3 a	140 ± 2 a	322 ± 6 b	85.7 ± 0.5 c	13.8 ± 0.8 a	69.1 ± 0.6 b	–
TF-A	47 ± 1 c	138 ± 2 a	277 ± 5 a	80.6 ± 0.9 a	19.2 ± 0.2 b	68.8 ± 0.2 ab	7.5 ± 0.6 b
TG-A	30 ± 3 b	144 ± 13 a	333 ± 23 b	85.0 ± 0.3 b	14.2 ± 0 a	68.4 ± 0.3 a	0.9 ± 0.2 a
UN-B	9 ± 1 a	83 ± 1 a	226 ± 6 a	88.3 ± 0.2 c	8.0 ± 0.1 a	74.2 ± 0.6 c	–
TF-B	33 ± 3 c	110 ± 2 c	296 ± 8 c	75.0 ± 0.4 a	9.9 ± 0.2 c	60.4 ± 0.3 a	13.6 ± 0.4 b
TG-B	21 ± 2 b	94 ± 2 b	256 ± 18 b	83.6 ± 0.2 b	8.2 ± 0.1 b	61.2 ± 0.1 b	5.1 ± 0.2 a
UN-Q	13 ± 3 a	113 ± 15 a	314 ± 19 a	90.7 ± 0.6 c	11.1 ± 0.2 a	82.1 ± 0.9 c	–
TF-Q	26 ± 1 c	143 ± 2 b	346 ± 4 b	85.8 ± 0.1 a	14.5 ± 0.1 c	74.4 ± 0.2 a	6.2 ± 0.2 b
TG-Q	21 ± 1 b	143 ± 7 b	354 ± 5 b	88.0 ± 0.2 b	11.8 ± 0.2 b	78.7 ± 0.5 b	2.9 ± 0.2 a
UN-S	17 ± 2 a	135 ± 12 a	345 ± 15 a	81.6 ± 0.6 c	9.8 ± 0.5 a	60.1 ± 0.3 a	–
TF-S	24 ± 1 c	142 ± 5 a	332 ± 11 a	74.8 ± 1.2 a	11.9 ± 0.2 c	64.1 ± 0.2 c	7.2 ± 0.9 b
TG-S	20 ± 1 b	141 ± 1 a	352 ± 3 a	79.3 ± 0.5 b	11.4 ± 0.4 b	63 ± 0.3 b	2.9 ± 0.5 a
Analysis of variance and significance (p-values)							
Matrix (F1)	***	***	***	***	***	***	***
Treatment (F2)	***	***	***	***	***	***	***
F1 x F2	***	**	***	***	***	***	***

Flours obtained from untreated-native grain (UN), microwave-treated grain (TG), and microwave-treated flour (TF) of amaranth (A), buckwheat (B), quinoa (Q), and sorghum (S). D_{10}, D_{50}, and D_{90}: diameter where 10%, 50%, and 90% of particles have a smaller size, respectively L*: luminosity; C*: chroma; h: hue; ΔE: colour difference from untreated-native flour. Data is expressed as mean ± standard deviation. Significant statistical differences ($p < 0.05$) are indicated by different letters for the same parameter and matrix. Analysis of variance and significance of matrix (A,B,Q,S), treatment (UN, TG, TF), and their interaction: ***$p < 0.001$, **$p < 0.01$, *$p < 0.05$, ns: non-significant.

with a size of 2–15 μm, and are packed in clusters of different sizes, with thin membranes and presenting attached or entwined protein particles (Perez-Rea & Antezana-Gomez, 2018). Sorghum endosperm has corneous and floury zones with starch granules of 5–30 μm organised in cells with a protein matrix, with higher protein abundance in the corneous endosperm (Khoddami et al., 2023). The application of MWT resulted in a disruption of the native structure of all matrices, causing partial fusion of the structures and a loss of integrity of both the starch and protein constituents. Other studies that have evaluated the MWT of flours or grains have reported similar observations in the surface aspect of particles after treatment with maize grain (An et al., 2023) and rice flour (Solaesa et al., 2022). These observations have been linked to the partial pasting of starch granules with amylose exudation, and to protein denaturation, resulting in the formation of larger agglomerates. The treatment of flour resulted in particles with a surface that had more fused structures than the grain treatment. This could be due to continuous exposure of the particle surface to the surrounding water. In contrast, the starch in the inner part of the grain would have been exposed to less water, especially towards the end of the treatment, limiting the modification. It is important to note that the modification may not be homogeneous within the flour particles and grains due to the non-uniform distribution of water inside them during treatment. These differences may lead to variations in the resulting flour characteristics given that water plays a key role in MWT modification.

3.5. Colour characteristics

Table 2 presents the colour characteristics of the flours obtained after MWT of flours and grains. The colour parameters were significantly ($p < 0.001$) influenced by matrix, treatment, and their interaction. For all matrices, MWT significantly reduced Luminosity (L*) and increased Chroma (C*) values, with a more pronounced effect observed when the treatment was performed in flour. Amaranth, buckwheat, and quinoa showed higher hue (h) in the native flour than in the treated flour, whereas sorghum showed the opposite effect. Obtaining a darker and more vivid colour is a common result after MWT of grains and flours, while the modification of hue varied depending on the matrix and treatment (Sharanagat et al., 2019; Solaesa et al., 2021; Vicente et al., 2023a, 2023b). The colour change has been associated with Maillard reaction and caramelisation (Solaesa et al., 2021), and polyphenol oxidation (Sharanagat et al., 2019) during MWT. The colour difference (ΔE) was higher when MWT was performed on flour, reaching up to 13.6 for TF-B, the matrix with the highest polyphenol content (Alvarez-Jubete et al., 2010). However, the ΔE was much lower for grain treatment, with a maximum of 5.1 for TG-B and less than 5 for the other matrices. Notably, $\Delta E < 5$ is barely perceptible to the human eye (García-Viguera & Zafrilla, 2001). Performing MWT on flours would have increased the contact between polyphenols and enzymes, and between proteins and reducing sugars, thus facilitating the oxidation of polyphenols and the Maillard reaction, respectively, resulting in more coloured compounds that increased ΔE. However, an intact grain structure would have limited the contact between the compounds involved in the reactions, thereby limiting them and reducing ΔE. Therefore, performing MWT on grains appears to be an effective approach for minimizing the effect of thermal treatment on colour.

3.6. Thermal properties

Table 3 presents the thermal properties of the flour samples obtained by DSC, and the thermograms are shown in Supplementary Fig. 3. The gelatinisation scan, performed on fresh samples, showed two endothermic peaks. The first peak, associated with gelatinisation, is related to the melting of amylopectin crystallites (Biliaderis, 2009). The gelatinisation enthalpy, ΔH_{gel}, decreased with treatment for all matrices, ranging from −9% (TF-A) to −29% (TF-Q). There were no significant differences ($p > 0.05$) between grain and flour treatments for the same matrix. The decrease in ΔH_{gel} was accompanied by an increase in the onset, peak, and endset temperatures ($T_{o\text{-}gel}$, $T_{p\text{-}gel}$, and $T_{e\text{-}gel}$, respectively) that varied between +1.9 °C (TG-Q) and +4.3 °C (TG-B) for $T_{o\text{-}gel}$. Amaranth and buckwheat exhibited a delay in $T_{o\text{-}gel}$ when comparing grain treatment with flour treatment (+0.5 °C for TG-A compared to TF-A, and +1.5 °C for TG-B compared to TF-B). In contrast, quinoa and sorghum showed the opposite behaviour, with greater delay when the treatment was applied to flour (+1.1 °C for TF-Q compared to TG-Q and +0.9 °C for TF-S compared to TG-S). This behaviour, previously reported for MWT of various matrices, may be mainly attributed to rearrangements of crystalline and amorphous regions of starch during MWT (Zhao et al., 2024). The starch would have partially gelatinised, with greater disruption of the weaker crystals and the double helix structure within the starch, resulting in a decrease in ΔH_{gel} and a delay in $T_{o\text{-}gel}$ (An et al., 2023; Cao et al., 2022). The rearrangements between the amorphous and crystalline regions may have improved

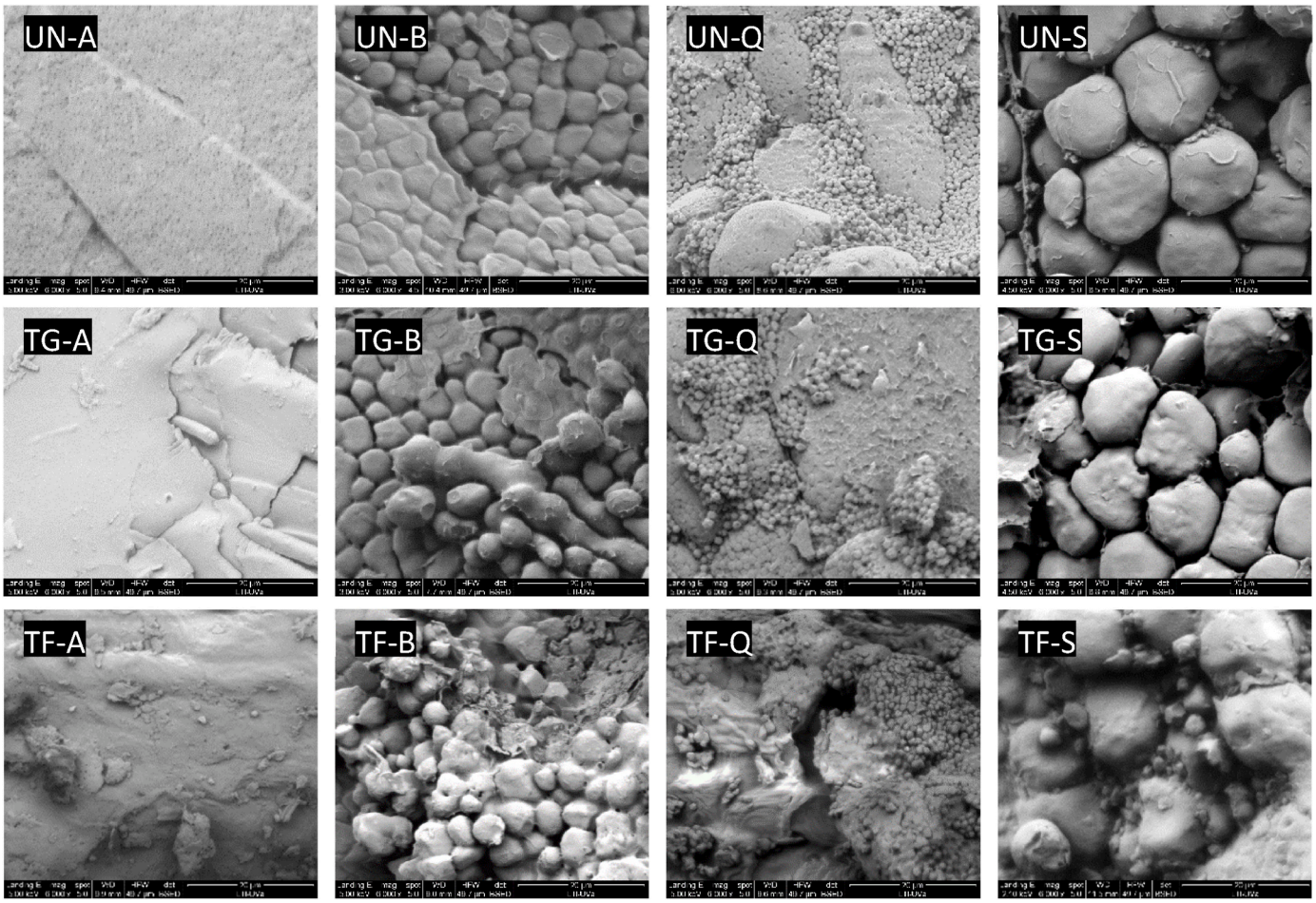


Fig. 3. Scanning electron microscopy (SEM) images of untreated-native grain (UN), microwave-treated grain (TG), and microwave-treated flour (TF) of amaranth (A), buckwheat (B), quinoa (Q), and sorghum (S) at a magnification of 6000×.

amylose-amylose and amylose-amylopectin interactions, resulting in increased $T_{o\text{-}gel}$, $T_{p\text{-}gel}$, and $T_{e\text{-}gel}$ (An et al., 2023; Zhao et al., 2024).

The second peak of the gelatinisation scan, which occurred at 94–98 °C, is typically associated with amylose-lipid dissociation (Biliaderis, 2009), although it could also be related to protein denaturation in flours with high protein content, as some proteins exhibit a denaturation peak at these temperatures (Janssen et al., 2017). This peak was detectable in all native samples, although it had a very low enthalpy (ΔH_2 1st) for amaranth and quinoa (<0.5 J/g db). MWT reduced ΔH_2 1st for sorghum, buckwheat, and quinoa (with no detectable peak for TF-Q and TG-Q), but no significant differences were observed for amaranth. In the retrogradation scan, the second peak was not detected in amaranth and quinoa samples, although it was observed in the first scan. The peak for amylose-lipid dissociation is reversible (Biliaderis, 2009). Consequently, the second peak observed in the gelatinisation scan of quinoa and amaranth may be attributed to protein denaturation, as it was not found to be reversible because it did not reappear in the retrogradation scan. Even after successive cooling and heating cycles, the complex failed to reform (data not shown). Quinoa and amaranth protein isolates have been shown to have a protein denaturation peak around 94–100 °C (Janssen et al., 2017). In addition, the disappearance of the peak for quinoa-treated samples and the slight reduction (not significant, $p > 0.05$) for amaranth-treated samples may be related to partial protein denaturation during the MWT. For UN-B and UN-S, a slightly higher enthalpy of the second peak was observed in the retrogradation scan than in the gelatinisation scan. This behaviour is consistent with that of amylose-lipid dissociation, as an increase in the enthalpy of this peak upon reheating has been commonly reported because of the better conditions for complex formation when amylose has already leaked from starch granules during gelatinisation (Eliasson, 1994). However, a significant reduction in enthalpy ($p < 0.05$) was observed after MWT of buckwheat and sorghum when applied in grain form, but not when applied to flour. This behaviour has already been reported for treated buckwheat grains and linked to the formation of amylose-polyphenol complexes (Vicente et al., 2023a).

The first peak in the retrogradation scan was identified in all samples and was related to the melting of recrystallised amylopectin (Biliaderis, 2009). Amaranth showed a minor retrogradation peak that was not affected by the treatment. Buckwheat, quinoa, and sorghum showed an increased degree of retrogradation after MWT. Increased retrogradation enthalpy has been reported after MWT of buckwheat (Vicente et al., 2023a), quinoa (Vicente et al., 2023b), and rice (Villanueva et al., 2018), whereas no significant increase was observed for tef (Calix-Rivera et al., 2023), quinoa (Vicente et al., 2023b), and rice (Solaesa et al., 2022). These results highlight the significant influence of both matrix and treatment conditions on starch retrogradation.

3.7. Techno-functional properties

Table 4 presents the techno-functional properties of flour samples obtained from native and treated matrices, including water absorption capacity (WAC), oil absorption capacity (OAC), water absorption index (WAI), water solubility index (WSI), emulsion activity (EA), and emulsion stability (ES). The matrix, type of treatment, and their interaction significantly affected all the techno-functional properties ($p < 0.001$).

The WAC of the flours increased significantly after the MWT for all

Table 3
Thermal properties of flour samples obtained from untreated and microwave-treated flours and grains.

Sample	ΔH_{gel} (J/g db)	$T_{p\text{-}gel}$ (°C)	$T_{o\text{-}gel}$ (°C)	$T_{e\text{-}gel}$ (°C)	ΔH_2 1st (J/g db)	$T_{p\text{-}2}$ 1st (°C)	ΔH_{ret} (J/g db)	$T_{p\text{-}ret}$ (°C)	%DR	ΔH_2 2nd (J/g db)	$T_{p\text{-}2}$ 2nd (°C)
UN-A	8.7 ± 0.5 b	73.7 ± 0.1 a	67.7 ± 0.2 a	80.6 ± 0.3 a	0.45 ± 0.08 a	96.3 ± 0.4 c	0.5 ± 0.1 a	nd	6 ± 2 a	nd	nd
TF-A	7.9 ± 0.1 ab	75.9 ± 0.2 b	70.3 ± 0.1 b	82.0 ± 0.2 b	0.32 ± 0.01 a	94.0 ± 0.1 a	0.5 ± 0.1 a	nd	7 ± 1 a	nd	nd
TG-A	7.7 ± 0.1 a	76.3 ± 0.2 b	70.9 ± 0.1 c	82.9 ± 0.4 b	0.37 ± 0.01 a	95.6 ± 0.1 b	0.8 ± 0.1 a	nd	10 ± 2 a	nd	nd
UN-B	10.7 ± 0.2 b	68.5 ± 0.2 a	61.3 ± 0.2 a	75.1 ± 0.1 a	1.10 ± 0.23 b	97.1 ± 0.5 a	2.1 ± 0.2 a	49.4 ± 0.5 a	20 ± 2 a	1.5 ± 0.4 b	96.0 ± 1.5 b
TF-B	8.1 ± 0.1 a	70.1 ± 0.1 b	64.1 ± 0.2 b	76.5 ± 0.1 b	0.89 ± 0.07 ab	96.2 ± 0.7 a	2.7 ± 0.1 ab	49.6 ± 1.3 a	33 ± 1 ab	0.9 ± 0.2 ab	91.5 ± 0.9 ab
TG-B	7.7 ± 0.1 a	70.9 ± 0.1 c	65.5 ± 0.1 c	76.6 ± 0.1 b	0.62 ± 0.05 a	96.1 ± 0.3 a	3.4 ± 0.5 b	48.3 ± 1.3 a	44 ± 7 b	0.4 ± 0.3 a	90.9 ± 2.1 b
UN-Q	10.6 ± 0.4 b	72.4 ± 0.3 a	64.0 ± 0.3 a	79.9 ± 0.5 a	0.28 ± 0.03	96.8 ± 0.7	1.0 ± 0.1 a	47.9 ± 0.9 ab	10 ± 2 a	nd	nd
TF-Q	7.6 ± 0.1 a	73.6 ± 0.3 b	67.1 ± 0.3 c	80.9 ± 0.2 a	nd	nd	1.6 ± 0.2 b	47.0 ± 0.4 a	21 ± 2 b	nd	nd
TG-Q	8.0 ± 0.3 a	73.2 ± 0.2 ab	65.9 ± 0.2 b	80.8 ± 0.2 a	nd	nd	2.1 ± 0.1 c	49.3 ± 0.7 b	27 ± 1 c	nd	nd
UN-S	10.7 ± 0.6 b	70.4 ± 0.1 a	63.5 ± 0.2 a	78.1 ± 0.4 a	1.12 ± 0.09 b	95.3 ± 0.2 a	4.9 ± 0.5 a	50.3 ± 0.1 a	43 ± 6 a	1.3 ± 0.3 b	94.4 ± 0.6 a
TF-S	8.7 ± 0.5 a	73.5 ± 0.3 b	67.6 ± 0.4 c	80.8 ± 0.1 b	0.71 ± 0.11 a	97.3 ± 0.4 b	6.0 ± 0.2 b	50.4 ± 0.1 a	69 ± 2 b	1.4 ± 0.1 b	94.4 ± 1.1 a
TG-S	8.1 ± 0.3 a	73.8 ± 0.1 b	66.7 ± 0.2 b	82.9 ± 0.5 c	0.61 ± 0.01 a	97.8 ± 0.6 b	5.8 ± 0.2 ab	50.5 ± 0.2 a	71 ± 5 b	0.7 ± 0.2 a	94.8 ± 0.5 a
Analysis of variance and significance (p-values)											
Matrix (F1)	***	***	***	***	***	***	***	***	***	***	*
Treatment (F2)	***	***	***	***	***	ns	***	ns	***	**	ns
F1 × F2	**	***	***	***	*	***	*	ns	**	**	*

Flours obtained from untreated-native grain (UN), microwave-treated grain (TG), and microwave-treated flour (TF) of amaranth (A), buckwheat (B), quinoa (Q), and sorghum (S). ΔH_{gel}: starch gelatinisation associated enthalpy; $T_{p\text{-}gel}$, $T_{o\text{-}gel}$, and $T_{e\text{-}gel}$: peak, onset, and endset temperatures for gelatinisation peak, respectively; ΔH_2 1st and $T_{p\text{-}2}$ 1st: enthalpy and peak temperature associated with the second peak observed on first scan on fresh sample, respectively; ΔH_{ret}: enthalpy associated with the melting of recrystallised amylopectin after 7 days of storage at 4 ± 2 °C; $T_{p\text{-}ret}$: peak temperature of melting of recrystallised amylopectin; DR: degree of retrogradation; ΔH_2 1st and $T_{p\text{-}2}$ 1st: enthalpy and peak temperature associated with the second peak observed on second scan on retrograded sample, respectively; nd: non-detectable; db: dry basis. Data is expressed as mean ± standard deviation. Significant statistical differences ($p < 0.05$) are indicated by different letters for the same parameter and matrix. Analysis of variance and significance of matrix (A,B,Q,S), treatment (UN, TG, TF), and their interaction: ***$p < 0.001$, **$p < 0.01$, *$p < 0.05$, ns: non-significant.

the matrices, indicating a greater affinity for water. WAC showed a higher increase when the treatment was performed on grains, with amaranth exhibiting the most notable change. The WAC of amaranth increased by 26% and 63% for TF-A and TG-A, respectively, compared with that of UN-A. The higher hydrophilicity of flour observed following MWT has been reported to be a result of the disruption of cell walls and membranes (An et al., 2023), disruption of hydrogen bonds between the amorphous and crystalline regions of starch (Solaesa et al., 2021), formation of a more porous structure, and increase in hydrophilic sites of protein due to its unfolding/dissociation (Kheto et al., 2022). Solaesa et al. (2021) observed a relationship between treatment at higher MC and an increase in the WAC of microwaved rice flours. Therefore, the higher impact of the treatment on grains could be explained by the higher water retention within the starch granules during the treatment. The OAC of the flours showed no significant differences when treatment was performed on amaranth and quinoa. However, significant differences were observed when buckwheat and sorghum were treated, highlighting an increase in OAC of 12% for TF-B and 7% for TF-S compared with their corresponding untreated flours. Previous studies have reported reductions in OAC in microwave-roasted sorghum (Sharanagat et al., 2019) and quinoa (Kheto et al., 2022). Conversely, Rao et al. (2023) observed an increase in MWT millet protein. Additionally, An et al. (2023) found slight/non-significant variations for MWT corn flour. These previous results, in addition to our findings, indicate that MWT has a limited effect on OAC, with the observed effects varying depending on the matrix and treatment conditions.

Amaranth and quinoa displayed the highest WAI values among the native samples, 8.6 g/g for UN-A and 7.3 g/g for UN-Q. Following MWT, a considerable reduction in this parameter was observed for these matrices, with values reaching 5.4 g/g for TF-A and TF-Q. However, sorghum displayed an inverse pattern, with an increase in WAI from 6.2 for UN-S to 6.9 for TF-S. The WSI followed an opposite trend to that of WAI. For amaranth and quinoa, an increase was observed with MWT, particularly high for TF-A (+49% compared to UN-A) and non-significant for TG-Q. Nevertheless, sorghum presented a pronounced decrease in WSI for TF-S compared with UN-S. Buckwheat presented intermediate behaviour, with small variations in WAI and WSI due to MWT. Both increases and decreases in WAI and WSI have been reported after MWT depending on the matrix and treatment conditions. The reduction in WAI has been attributed to the structural damage and long-chain rupture of amylopectin, an increase in intermolecular and intramolecular forces through hydrogen bonding, and the formation of amylose-lipid complexes (Vicente et al., 2023a; Zhi et al., 2022). However, the increase in WAI has been attributed to the weakening of intramolecular bonds between amylose and amylopectin molecules, reduced crystallinity, enhanced amylose-water interactions, and interactions of starch with proteins and lipids (An et al., 2023; Solaesa et al., 2021). The differing compositions and starch and protein characteristics, in addition to the varying water availability during MWT, may have led to the observed differences between treatments and matrices, which in turn favoured one or another phenomenon and resulted in different impacts on the values of WAI and WSI.

All flours exhibited a significant reduction or complete loss of their emulsion properties (EA and ES) following MWT. The emulsion

Table 4
Techno-functional properties of flour samples obtained from untreated and microwave-treated flours and grains.

Sample	WAC (g/g)	OAC (g/g)	WAI (g/g)	WSI (g/100g)	EA (%)	ES (%)
UN-A	1.20 ± 0.03 a	1.13 ± 0.01 ab	8.6 ± 0.1 c	5.3 ± 0.2 a	54.5 ± 0.4 c	50.9 ± 2.3 b
TF-A	1.51 ± 0.02 b	1.15 ± 0.02 b	5.4 ± 0.2 a	7.8 ± 0.3 c	<0.1 a	–
TG-A	1.96 ± 0.04 c	1.11 ± 0.02 a	6.5 ± 0.3 b	5.8 ± 0.2 b	12.1 ± 0.1 b	6.4 ± 1.8 a
UN-B	1.14 ± 0.02 a	0.97 ± 0.01 a	6.8 ± 0.2 b	4.2 ± 0.1 b	58.6 ± 0.6 c	34.8 ± 0.8 c
TF-B	1.67 ± 0.04 b	1.08 ± 0.01 c	6.8 ± 0.1 b	3.9 ± 0.2 a	9.5 ± 0.7 b	6.1 ± 0.5 b
TG-B	1.70 ± 0.04 b	1.01 ± 0.01 b	6.3 ± 0.1 a	4.5 ± 0.1 c	7.1 ± 0.9 a	3.7 ± 0.8 a
UN-Q	1.09 ± 0.01 a	1.09 ± 0.02 a	7.3 ± 0.2 b	5.1 ± 0.2 a	52.6 ± 0.3 c	44.9 ± 0.5 b
TF-Q	1.84 ± 0.04 b	1.10 ± 0.02 a	5.4 ± 0.3 a	5.8 ± 0.3 b	<0.1 a	–
TG-Q	1.92 ± 0.02 c	1.11 ± 0.01 a	5.5 ± 0.3 a	5.2 ± 0.1 a	8.5 ± 0.3 b	4.7 ± 0.4 a
UN-S	1.34 ± 0.02 a	1.05 ± 0.01 b	6.2 ± 0.2 a	3.7 ± 0.1 b	39.3 ± 1.4 b	8.6 ± 0.5
TF-S	1.62 ± 0.03 b	1.12 ± 0.01 c	6.9 ± 0.1 b	2.1 ± 0.1 a	<0.1 a	–
TG-S	1.73 ± 0.02 c	1.02 ± 0.02 a	6.2 ± 0.2 a	3.5 ± 0.1 b	<0.1 a	–
Analysis of variance and significance (p-values)						
Matrix (F1)	***	***	***	***	***	***
Treatment (F2)	***	***	***	***	***	***
F1 × F2	***	***	***	***	***	***

Flours obtained from untreated-native grain (UN), microwave-treated grain (TG), and microwave-treated flour (TF) of amaranth (A), buckwheat (B), quinoa (Q), and sorghum (S). WAC: water absorption capacity, OAC: oil absorption capacity, WAI: water absorption index, WSI: water solubility index, EA: emulsifying activity, ES: emulsion stability. Data is expressed as mean ± standard deviation. Significant statistical differences ($p < 0.05$) are indicated by different letters for the same parameter and matrix. Analysis of variance and significance of matrix (A,B,Q,S), treatment (UN, TG, TF), and their interaction: ***$p < 0.001$, **$p < 0.01$, *$p < 0.05$, ns: non-significant.

properties of flours are highly related to their protein fraction characteristics (Vicente et al., 2023a). Both increases and decreases in EA and ES have been reported after MWT, depending on the matrix and the treatment. MWT may induce partial unfolding of proteins, resulting in greater exposure of hydrophobic groups and secondary structural modification, which enhances protein absorption at the oil/water interface (Rao et al., 2023; Vicente et al., 2023a). However, for more intense treatment conditions, such as high moisture content, high temperature, and/or long duration, as those in our study, the ability of the protein to form and stabilise emulsions may be reduced by heat-induced denaturation and aggregation, resulting in the formation of larger protein aggregates that are unable to stabilise the oil/water interface (Kheto et al., 2022; Vicente et al., 2023b).

4. Conclusions

The outcomes of this study indicate that the efficacy of MWT in modifying the properties of a matrix is greatly influenced by the botanical origin of the treated system and its form during treatment, whether it is in flour or grain form. The MWT proved effective in changing the microstructure, thermal properties, and techno-functional properties of the resulting flours. All native samples displayed comparable microwave absorption capacities, with water being the only significant absorber in the studied matrices. This was corroborated by the observation that equivalent temperature curves were obtained when different samples were subjected to the same treatment conditions, with the same microwave effective time being required. However, significant differences were observed in the mobility of water during heating. The rate of water loss during heating was remarkably higher in the flours, with the grains exhibiting significant differences depending on the matrix. Therefore, the observed differences in flour microstructure, thermal properties, and techno-functional properties may be attributed not only to the different matrix compositions but also to the different distribution and mobility of water within the grains during MWT. Moreover, this study revealed that the selection of the matrix form (grain or flour) subjected to treatment may potentiate a specific property, thereby enabling the production of a wider range of modified flours with adapted functionality. For instance, grain treatment was found to be effective in mitigating the evident difference in colour associated with flour treatment, while simultaneously enhancing the water absorption capacity of the flour. Additional research is still required to clarify the molecular/structural basis of the distinct impacts of the MWT on matrices of varying sources and forms, with a particular focus on the relationship between structure and function in microwave modification.

CRediT authorship contribution statement

Ainhoa Vicente: Writing – review & editing, Writing – original draft, Visualization, Methodology, Investigation, Formal analysis, Conceptualization. **Marina Villanueva:** Writing – review & editing, Visualization, Validation, Methodology, Conceptualization. **Jose María Muñoz:** Writing – review & editing, Software, Methodology, Formal analysis, Conceptualization. **Pedro A. Caballero:** Writing – review & editing, Supervision, Resources, Conceptualization. **Felicidad Ronda:** Writing – review & editing, Validation, Supervision, Resources, Project administration, Methodology, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors confirm that they have no conflicts of interest with respect to the work described in this manuscript.

Data availability

Data will be made available on request.

Acknowledgements

The authors thank the Spanish Ministerio de Ciencia e Innovación (PID2019-110809RB-I00 and EQC2021-006985-P), Ministerio de Ciencia, Innovación y Universidades (PID2023-153330OB-I00), and the Junta de Castilla y León/FEDER (VA195P20 and CLU 2019–04 – BIO-ECOUVA Unit of Excellence of the University of Valladolid) for their financial support. A. Vicente thanks the Spanish Ministerio de Ciencia, Innovación y Universidades for her FPU doctorate grant. The authors thank Joanna Harasym for assisting with the supply of buckwheat grain.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.foodhyd.2024.110680.

References

AACC. (2010). Approved methods of analysis. *Cereals & grains association* (11th ed.).
Abebe, W., Collar, C., & Ronda, F. (2015). Impact of variety type and particle size distribution on starch enzymatic hydrolysis and functional properties of tef flours. *Carbohydrate Polymers, 115*, 260–268. https://doi.org/10.1016/j.carbpol.2014.08.080
Alvarez-Jubete, L., Arendt, E. K., & Gallagher, E. (2010). Nutritive value of pseudocereals and their increasing use as functional gluten-free ingredients. *Trends in Food Science and Technology, 21*(2), 106–113. https://doi.org/10.1016/j.tifs.2009.10.014

An, N., Li, D., Wang, L., & Wang, Y. (2023). Microwave irradiation of corn kernels: Effects on structural, thermal, functional and rheological properties of corn flour. *Food Hydrocolloids, 143*, Article 108939. https://doi.org/10.1016/j.foodhyd.2023.108939

AOAC. (2023). In G. W. Latimer (Ed.), *Official methods of analysis of AOAC international* (22nd ed.). Oxford University Press.

Biliaderis, C. G. (2009). Structural transitions and related physical properties of starch. In R. L. Whistler, & J. N. BeMiller (Eds.), *Starch: Chemistry and technology* (3rd ed., pp. 293–372). Academic Press. https://doi.org/10.1016/B978-0-12-746275-2.00008-2.

Calix-Rivera, C. S., Villanueva, M., Náthia-Neves, G., & Ronda, F. (2023). Changes on techno-functional, thermal, rheological, and microstructural properties of tef flours induced by microwave radiation—development of new improved gluten-free ingredients. *Foods, 12*(6), 1345. https://doi.org/10.3390/foods12061345

Cao, H., Sun, R., Liu, Y., Wang, X., Guan, X., Huang, K., & Zhang, Y. (2022). Appropriate microwave improved the texture properties of quinoa due to starch gelatinization from the destructed cyptomere structure. *Food Chemistry X, 14*, Article 100347. https://doi.org/10.1016/J.FOCHX.2022.100347

Eliasson, A. C. (1994). Interactions between starch and lipids studied by DSC. *Thermochimica Acta, 246*(2), 343–356. https://doi.org/10.1016/0040-6031(94)80101-0

Englyst, K. N., Hudson, G. J., & Englyst, H. N. (2006). Starch analysis in food. In *Encyclopedia of analytical chemistry*. John Wiley & Sons, Ltd. https://doi.org/10.1002/9780470027318.a1029.

García-Viguera, C., & Zafrilla, P. (2001). Changes in anthocyanins during food processing: Influence on color. *ACS Symposium Series, 775*, 56–65. https://doi.org/10.1021/BK-2001-0775.CH004

Gibson, T. S., Solah, V. A., & McCleary, B. V. (1997). A procedure to measure amylose in cereal starches and flours with Concanavalin A. *Journal of Cereal Science, 25*(2), 111–119. https://doi.org/10.1006/jcrs.1996.0086

Janssen, F., Pauly, A., Rombouts, I., Jansens, K. J. A., Deleu, L. J., & Delcour, J. A. (2017). Proteins of amaranth (*Amaranthus* spp.), buckwheat (*fagopyrum* spp.), and quinoa (*Chenopodium* spp.): A food science and technology perspective. *Comprehensive Reviews in Food Science and Food Safety, 16*(1), 39–58. https://doi.org/10.1111/1541-4337.12240

Kaushal, P., Kumar, V., & Sharma, H. K. (2012). Comparative study of physicochemical, functional, antinutritional and pasting properties of taro (*Colocasia esculenta*), rice (*Oryza sativa*) flour, pigeonpea (*Cajanus cajan*) flour and their blends. *LWT - Food Science and Technology, 48*(1), 59–68. https://doi.org/10.1016/j.lwt.2012.02.028

Kheto, A., Joseph, D., Islam, M., Dhua, S., Das, R., Kumar, Y., Vashishth, R., Sharanagat, V. S., Kumar, K., & Nema, P. K. (2022). Microwave roasting induced structural, morphological, antioxidant, and functional attributes of Quinoa (*Chenopodium quinoa* Willd). *Journal of Food Processing and Preservation, 46*(5), Article e16595. https://doi.org/10.1111/jfpp.16595

Khoddami, A., Messina, V., Vadabalija Venkata, K., Farahnaky, A., Blanchard, C. L., & Roberts, T. H. (2023). Sorghum in foods: Functionality and potential in innovative products. *Critical Reviews in Food Science and Nutrition, 63*(9), 1170–1186. https://doi.org/10.1080/10408398.2021.1960793

Martínez-Villaluenga, C., Peñas, E., & Hernández-Ledesma, B. (2020). Pseudocereal grains: Nutritional value, health benefits and current applications for the development of gluten-free foods. *Food and Chemical Toxicology, 137*, Article 111178. https://doi.org/10.1016/J.FCT.2020.111178

Onwude, D. I., Hashim, N., Janius, R. B., Nawi, N. M., & Abdan, K. (2016). Modeling the thin-layer drying of fruits and vegetables: A review. *Comprehensive Reviews in Food Science and Food Safety, 15*(3), 599–618. https://doi.org/10.1111/1541-4337.12196

Perez-Rea, D., & Antezana-Gomez, R. (2018). The functionality of pseudocereal starches. In *Starch in food: Structure, function and applications* (2nd ed., pp. 509–542). Elsevier Inc. https://doi.org/10.1016/B978-0-08-100868-3.00012-3.

Rao, M. V., Sunil, C. K., & Venkatachalapathy, N. (2023). Effect of microwave and hot air radiofrequency treatments on physicochemical and functional properties of foxtail millet flour and its protein isolate. *Journal of Cereal Science, 114*, Article 103774. https://doi.org/10.1016/J.JCS.2023.103774

Sadaka, S. (2022). Impact of grain layer thickness on rough rice drying kinetics parameters. *Case Studies in Thermal Engineering, 35*, Article 102026. https://doi.org/10.1016/J.CSITE.2022.102026

Schafranski, K., Ito, V. C., & Lacerda, L. G. (2021). Impacts and potential applications: A review of the modification of starches by heat-moisture treatment (HMT). *Food Hydrocolloids, 117*, Article 106690. https://doi.org/10.1016/j.foodhyd.2021.106690

Sharanagat, V. S., Suhag, R., Anand, P., Deswal, G., Kumar, R., Chaudhary, A., Singh, L., Singh Kushwah, O., Mani, S., Kumar, Y., & Nema, P. K. (2019). Physico-functional, thermo-pasting and antioxidant properties of microwave roasted sorghum [*Sorghum bicolor* (L.) Moench]. *Journal of Cereal Science, 85*, 111–119. https://doi.org/10.1016/j.jcs.2018.11.013

Solaesa, Á. G., Villanueva, M., Muñoz, J. M., & Ronda, F. (2021). Dry-heat treatment vs. heat-moisture treatment assisted by microwave radiation: Techno-functional and rheological modifications of rice flour. *Lebensmittel-Wissenschaft und -Technologie, 141*, Article 110851. https://doi.org/10.1016/j.lwt.2021.110851

Solaesa, Á. G., Villanueva, M., Vela, A. J., & Ronda, F. (2022). Impact of microwave radiation on in vitro starch digestibility, structural and thermal properties of rice flour. From dry to wet treatments. *International Journal of Biological Macromolecules, 222*, 1768–1777. https://doi.org/10.1016/j.ijbiomac.2022.09.262

Sosa-Morales, M. E., Valerio-Junco, L., López-Malo, A., & García, H. S. (2010). Dielectric properties of foods: Reported data in the 21st Century and their potential applications. *LWT - Food Science and Technology, 43*(8), 1169–1179. https://doi.org/10.1016/J.LWT.2010.03.017

Tao, Y., Yan, B., Fan, D., Zhang, N., Ma, S., Wang, L., Wu, Y., Wang, M., Zhao, J., & Zhang, H. (2020). Structural changes of starch subjected to microwave heating: A review from the perspective of dielectric properties. *Trends in Food Science & Technology, 99*, 593–607. https://doi.org/10.1016/j.tifs.2020.02.020

Togrul, I. T., & Pehlivan, D. (2002). Mathematical modelling of solar drying of apricots in thin layers. *Journal of Food Engineering, 55*(3), 209–216. https://doi.org/10.1016/S0260-8774(02)00065-1

Vicente, A., Villanueva, M., Caballero, P. A., Muñoz, J. M., & Ronda, F. (2023b). Microwave modification of quinoa grains at constant and varying water content modulates changes in structural and physico-chemical properties of the resulting flours. *Foods, 12*(7), 1421. https://doi.org/10.3390/foods12071421

Vicente, A., Villanueva, M., Caballero, P. A., Muñoz, J. M., & Ronda, F. (2023a). Buckwheat grains treated with microwave radiation: Impact on the techno-functional, thermal, structural, and rheological properties of flour. *Food Hydrocolloids, 137*, Article 108328. https://doi.org/10.1016/j.foodhyd.2022.108328

Villanueva, M., Harasym, J., Muñoz, J. M., & Ronda, F. (2018). Microwave absorption capacity of rice flour. Impact of the radiation on rice flour microstructure, thermal and viscometric properties. *Journal of Food Engineering, 224*, 156–164. https://doi.org/10.1016/j.jfoodeng.2017.12.030

Yang, Q., Qi, L., Luo, Z., Kong, X., Xiao, Z., Wang, P., & Peng, X. (2017). Effect of microwave irradiation on internal molecular structure and physical properties of waxy maize starch. *Food Hydrocolloids, 69*, 473–482. https://doi.org/10.1016/j.foodhyd.2017.03.011

Zhao, Y., Tu, D., Wang, D., Xu, J., Zhuang, W., Wu, F., & Tian, Y. (2024). Structural and property changes of starch derivatives under microwave field: A review. *International Journal of Biological Macromolecules, 256*, Article 128465. https://doi.org/10.1016/J.IJBIOMAC.2023.128465

Zhi, W., Zhou, Y., Wang, R., Wang, M., Wang, W., Hu, A., & Zheng, J. (2022). Effect of microwave treatment on the properties of starch in millet kernels. *Starch - Stärke, 74*(9–10), Article 2200063. https://doi.org/10.1002/STAR.202200063