

Sustainable Local Gluten-free Flours: Nutritional, Physical, and Functional Characterization

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Abstract Most of gluten-free (GF) bakery products available on the market are made with a restricted number of grains. Flours and starches from rice and maize are mainly used; for this reason, people affected by celiac disease frequently suffer from nutritional deficiencies. The use of a wider range of GF flours, rich in nutrients and phytochemicals, may improve the nutritional quality of GF products. In this work, an investigation of the physicochemical, mineral, and functional profiles in 16 GF flours belonging to different local varieties of legumes (chickpea, fava bean, and lentil), cereals (rice and maize) and pseudo-cereals (white quinoa, red quinoa and chia) was achieved. Significant differences could be observed across samples, legume flours presented very interesting nutritional characteristics: proteins (22 % - 28.71%), mineral composition especially in iron (52.05ppm to 152.45ppm), Zinc (36.96% - 51.28%) and Potassium (0.83% -1.13%); moreover, pulses were characterized by their high antioxidant activity (88.63%). Besides the important functional properties, such as the emulsifying activity and stability as well as the foaming capacity and foam stability, were noticed especially for lentil and fava bean flours. Finally, such interesting properties contribute to the selection of flours for healthier GF bakery products.

Keywords: flours, gluten-free, functional properties, physicochemical characterization

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1. Introduction

Celiac disease (CD) is an autoimmune system disorder and inflammatory disease triggered by gluten in the upper small intestine [1,2]. The symptoms of CD are malnutrition, diarrhea, growth retardation, anemia, and fatigue [3]. CD is a global health problem affecting approximately 1% of the world population [4,5]. Thus, cereals containing gluten include mainly wheat, rye, barley and probably oat, shall not be included in diets for CD patients. The “Codex Alimentarius” [6] defines GF food as the ingredients that don’t contain gluten or the food that has been specially treated to remove gluten, where the GF foods should not contain gluten, or the presence of gluten should not exceed 20 mg/kg in total. To date, the only treatment for people suffering from CD is to follow a GF diet [7]. Many GF flours/starches have been used to substitute gluten-containing ingredients to produce GF food, such as cereal, pseudo cereals, and legumes. Rice (*Oryza sativa* L.), maize (*Zea mays* L.), and sorghum

(*Sorghum bicolor* L. Moench) are GF cereals, distantly related to wheat, and are considered safe for individuals with celiac disease. In addition, a number of species of millets and the Ethiopian cereal teff are also GF flours [8]. Besides of GF cereals, pseudo cereals (quinoa, buckwheat, amaranth, and chia) are a current trend in human diets as they are GF grains and have an excellent nutritional value. It has been shown that pseudo cereals contain high concentration (13.2–18.2%) of protein and a good balanced amino acid composition characterized by abundant amounts of sulfur-rich amino acids [9]. They are rich in starch, fiber and minerals (calcium, iron and zinc), vitamins, and phytochemicals such as saponins, polyphenols, phytosterols, phytosteroids, and betalains with potential health benefits [10].

Recently, research focused the attention on the use of legumes as ingredients in GF products [11,12,13,14] because of their functional and nutritional properties, as well as of their role in sustainable diets. Chickpea (*Cicer arietinum* L.) is a legume rich in protein, dietary fiber, carbohydrates, folate and trace minerals (Fe, Mo, Mn) [15]. Some authors have studied functional properties of

chickpea proteins, reporting good emulsifying and foaming characteristics [16,17] as well as high oil absorption capacity [18]. Besides of chickpea, Fava bean (*Vicia faba*) is a legume rich in essential amino acids: isoleucine, leucine, lysine, phenylalanine, threonine and valine [19,20] and when combined with cereal ingredients make a product with well-balanced amino acid composition [21]. Fava bean is also rich in dietary fiber, minerals, and non-nutrient secondary metabolites recognized as beneficial in human health [22]. Moreover, lentil (*Lens culinaris*) flour is GF and may be added to cereal flour to make bread, cakes, and baby foods [23]. Beyond their nutrition functions, lentils have several potential health-promoting effects. Furthermore, Siddique, Johansen [24] widely documented the importance of legumes in sustainable cropping systems. It is expected that future climatic conditions will be more favorable for the cultivation of common beans in the Northern Hemisphere than in the Southern Hemisphere [25]. New areas for pulse crops are expected in U.S.A, Canada, Europe and northern Asia. In Finland, the cultivation of peas and fava beans is already gaining in importance [26]. Predicted climate changes should increase yields of dry peas, chickpeas, beans, lentils, lupine and grass pea in developed countries like Canada and France [27]. Besides, the transition from animal-based proteins toward greater use of legume-based foods has the potential to generate less greenhouse gas emissions, and requires less azote fertilizer, land and water resources [28].

The ultimate success of utilizing pseudo cereals and legumes as ingredients depends largely upon their nutritional qualities especially their high protein content. Functional properties also constitute the major criteria for the adoption and acceptability of proteins in the food systems. Hydration properties, water absorption and binding are known to directly influence the characteristics of a food system [29]. The aim of this research was to characterize different GF flours of cereals (rice, maize), pseudo cereals (white and red quinoa, chia) and several local varieties of legumes: chickpea (Rebha, Neyer, Joud, Nour, Beja, and Chetoui), fava bean (Badii, Bachar, and Najeh), lentil (Kef, Krib). The present study provides useful information on the efficient incorporation alone or combined of pseudo cereals and/or legumes with cereals in order to fortify and to improve GF products.

2. Materials and Methods

2.1. Materials

Sixteen raw samples of cereals rice (*Oryza sativa* L.) and maize (*Zea mays*), pseudo cereal including Quinoa (*Chenopodium quinoa* Willd.) and Chia (*Salvia hispanica* L.), legumes including chickpea (*Cicer arietinum* L.), fava bean (*Vicia faba* L.) and lentils (*Lens culinaris*) were provided by the National Institute of Agronomic Research of Tunisia (INRAT). The different varieties of legumes listed in Table 1 were ground by a hammer grinder (Retschmühle, 7311 Dettigen-Tech, Germany). Rice grain pack was bought from commercial market and transformed to flour using laboratory grinder (Cyclotec, Foss). Maize flour (Napolis, Tunisia) bought from a local

market. Red and white quinoa and chia were obtained from the INRGREF (National Research Institute for Rural Engineering, Water and Forest) and ground using a Cyclotec grinder (Foss). Flour samples were stored in refrigerator at 4°C prior to analyses.

Table 1. List of evaluated samples

Class	Variety/cultivar	Source/provider	Origin/location
Rice	Not specified (NS)	Supermarket, Tunisia	NS
Maize	NS	Napolis, Tunisia	NS
Quinoa	Red	National Research Institute for Rural Engineering, Water and Forest (INRGREF, Tunisia)	Kef, Tunisia
	White		
Chia	NS	National Research Institute for Rural Engineering, Water and Forest (INRGREF, Tunisia)	Kef, Tunisia
Lentil	Kef / Krib	National Institute of Agronomic Research of Tunisia	Kef, Tunisia
Chickpea	Neyer/ Nour / Chetoui / Beja1/ Rebha / Joud	National Institute of Agronomic Research of Tunisia	Kef, Tunisia
Fava bean	Badii / Bachar / Najeh	National Institute of Agronomic Research of Tunisia	Kef, Tunisia

2.2. Proximate Composition

Flour samples were estimated for their moisture, ash, fat and protein (N×6.25 for the legumes and N×5.7 for cereals and pseudo cereals) contents using standard methods [30]. Carbohydrates were calculated by subtracting 100 from the protein, ash, moisture and lipid levels.

2.2.1. Mineral Composition

Calcium (Ca), magnesium (Mg), potassium (K), iron (Fe) and zinc (Zn) were determined using inductively coupled plasma optical emission spectrometry (ICP-AES) using an atomic-emission spectrometer (Perkin Elmer/ Avio 200, USA). Prior to the ICP-AES measurement, the samples were mineralized by treatment with HNO₃ and H₂O₂ at 100°C until digestion was complete. These procedures were performed according to the standard methods [31].

2.2.2. Phytochemical Properties of Flours

2.2.2.1. Total Phenols

The total polyphenols content (TPP) were determined by a Folin-Ciocalteu assay [32] using Gallic Acid (GA) as the standard. The absorbance was measured at 760 nm against distilled water as a blank. The TPP was expressed as Gallic acid equivalents (mg of GAE/g DM sample) through the calibration curve of GA.

2.2.2.2. Total Flavonoids

Total flavonoid content (TF) was determined using a colorimetric method as described previously by Dewanto, Wu [32]. The results were expressed as milligram Catechin equivalents (CE) per gram of dry matter sample (DM) using the calibration curve of Catechin.

2.2.2.3. Condensed Tannins

Condensed tannin (CT) was analyzed according to the method of Sun, Ricardo-da-Silva [33] using HCl and vanillin (4%). The absorbance was measured at 500 nm against distilled water as a blank. The CT was expressed as Catechin equivalents (mg of CE/g DM sample) through the calibration curve of Catechin.

2.2.2.4. Radical Scavenging Assay

The radical-scavenging activity of the analyzed extracts was determined spectrophotometrically against DPPH radical [34,35]. Samples were prepared by mixing 2 ml of DPPH solution and 1 ml of the extracts. The measurement is based on change of absorbance which corresponds to free radical scavenging activity of the extracts. Each measurement was repeated three times at 517 nm at room temperature. The final result is the average of three replicates. The antioxidant activity can be calculated with the following formula (1):

$$\% = \frac{A_0}{A_0 - A_1} \times 100 \quad (1)$$

Where, A₀: absorbance of the reference sample, A₁: absorbance of the sample with tested extracts.

2.3. Physical Properties of Flours

2.3.1. Bulk Density:

Bulk density of flours was determined by the method suggested by Kaur and Sandhu [36]. In this method, the flour samples were gently filled into 10 ml graduated cylinders, previously tared. The bottom of each cylinder was gently tapped on a laboratory bench several times until there was no further diminution of the sample level after filling to the 10 ml mark. Bulk density was calculated as weight of sample per unit volume of sample (g/ml).

2.3.2. Water Absorption Index and Water Solubility Index

Water absorption index (WAI) and water solubility index (WSI) of flours were determined by slightly modifying the method of Anderson, Conway [37]. Flour sample (2.5 g) was dispersed in 30 ml of distilled water, using tared centrifuge tubes, and heated in a water bath at 90°C for 15 minutes. Then it was centrifuged at 3000*g for 10 min at room temperature. The supernatant was decanted for determination of its solid content into a tared evaporating dish and the sediment was weighed. The weight of dry solids was recovered by evaporating the supernatant overnight at 110°C. WAI and WSI were calculated by the equations (2 and 3):

$$WAI \left(\frac{g}{g} \right) = \frac{\text{Weight of sediment}}{\text{Weight of Flour Sample}} \quad (2)$$

$$WSI \left(\frac{g}{100g} \right) = \frac{\text{Weight of Dissolved Solids in Supernatant}}{\text{Weight of Flour Sample}} \times 100 \quad (3)$$

2.3.3. CIE Lab Color Characteristics:

Color measurements of the flour samples were carried

out in triplicate using Chroma meter CR-400 (Minolta, Japan), on the basis of CIE L^{*}, a^{*} and b^{*} values. L^{*} value indicates the lightness, 0-100 representing dark to light; the a^{*} value gives the degree of the red-green color, with a higher positive a^{*} value indicating more red. The b^{*} value indicates the degree of the yellow-blue color, with a higher positive b^{*} value indicating more yellow.

2.4. Functional Properties of Flours

2.4.1. Water and Oil Absorption Capacity

Water absorption of different flours was measured by the centrifugation method of Sosulski [38]. The sample (3 g) was dispersed in 25 ml of distilled water and placed in pre-weighed centrifuge tubes. The dispersions were stirred occasionally, held for 30 min, followed by centrifugation for 25 min at 3000*g. The supernatant was decanted, and the excess moisture was removed from centrifuge tubes when dried at 50°C/25 min in hot air oven and the sample was reweighed. For the determination of oil absorption, the method of Lin, Humbert [39] was used. Sample (0.5 g) was mixed with 6 ml of corn oil. The contents were stirred for 1 min to disperse the sample in the oil. After a holding period of 30 min, the tubes were centrifuged for 25 min at 3000*g. The separated oil was then removed with a pipette and the tubes were inverted for 25 min to drain the oil prior to reweighing. The water and oil absorption capacities were expressed as gram of water or oil bound per gram of the sample on a dry basis. WAC and OAC were calculated by the equations (4 and 5):

$$WAC = \frac{(\text{Weight of tube after drying} - \text{tube weight}) - \text{Sample weight}}{\text{Sample weight}} \quad (4)$$

$$OAC = \frac{\text{Weight of dry tube} - (\text{tube weight} + \text{sample weight})}{\text{Sample weight}} \quad (5)$$

2.4.2. Least Gelation Concentration

Least gelation concentration of flours was determined by the method of Sathe, Deshpande [40]. Test tubes containing suspensions of 2, 4, 6, 8, 10, 12, 14, 16, 18 and 20 g/100 ml of material in 5 ml distilled water were heated for one hour in boiling water, followed by rapid cooling under running water. The tubes were then cooled at 4°C for two hours. Least gelation concentration is the concentration above which the sample did not fall down or slip when the test tube was inverted.

2.4.3. Emulsifying Activity and Stability

Emulsion properties of flours were determined according to the method given by Naczka, Diosady [41] slightly modified. Flour sample (1.75 g) was homogenized for 30 s in 25 ml water using a vortex (Velp scientifica, made in Europe) at high setting. Corn oil (12.5 ml) was added, and the mixture was again homogenized for 30 seconds. Then another 12.5 ml of corn oil was added, and the mixture homogenized for 90 seconds. The emulsion was centrifuged at 1100*g for 5 min. Emulsifying activity was calculated by dividing the volume of the emulsified

layer by the volume of the emulsion before centrifugation $\times 100$. The emulsion stability was determined using the samples prepared for the measurement of emulsifying activity. They were heated for 15 min at 85°C, cooled and centrifuged again at 1100*g for 5 min. The emulsion stability was expressed as the percentage of emulsifying activity remaining after heating.

2.4.4. Foaming Capacity and Foaming Stability

Foaming properties of flours were determined according to the method given by Lin, Humbert [39]. The dispersions of sample (1.5 g in 50 ml distilled water) were homogenized, using a hand blender at high setting for 2-3 min. The blend was immediately transferred into a graduated cylinder and the homogenizer cup was rinsed with 10 ml of distilled water, which was then added to the graduated cylinder. The volume was recorded before and after whipping. Foaming capacity (FC) and Foam stability (FS) were calculated as follows (6):

$$FC = \frac{\text{Increase in Volume after homogenization} - \text{Initial volume}}{\text{Initial Volume}} \quad (6)$$

FS= Foam volume changes in the graduated cylinder was recorded at an interval of 20, 40, 60 and 120 min of storage.

2.5. Statistical Analysis

Data were expressed as mean $\pm$ standard deviations of 3 samples, Analysis of variance (ANOVA) was carried out using SPSS (IBM SPSS statistics version 25), and statistical significance was considered at $p < 0.05$. Pearson correlations analysis was done to calculate the correlations among data. Furthermore, Principal Component Analysis (PCA) was performed to visualize differences among samples using Minitab19® Statistical Software (Minitab Ltd., Coventry, UK).

3. Results and Discussion

3.1. Chemical Composition

3.1.1. Proximate Composition

Chemical composition of the 16 samples of flours from cereals, pseudo-cereals and legumes was presented in Table 2. Significant variations in chemical composition were observed ($p < 0.05$) among studied samples. Carbohydrates were the predominant nutrient in all studied flours, ranging from 32.86 to 75.25g/100 g, with statistically significant differences. The highest value was for rice flour while the lowest was for chia flour. The results are in agreement with those of Kaushal, Kumar [42] investigating the proximate composition of rice (78.45 % of total carbohydrate). High carbohydrate content makes flours a good source of metabolizable energy and assists in fat metabolism. Legume flours presented lower carbohydrate content (55.72-60.20 g/100 g) compared with rice and maize flours (75.25-71.90 g/100 g). Millar et al. [43] reported similar carbohydrate content in different varieties of pulse flours purchased from United Kingdom, of fava beans (*Vicia fava*, cv. Victor), yellow peas (*Pisum*

sativum L.) and green peas (*Pisum sativum*, cv. Large blue), (54.7-63.9 g/100 g). Fujiwara, Hall [44] have showed that replacing wheat flour by pulse ingredients (up to 50 %) has led to a reduction of total carbohydrates value. Regarding flour protein contents, it ranged from 10.26% to 28.71% with significant differences (Table 2). All three pulse flours were the richest in proteins compared to cereal and pseudo cereal flours ($p < 0.05$). These results fit with those of Rocchetti, Lucini [45] comparing chemical and antioxidant properties of GF flours from cereals, pseudo cereals and legumes. Flours from fava bean cultivars exhibited the highest average level of proteins, in particular Bachar variety, followed by lentil and chickpea flours. Masey O'Neill, Rademacher [46] reported protein contents in the proportions of 22-24.6 g/100 g in several different cultivars of fava bean. Moreover, Ghribi, Maklouf [47] exhibited similar results of protein content (20.29-24.51 g/100g) when studying nutritional and compositional properties of Desi and Kabuli Chickpea (*Cicer Arietinum* L.) flours from Tunisian cultivars. Interestingly, substituting wheat flour with 40% of fava bean flour in baked crackers led to increase protein content from 8.36 g/100 g to 13.02 g/100 g, without affecting cracker texture and consumers sensory acceptability [48]; which supports the work of Tazart, Lamacchia [49] who observed similar increase in protein content of pasta following addition of broad-bean.

As shown in Table 2, significant differences in fat were found among cereal, pseudo cereal and legume flour samples, and among individual varieties of chickpeas. The lowest value was observed for rice flour and the highest amount was for chia flour. Legume flours presented low fat content in comparison with pseudo cereals where the lentil flours (especially Krib variety) presented the lowest fat content in comparison with the other legume flours. Those results were in agreement with those of Millar, Gallagher [43], reporting fat content less than 2% for pulse flours (Fava-bean, green and yellow-pea flour). Consumption of pulses is thus recommended to prevent and to manage obesity and non-communicable diseases such as cardiovascular diseases and type 2 diabetes.

As illustrated in Table 2, ash content varied from 0.71 to 4.14% of dry matter for all flours. The lowest value was observed for rice flour and the highest was for chia flour. Similar values were found by Kaushal, Kumar [50] in rice flour. Our study clearly highlighted that legume flours constitute a good source of minerals, with chickpea (Nour variety) having the highest mineral content, in comparison with cereal and pseudo cereal flours. This confirmed findings of Millar, Gallagher [43] indicating high levels of minerals in pulse flours (2.8-3.5 g/100 g). Dietary intake of minerals can be increased in GF food products by combining pulse flours with low mineral GF cereal flours.

3.1.2. Phenolic Compounds and Antioxidant Activity

Phenolic compounds are secondary metabolites displaying antimicrobial, antioxidant, and anti-inflammatory properties [51]. They have been shown to prevent from degenerative diseases such as cancer and cardiovascular diseases [52]. In order to evaluate their potential role on the antioxidant activity of selected flours, total polyphenols (TPP) and total flavonoids (TF) contents were analyzed (Table 2). A significant variation in TPP

was found among all samples, and it ranged from 0.2 mg GAE/g to 2.11 mg GAE/g. Rice flour showed the lowest concentration in TPP (0.2 mg GAE/g), whereas Chia flour showed the highest TPP followed by fava bean (Badii variety), quinoa, and maize flours. These results are in agreement with those of Di Cairano, Condelli [53] and Millar, Barry-Ryan [48] pointing out low TPP in rice flour, and high values for pseudo cereals followed by legume flours. Interestingly, significant differences in TPP were also identified among fava bean cultivars flour as well as among chickpea cultivars. Similar results were observed

in different fava bean varieties grown in Australia, which ranged from 258 to 570 mg GAE/100 g (DW) [54]. Changes in TPP contents could be probably due to growing location and year, post-harvest storage, genotype, geographic and climatic conditions [55]. In contrast, Table 2 indicated no significant differences in TF (0.14-0.31 mg CE/g DM) among quinoa, rice, maize, and legume flours. Xu, Yuan [56] reported similar results for legume flours using different extraction methods for chickpea and lentil flours.

Table 2. Chemical characterization of cereals (rice and maize), pseudo cereals (white and red quinoa, chia) and legumes flours (lentils, fava bean, chickpea) flours

	Flours	Variety	Carbohydrate (g/100g DM)	Protein (g/100g DM)	Fat (g/100g DM)	Ash (g/100g DM)	Moisture (g/100g DM)	TPP (mg GAE/g DM)	TF (mgC/g DM)	CT (mg CE/g DM)	DPPH scavenging Activity %
Cereals	Rice <i>Oryza sativa L.</i>	NS	75.25 ±0.63 ^f	10.26 ±0.62 ^a	0.40 ±0.03 ^a	0.71 ±0.03 ^a	13.36 ±0.01 ^h	0.20 ±0.01 ^a	0.18 ±0.01 ^a	1.68 ±0.16 ^{abc}	34.65 ±1.53 ^a
	Maize <i>Zea mays</i>	NS	71.90 ±0.25 ^{ef}	10.42 ±0.20 ^a	4.15 ±0.01 ^{bcd}	0.95 ±0.0004 ^b	12.18 ±0.54 ^{gh}	1.04 ±0.06 ^{fg}	0.21 ±0.04 ^a	0.28 ±0.02 ^a	47.64 ±6.18 ^{ab}
Pseudo cereals	Quinoa <i>Chenopodium quinoa</i>	White quinoa	66.84 ±0.27 ^{de}	13.63 ±0.37 ^a	7.24 ±0.66 ^{ef}	2.02 ±0.003 ^c	10.25 ±0.004 ^b	0.95 ±0.03 ^{efg}	0.19 ±0.06 ^a	0.64 ±0.04 ^{ab}	78.19 ±0.44 ^{def}
		Red quinoa	65.22 ±1.78 ^{cd}	14.54 ±0.89 ^a	8.019 ±0.99 ^f	2.02 ±0.02 ^c	10.14 ±0.07 ^b	0.83 ±0.05 ^{defg}	0.31 ±0.02 ^a	0.79 ±0.03 ^{ab}	84.37 ±1.18 ^f
	Chia <i>Salvia hispanica</i>	NS	32.86 ±3.36 ^a	22.30 ±2.85 ^b	35.98 ±0.46 ^g	4.14 ±0.01 ⁱ	6.00 ±0.41 ^a	2.11 ±0.08 ^h	1.83 ±0.16 ^b	0.58 ±0.04 ^{ab}	88.63 ±1.69 ^f
Legumes	Lentil <i>Lens culinaris</i>	Kef	57.32 ±0.31 ^b	26.35 ±0.37 ^{bcd}	1.6 ±0.08 ^{ab}	2.43 ±0.02 ^{de}	12.13 ±0.46 ^{fg}	0.29 ±0.008 ^{ab}	0.30 ±0.01 ^a	3.77 ±0.12 ^{ef}	79.90 ±1.97 ^f
		Krib	58.80 ±0.49 ^b	26.76 ±0.48 ^{bcd}	1.01 ±0.09 ^a	2.56 ±0.05 ^{def}	10.93 ±0.004 ^{bcd}	0.24 ±0.002 ^{ab}	0.13 ±0.01 ^a	2.55 ±0.03 ^{cdef}	87.62 ±0.35 ^f
	Fava bean <i>Vicia faba</i>	Badii	56.93 ±0.46 ^b	27.46 ±0.15 ^{de}	1.70 ±0.26 ^{abc}	2.40 ±0.10 ^{de}	11.49 ±0.02 ^{def}	1.12 ±0.26 ^g	0.16 ±0.03 ^a	8.56 ±0.54 ^g	86.76 ±0.61 ^f
		Bachar	55.72 ±0.51 ^b	28.71 ±0.16 ^e	1.88 ±0.01 ^{abc}	2.37 ±0.005 ^d	11.85 ±0.03 ^{efg}	0.43 ±0.02 ^{abc}	0.18 ±0.03 ^a	1.34 ±0.03 ^{abc}	87.78 ±0.61 ^f
		Najeh	58.75 ±0.50 ^b	24.91 ±0.02 ^{bcd}	2.55 ±0.49 ^{abcd}	2.47 ±0.02 ^{def}	11.30 ±0.001 ^{cde}	0.57 ±0.02 ^{abcde}	0.14 ±0.01 ^a	1.63 ±0.10 ^{abc}	84.31 ±6.51 ^f
	Chickpea <i>Cicer arietinum</i>	Neyer	56.91 ±1.16 ^b	26.56 ±0.07 ^{bcd}	4.80 ±0.10 ^{cde}	2.81 ±0.0005 ^{gh}	10.22 ±0.03 ^b	0.62 ±0.04 ^{bcd}	0.31 ±0.02 ^a	1.95 ±0.53 ^{bcd}	78.4 ±0.64 ^{ef}
		Chetoui	60.16 ±0.60 ^{bc}	22.43 ±0.63 ^{bc}	4.59 ±0.01 ^{bcd}	2.61 ±0.01 ^{efg}	10.20 ±0.01 ^b	0.73 ±0.004 ^{cdef}	0.20 ±0.007 ^a	1.82 ±0.11 ^{abc}	83.76 ±1.67 ^f
		Beja 1	59.22 ±0.57 ^{bc}	22.67 ±0.64 ^b	5.06 ±0.05 ^{def}	2.64 ±0.001 ^{fgh}	10.40 ±0.01 ^{bc}	0.65 ±0.01 ^{bcd}	0.23 ±0.04 ^a	4.08 ±0.12 ^f	64.57 ±0.50 ^{cde}
		Rebha	56.43 ±1.30 ^b	25.99 ±0.14 ^{bcd}	4.31 ±1.43 ^{bcd}	2.52 ±0.035 ^{def}	10.73 ±0.009 ^{bcd}	0.50 ±0.05 ^{abcd}	0.22 ±0.03 ^a	2.15 ±0.32 ^{bcd}	79.21 ±0.48 ^{ef}
		Nour	61.04 ±0.31 ^{bcd}	22.74 ±0.05 ^{bc}	4.54 ±0.33 ^{bcd}	2.84 ±0.032 ^h	10.82±0.003 ^{bcd}	0.76 ±0.12 ^{cdefg}	0.16 ±0.003 ^a	3.64 ±0.47 ^{def}	63.15 ±3.80 ^{cd}
		Joud	55.81 ±0.32 ^b	23.10 ±0.57 ^{bc}	7.98 ±0.88 ^f	2.47 ±0.002 ^{def}	10.61 ±0.007 ^{bcd}	0.80 ±0.08 ^{cdefg}	0.20 ±0.02 ^a	3.14 ±0.27 ^{cdef}	57.12 ±1.65 ^{bc}

TPP: Total Polyphenols, GAE: Gallic Acid Equivalent, TF: Total flavonoids, CT: Condensed Tannins, CE: Catechin Equivalent. Mean ± S.D. with different superscripts in a column differ significantly (p < 0.05).

Tannins are bioactive phenolic compounds and may have beneficial or adverse nutritional effects, depending upon its concentration. However, tannins reduce the digestibility of nutrients when consumed through foods, but they can be significantly reduced by various domestic processing treatments such as soaking, germination, cooking etc. [57]. The tannins are categorized into two groups, namely, condensed tannins (non-hydrolysable) and hydrolysable tannins. Condensed tannins are the most commonly existing in stems, legumes, trees, forages, etc. [58]. To explore their potential role as antioxidants in food products, contents of condensed tannins were analyzed (Table 2). Results showed significant differences between studied samples whereby levels of CT oscillated from 8.56 mg CE/g to 0.28 mg CE/g. Fava bean flour (Badii variety) showed the highest concentration, whereas maize flour, the lowest content. Concentrations in CT were higher in

legume flours than in cereal and pseudo cereal flours. Moreover, similar to TPP, Fava bean cultivars as well as chickpeas cultivars displayed significant differences in CT concentrations. These results are in agreement with those found by Xu, Yuan [56] for condensed tannin content in different chickpea and lentil cultivars. Similar results were also observed for six fava bean varieties from Germany, the content of condensed tannins varied between 0.1 and 1.15 g/100g [59]. Changes in CT contents may be attributed to agricultural conditions, climate, or soil [60]. The antioxidant capacity of extracts was widely investigated using different methodological approaches such as the determination of the scavenging effect on 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals. The DPPH scavenging assay is extensively used to evaluate the free-radical scavenging of plant extracts because of its simple, rapid, sensitive, and reproducible procedure [61]. The

extracts of different flours displayed high antioxidant activity, and the DPPH scavenging activity values oscillated significantly from 88.63% to 34.65% (Table 2). The highest values were observed for chia with no significant differences in comparison with lentil, fava bean varieties and Chetoui variety for chickpea, while the lowest was found for rice flour (34.65%). Our results are contrasting with those reported by Millar, Gallagher [43] where the highest antioxidant activity was found in Fava bean flour (250.81 mg AAE/100g DPPH) amongst other pulse flours. Furthermore, Šibul, Orčić [57] observed the highest antioxidant activity in fava bean extracts compared with 6 other pulse vegetable extracts. This could be explained not only by plant growing location, year, and post-harvest storage, but also by the use of different analytical methods. Interestingly, the results showed that legume and pseudo cereal flours presented high antioxidant activity, in accordance with results of Marathe, Rajalakshmi [62], Zhao, Du [63] and Morales, Miguel [64]. Significant differences were noted amongst chickpeas cultivars, Kwon, Kim [65] reported similar results when comparing the phenolic compounds and the antioxidant activity of different fava bean genotypes from for instance, Turkey, Italy, Argentina, USA, and Hungary.

Based on their chemical analysis, our results highlighted the potential of incorporating such flours into cereal-based foods to increase the level of such beneficial plant chemicals in the diet. In addition to their protein content, pulse flours had the potential to play a preventative role in inflammatory conditions and metabolic syndrome, as highlighted in several reviews [66,67,68]. Interestingly Thongram, Tanwar [69] have showed a significant effect of addition of legume flour blends on the physicochemical and organoleptic properties of cookies. They specifically pointed out an increase in free radical scavenging activity due to the incorporation of legume flour into wheat flour. Similarly, Millar, Barry-Ryan [48] used wheat-pulse flour composites in the formulation of unleavened crackers. Besides the impact of the product quality, the substitution of wheat flour by fava bean and other pulse flours led to an increase of up to 162% in TPP, and of 182% in antioxidant activity (DPPH). Moreover, Drakula, Novotni [70] showed an increase of TPP (44%) and antioxidant capacity (30% DPPH) when replacing (25%) of whole meal rice flour with yellow pea flour in the formulation of GF sourdough and bread. Interestingly, antioxidant properties of pseudo cereals and legumes added to a food product may be beneficial not only for the consumers but also for enhancing shelf life of food product by preserving dietary lipids, biologically active substances, and color during storage [71].

3.1.3. Mineral Composition

In general, calcium, magnesium, and iron are scarce in the GF diet [73]. Studies underlined the importance of calcium intake and its contribution to bone metabolism in people with celiac disease. In recent years, there is an increase in non-communicable diseases, such as osteoporosis and anemia, caused by low and disorderly mineral intake. For this reason, fortification of some food products with minerals is an important nutritional approach [74]. Mineral composition of the different flour samples is showed in the Table 3. Amongst flour samples,

all three legume flours varieties had significantly higher content of zinc, calcium (except of chia), iron, and potassium compared to cereal flours ($p < 0.05$; Table 3). Magnesium assists in preserving normal muscle and nerve functions and maintains the heart rhythm steady. The Mg content differed significantly among the different flour samples and fluctuated between 0.05 % and 0.18 %. Pseudo cereal flours had the most important Mg value followed by legume flours, while cereal flours had the lowest Mg amount. Mg content of chickpea varieties differed significantly and presented the highest value in comparison with the other legume flours followed by lentil and fava bean which fluctuated between 0.089 and 0.132 g/100g DM. This result disagreed with those found by Levent and Yeşil [75] when studying the characteristics of different legume flours from Turkey to prepare noodles; they reported highest Mg content for fava bean flour followed by lentil then chickpea (0.076 g/100g to 0.121 g/100 g).

The iron composition of the different flours varied significantly from 24 ppm to 152.54 ppm. Legume flours presented higher iron amounts in comparison with cereal and pseudo cereal especially lentil flour. The importance of iron in the diet has been widely reported and presented multitude benefits on health including its contribution to normal cognitive function and metabolism, and reducing tiredness and fatigue [72]. Likewise, Ray, Bett [73] reported higher levels of iron and zinc in lentils and the most abundant amount of magnesium in chickpeas when comparing mineral composition of different pulse flours. Our data have also showed a variation in levels of zinc and Potassium amongst flour samples (Table 3). Rice flour had the lowest zinc and potassium contents while zinc content was the highest in chickpea (Beja 1 variety) and chia flours. Legume flours showed the highest zinc and potassium contents in comparison with cereal and pseudo cereal flours (except of chia flour).

As shown in Table 3, chickpea flour had the most important amount of potassium, followed by fava bean then lentil flours. In this concern, De Angelis, Pasqualone [74] observed similar results showing that potassium was the most abundant element in pulse flours. On the other hand, Millar, Gallagher [43] reported higher content of zinc, magnesium, iron and potassium of three pulse flours (fava bean, green pea, yellow pea) compared to wheat flour ($p < 0.05$), the highest content of potassium being detected in fava bean flour. Zinc has also been established to prevent the formation and reactive response of free radicals, which are unstable atoms that can damage cells, as well as to contribute to normal cognitive functions and to carbohydrate and fatty acid metabolisms [72]. Considering the recommended mineral intake laid out by the European Food Authority [75], our data corroborate that the use of legume flours in food product development could make a substantial contribution to meeting these requirements through the diet. In this study, the antioxidant activity determined with the DPPH assay proved a significant positive correlation with Zinc ($r = 0.55$, $p < 0.01$), Potassium ($r = 0.557$, $p < 0.01$), Magnesium ($r = 0.398$, $p < 0.05$), and non-significant correlation with Calcium ($r = 0.330$, $p > 0.05$). Similarly, total phenols were highly correlated with minerals content, Ca ($r = 0.702$, $p < 0.01$), Mg ($r = 0.781$, $p < 0.01$) and Zn ($r = 0.391$, $p < 0.05$), confirming observations of Grela, Samolińska [76] in different

leguminous seeds. These significant correlations might be due to the role of minerals in activating enzymes that enhance biosynthesis of flavonoids and phenolic compounds [77].

Minerals can also contribute to antioxidant activity by acting as cofactors for antioxidant enzymes.

Table 3. Mineral composition of cereals (rice and maize), pseudo cereals (white and red quinoa, chia) and legumes flours (Lentil, Fava bean, chickpea)

Flours		Variety	Minerals				
			Ca (%)	Mg (%)	Fe (ppm)	Zn (ppm)	K (%)
Cereals	Rice <i>Oryza sativa</i> L.	NS	0.01±0.0005 ^{ab}	0.05±0.003 ^a	26,98±0.45 ^a	18±0.65 ^a	0.15±0.001 ^a
	Maize <i>Zea mays</i>	NS	0.0065±0.0005 ^a	0.080±0.001 ^b	24±0.3 ^a	25±0.02 ^a	0.23±0.002 ^b
Pseudo cereals	Quinoa	White Quinoa	0.03±0 ^{abc}	0.180±0.001 ^e	58.7±0.94 ^{bcd}	35.14±0.17 ^{bc}	0.59±0.002 ^c
	<i>Chenopodium quinoa</i>	Red Quinoa	0.05±0 ^{abcd}	0.170±0.001 ^e	63.89±0.18 ^{cde}	33.75±0.06 ^b	0.64±0.008 ^c
	Chia <i>Salvia hispanica</i> L.	NS	0.68±0.03 ^f	0.34±0.02 ^f	63.69±3.21 ^{cde}	50.74±3.11 ^f	0.77±0.003 ^d
Legumes	Lentil <i>Lens culinaris</i>	Kef	0.15±0.0005 ^e	0.126±0.0005 ^{cd}	152.45±2.85 ^j	39.18±0.25 ^{bc}	0.87±0.004 ^{ef}
		Krib	0.09±0.001 ^d	0.110±0.002 ^{bcd}	97.44±0.97 ^h	36.96±0.09 ^{bc}	0.83±0.01 ^{de}
	Fava bean <i>Vicia faba</i>	Badii	0.06±0.001 ^{bcd}	0.090±0.003 ^{bc}	66.44±3.08 ^{def}	47.94±2.79 ^{ef}	0.94±0.006 ^{fg}
		Bachar	0.07±0.002 ^{cd}	0.099±0.002 ^{bcd}	53.97±2.45 ^{bc}	48.22±1.98 ^{ef}	1.10±0.002 ^{ij}
		Najeh	0.07±0.0005 ^{cd}	0.089±0.001 ^b	52.05±1.11 ^b	38.17±0.42 ^{bc}	0.97±0.01 ^{gh}
	Chickpea <i>Cicer arietinum</i>	Neyer	0.06±0.0005 ^{bcd}	0.118±0.003 ^{bcd}	70.45±0.27 ^{ef}	40.47±1.49 ^{bcd}	1.139±0.01 ^j
		Chetoui	0.079±0.002 ^{cd}	0.114±0.0005 ^{bcd}	70.19±0.95 ^{ef}	40.28±0.79 ^{bc}	1.05±0.005 ^{hi}
		Beja 1	0.091±0.002 ^d	0.131±0.001 ^{de}	119.75±0.75 ^j	51.28±0.44 ^f	1.072±0.039 ^{ij}
		Rebha	0.093±0.001 ^d	0.119±0.001 ^{bcd}	118.05±2.05 ⁱ	37.11±0.86 ^{bc}	0.978±0.007 ^h
		Nour	0.078±0.0005 ^{cd}	0.124±0.002 ^{cd}	73.97±0.99 ^{fg}	41.33±0.25 ^{cde}	1.042±0.017 ^{hi}
		Joud	0.088±0.0015 ^d	0.128±0.001 ^{cd}	80.575±2.375 ^g	47.63±0.68 ^{def}	1.075±0.014 ^{ij}

Mean ± S.D. with different superscripts in a column differ significantly (p<0.05).

3.2. Physical Analysis

Results of analysis of physical properties of flours were presented in Table 3. Bulk density (BD) gives an indication of flour heaviness. The bulk density for different flours varied from 0.61 to 0.87 g/ml (Table 4), the highest value was observed for lentil flour (Krib) and the lowest one for chickpea flour (Rebha variety). Lentil flour had the highest bulk density followed by fava bean flour; instead, chickpea flour had the lowest BD. Similarly, De Angelis et al. [74] have observed the highest BD for red lentil, indicating a denser flour, whereas a significantly lower BD was found in chickpea. Bulk density and fat content of the flours were slightly negatively correlated ($r = -0.055$, $p > 0.05$). This result confirmed those of Culetu, Susman [78]. However, Joshi, Liu [79] stated that flours tended to have higher bulk density as fats may act as adhesives in the aggregation of the flour particles, resulting to a density increase. Di Cairano, Condelli [80] reported that the distinct results in the bulk density of the different studies of GF flours characterization are related to non-homogeneous granulometry distribution of flours.

WAI represents the volume occupied by the starch after swelling in excess water, which maintains the integrity of starch in aqueous dispersion. WAI of different flours ranged between 4.03 g/g for chickpea (Neyer variety) and 9.25 g/g for chia flour (Table 4). Legume flours showed lower WAI than cereal and pseudo cereal flours. Those results are related to the hydrophilicity and gelation capacity of bio macromolecules, such as starch and protein, in flour [81]. De Angelis et al. [82] reported slightly lower WAI for lentil and chickpea flours. In contrast, Du, Jiang [83] have determined the highest WAI

value in chickpea flour, when compared to other tested legume flours (4.09 g/g to 6.13 g/g). Water solubility index (WSI) is highly related to WAI and quantifies the percentage of soluble solids that persist in the aqueous phase after the heating process. The WSI is an indicator of the solubility of molecules and differed significantly among different flours. WSI of flours varied from 3.20 to 24.62 g/100 g (Table 4). Legume flours showed distinctly higher WSI than cereal and pseudo cereal flours, where fava bean (Bachar variety) had the highest value. This is partially comparable to those previously reported by Di Cairano, Condelli [53], they showed a higher WSI for legume and pseudo cereal flours than cereal flours, with higher values of WSI for pseudo cereals. This could be explained by the establishment of amylose-lipid and protein-starch complexes in the process of heating which may have an impact on the WSI [40].

Color CIE values (L^* , a^* , b^*) of different flours are presented in the Table 4. Significant variation in L^* value, was observed amongst tested flours. Chickpea, fava bean, rice and white quinoa flours presented high L^* value: chia flour was darker with the lowest L^* , whereas chickpea flour (Neyer variety) was lighter with the highest L^* value. Mostly, final product color is an important parameter for its quality assessment, and consumers' acceptance. Therefore, Bouasla, Wójtowicz [84] showed that the lightness of dry pasta samples decreased as the amount of lentil flour in the recipe increased. The a^* and b^* values of different flours varied between -2.88 and 3.18 and 3.86 to 39.39 respectively. Red quinoa, chia and Krib lentil flours displayed positive a^* value, implying a pinkish color, whereas negative a^* values were found in other flours indicating a slight green tint. Maize flour had the highest b^* value, meaning a brighter yellow, followed by chickpea

flours, whereas rice flour displayed the lowest b^* value. The b^* value, an indicator of (-) blue and yellow (+); it has been attributed that variation in b^* among the samples may be attributed to the amount of carbohydrate and protein content [85]. This significant difference has a direct impact on the surface color of a cereal product which is an important parameter for initial consumer acceptability. In many studies, an increase in red and yellow tints were observed when flours, characterized with high a^* and b^* parameters, were added; especially, in the research mentioned above on rice pasta enriched with legume

flours, Bouasla, Wójtowicz [84] showed that the significant increase in red tint (a^*) for dry pasta enriched with 30 g/100 g of lentil flour was attributed to the red color of the lentil variety used in the study. Moreover, pasta enriched with 30 g/100 of yellow pea exhibited the highest yellow color (b^*), reflecting the color of the pea used in this study. The difference in the color characteristics of different flours may be attributed to differences in colored pigments of the flours, which in turn depends on the botanical origin of the plant and also to the composition of the flour [86].

Table 4. Physical properties of cereals (rice and maize), pseudo cereals (white and red quinoa, chia) and legumes flours (Lentils, Fava bean, Chickpea) flours

Flours		Variety	Bulk Density (g/ml)	WAI (g/g DM)	WSI (%)	L*	a*	b*
Cereals	Rice <i>Oryza sativa</i> L.	NS	0,75±0,009 ^{def}	9,004±0,46 ^f	3,20±0,003 ^a	93,57±0,17 ^d	-0,175±0,005 ^e	3,86±0,002 ^a
	Maize <i>Zea mays</i>	NS	0,80±0,002 ^{efg}	7,45±0,06 ^{de}	5,02±0,59 ^a	85±0,42 ^c	-2,45±0,11 ^a	39,39±0,005 ^h
Pseudo cereals	Quinoa <i>Chenopodium quinoa</i>	White quinoa	0,81±0,007 ^{efg}	8,09±0,13 ^{ef}	4,92±0,15 ^a	91,80±0,77 ^d	-0,53±0,01 ^{de}	8,9±0,93 ^{bc}
		Red quinoa	0,82±0,007 ^{fg}	7,42±0,71 ^{de}	5,24±0,04 ^a	68,47±0,34 ^b	3,18±0,16 ^g	12,9±0,4 ^e
	Chia <i>Salvia hispanica</i> L.	NS	0,72±0,001 ^{bcd}	9,25±0,31 ^f	2,89±0,22 ^a	31,19±1,3 ^a	1,85±0,06 ^f	17,21±0,02 ^{fg}
Legumes	Lentil <i>Lens culinaris</i>	Kef	0,84±0,01 ^g	5,35±0,05 ^{ab}	15,58±0,20 ^c	85,15±0,35 ^c	-1,21±0,11 ^c	11,72±0,23 ^{de}
		Krib	0,87±0,012 ^g	6,02±0,03 ^{bcd}	18,75±0,34 ^d	84,81±0,60 ^c	2,95±0,21 ^g	13,07±0,37 ^e
	Fava bean <i>Vicia faba</i>	Badii	0,67±0,007 ^{abc}	6,18±0,17 ^{bcd}	15,17±0,84 ^c	92,90±0,41 ^d	-1,88±0,04 ^b	11,17±0,26 ^{de}
		Bachar	0,74±0,01 ^{cde}	6,04±0,22 ^{bcd}	24,62±0,89 ^e	92,93±0,17 ^d	-1±0,05 ^{cd}	7,4±0,19 ^b
		Najeh	0,66±0,0098 ^{ab}	6,86±0,05 ^{cde}	17,16±0,45 ^{cd}	92,06±0,37 ^d	-1,40±0,09 ^{bc}	10,69±0,33 ^{cd}
	Chickpea <i>Cicer arietinum</i>	Neyer	0,67±0,02 ^{abc}	4,03±0,01 ^a	17,43±0,72 ^{cd}	94,33±0,16 ^d	-2,67±0,06 ^a	17,49±0,38 ^{fg}
		Chetoui	0,67±0,0001 ^{abc}	6,40±0,05 ^{bcd}	12,03±0,08 ^b	94,23±0,07 ^d	-2,65±0,04 ^a	17,42±0,04 ^{fg}
		Beja 1	0,66±0,002 ^{ab}	5,83±0,21 ^{bc}	16,86±0,17 ^{cd}	93,81±0,25 ^d	-2,57±0,02 ^a	18,49±0,4 ^{fg}
		Rebha	0,61±0,029 ^a	5,85±0,18 ^{bc}	14,93±0,16 ^c	92,97±0,13 ^d	-2,43±0,005 ^a	16,5±0,36 ^f
		Nour	0,660±0,0034 ^{ab}	5,08±0,10 ^{ab}	16,87±0,85 ^{cd}	93,93±0,26 ^d	-2,88±0,01 ^a	18,91±0,58 ^g
		Joud	0,660±0,015 ^{ab}	5,34±0,08 ^{ab}	16,26±0,03 ^{cd}	93,38±0,29 ^d	-2,76±0,005 ^a	19,12±0,19 ^g

BD: Bulk Density, WAI: Water Absorption Index, WSI: Water Solubility Index. Mean ± S.D. with different superscripts in a column differ significantly ($p < 0.05$).

3.3. Functional Properties

Functional properties qualified as the nonnutritive characterization that food components play in a food system. Functionality of flour is important in the preparation, processing, storage, quality, and organoleptic attributes of foods. Knowledge of functional properties is essential to developing new products and improving those that already exist. Table 5 summarizes functional properties of the different flours. Functional properties are strongly affected by the composition of the flour and reflect the interactions between flour components. Water Absorption Capacity (WAC) indicates the amount of water that can be bound by a gram of flour. Variations in WAC may be related to proteins structure as well as to the presence of hydrophilic carbohydrates [87]; high WAC may help to retain a softer texture in bakery products [88]. Examining Table 5, WAC was ranged between 0.94 g/g DM to 7.35 g/g DM. Chia flour had the highest value and rice has the lowest WAC. Pulse flours had significant different values of WAC, where lentils exhibited the highest followed by fava bean while chickpea showed the lowest values. In this line, the results obtained are similar to those obtained by Du, Jiang [88] who found WAC values of legume flours ranging from 1.12 g/g to 1.89 g/g. In the present study, no significant correlation was found

between water absorption and protein content ($r = 0.027$, $p > 0.05$); and high negative correlation with carbohydrates content ($r = -0.752$, $p < 0.01$) was observed. Those findings were in accordance with the results found by Culetu, Susman [78] who stated no good correlation between water absorption and protein content ($r = 0.2099$) or starch content ($r = -0.4392$). However, Patil and Arya [89] stated that higher protein content tends to increased water absorption. The type of proteins and the presence of non-polar side chains have also an effect on Oil Absorption Capacity (OAC). It indicates the weight of oil retained by a gram of flour. OAC varied between 0.79 g/g DM to 1.38 g/g DM (Table 5). Chia flour had the lowest OAC, and fava bean flour (Badii variety) had the highest value; legume flours presented higher OAC than cereal flours. The highest OAC of legumes flour is potentially useful in structural interactions in food, especially in flavor retention, improvement of palatability and extension of shelf life particularly in bakery products where fat absorption is desired. Those findings agreed with those showed by Culetu, Susman [78], these authors obtained higher OAC values for chickpea than maize and rice flours. However, Di Cairano, Condelli [80] found no significant differences between OAC of different grains of cereals, pseudo cereals and legumes. The significant difference between the flours could be explained by the

different type of proteins and amino acid composition and protein polarity and hydrophobicity [81].

The least gelation concentration (LGC) was used as an index of gelation capacity. The data for LGC of different flours are given in Table 5. LGC fluctuated between 8 and 18 %. Rice, chia and Badii fava bean variety had the lowest value. Red quinoa flour had the highest LGC followed by chickpea (Nour variety) and lentil flours. Kaur and Sandhu [36] found similar results when studied the LGC for various lentil flours which ranged between 12 and 14%. Previous studies have indicated LGC of 10-14% in chickpea flours [29]. Legume flours contain high protein and starch contents, and the gelation capacity of flours is influenced by a physical competition for water between protein gelation and starch gelatinization. The flours having the low gelation concentration may be an asset for the formulation of curd or as an additive to other gel forming materials in food products.

Proteins being the surface-active agents can form and stabilize the emulsion by creating electrostatic repulsion on oil droplet surface. Flours differed significantly in their abilities to emulsify corn oil. EA of different flours ranged

between 36.62 and 74.17 ml/100 ml. Maize flour had the lowest EA and chickpea's Chetoui variety had the highest EA. Du, Jiang [83] reported close results for chickpea flour of 61.14 %, and when comparing functional properties of different pulses, they concluded higher EA for chickpea compared to lentil. Contrary, in our study, chickpea had higher EA relative to lentil and fava bean. The protein content of chickpea flour is lower than the other kinds of legume flour and indicating relatively higher emulsion activity; this result confirms that EA is strongly related to the Protein-water interactions that occur in the polar amino acid regions of protein molecules.

In contrast, ES varied from 51.58 to 88.33 ml/100 ml. The highest ES was registered for Chetoui's chickpea flour, and the lowest value was registered for maize flour. The difference in protein composition as well as possibly carbohydrates, may contribute substantially to the emulsification properties of protein-containing products like legume flours. High emulsion properties of legume flours especially chickpea indicated that it could be useful as an additive for the stability of fat emulsion in the production of sausage, soup and cake [42].

Table 5. Functional characterization of cereals (rice and maize), pseudo cereals (white and red quinoa, chia) and legumes (Lentils, Fava bean, Chickpea) flours

Flours		Variety	WAC (g/g DM)	OAC (g/g DM)	MGC (%)	EC ml/100ml	ES ml/100ml	FC ml/100ml	FS (20 min) ml/100ml	FS (40 min) ml/100ml	FS (60 min) ml/100ml	FS (120 min) ml/100ml
Cereals	Rice <i>Oryza sativa L.</i>	NS	0.94 ±0.11 ^a	0.96 ±0.06 ^{ab}	8	70.67 ±0.09 ^{cde}	83.33 ±0 ^c	21.12 ±1.51 ^b	19.20 ±1.55 ^{bc}	19.20 ±1.55 ^b	17.27 ±1.59 ^{bcd}	7.69 ±0.14 ^{ab}
	Maize <i>Zea mays</i>	NS	1.45 ±0.15 ^{defg}	0.79 ±0.005 ^a	10	36.62 ±0.51 ^a	51.58 ±5.61 ^a	7 ±1 ^a	3 ±1 ^a	1 ±1 ^a	0 ±0 ^a	0 ±0 ^a
Pseudo cereals	Quinoa <i>Chenopodium quinoa</i>	White quinoa	1.73 ±0.007 ^{ghi}	1.15 ±0.004 ^{bcd}	12	69.16 ±5.83 ^{cde}	88.31 ±2.59 ^c	31.72 ±0.35 ^{bcd}	26.89 ±1.40 ^{bcd}	22.14 ±1.38 ^b	19.23 ±0.36 ^{bcd}	8.71 ±3.05 ^{abc}
		Red quinoa	1.61 ±0.01 ^{igh}	1.18 ±0.01 ^{bcd}	18	70.29 ±2.43 ^{bcd}	84.61 ±0 ^c	29.09 ±3.60 ^{bc}	29.09 ±3.60 ^{bcd}	26.20 ±0.71 ^{bcd}	24.26 ±0.73 ^{bcd}	16.51 ±1.13 ^{cde}
	Chia <i>Salvia hispanical.</i>	NS	7.35 ±0.11 ^j	0.75 ±0.03 ^a	8	60.71 ±3.57 ^{bcd}	77.27 ±4.54 ^{bc}	4 ±0 ^a	2 ±0 ^a	2 ±0 ^a	2 ±0 ^a	1 ±0 ^a
Legumes	Lentil <i>Lens culinaris</i>	Kef	2.007 ±0.007 ⁱ	1.14 ±0.01 ^{bc}	12	55.73 ±0.73 ^b	70.83 ±4.16 ^{abc}	43.79 ±1.48 ^d	43.79 ±1.48 ^f	40 ±0.38 ^e	39.06 ±1.32 ^f	35.26 ±3.19 ^f
		Krib	1.82 ±0.004 ^{hi}	1.20 ±0.01 ^{cd}	12	57.98 ±0.84 ^{bc}	71.48 ±2.04 ^{bc}	45 ±3 ^d	41 ±3 ^{ef}	35 ±5 ^{cde}	27 ±3 ^{de}	20 ±0 ^e
	Fava bean <i>Vicia faba</i>	Badii	1.50 ±0.001 ^{efg}	1.38 ±0.06 ^d	8	65.78 ±0 ^{bcd}	81.25 ±6.25 ^{bc}	44.64 ±1.5 ^d	38.81 ±1.56 ^{def}	35.91 ±0.62 ^{de}	32.99 ±1.62 ^{ef}	29.11 ±1.65 ^f
		Bachar	1.30 ±0.01 ^{bcd}	1.30 ±0.09 ^{cd}	10	56.87 ±0.62 ^{bc}	73.39 ±1.60 ^{bc}	33.33 ±1.96 ^{bcd}	28.43 ±4.90 ^{bcd}	26.47 ±4.90 ^{bcd}	24.50 ±4.90 ^{cde}	15.68 ±1.96 ^{bcd}
		Najeh	1.36 ±0.03 ^{cdef}	1.32 ±0.02 ^{cd}	8	57.64 ±1.17 ^{bc}	75.96 ±0.96 ^{bc}	32.01 ±2.60 ^{bcd}	27.16 ±1.67 ^{bcd}	23.28 ±1.71 ^{bc}	22.30 ±2.69 ^{bcd}	18.42 ±2.73 ^{de}
	Chickpea <i>Cicer arietinum</i>	Neyer	1.03 ±0.003 ^{ab}	1.35 ±0.01 ^{cd}	14	63.26 ±6.15 ^{bcd}	78.46 ±1.53 ^{bc}	26.92 ±0 ^{bc}	21.15 ±1.92 ^{bc}	17.30 ±1.92 ^b	16.34 ±0.96 ^{bc}	14.42 ±0.96 ^{bcd}
		Chetoui	1.20 ±0.007 ^{abcd}	1.14 ±0.007 ^{bc}	10	74.17 ±2.74 ^e	88.33 ±5 ^c	25 ±5 ^{bc}	19 ±3 ^{bc}	17 ±1 ^b	16 ±2 ^{bc}	13 ±1 ^{bcd}
		Beja 1	1.12 ±0.004 ^{abc}	1.13 ±0.01 ^{bc}	12	55.71 ±1.42 ^b	63.94 ±2.72 ^{ab}	28 ±0 ^{bc}	24 ±0 ^{bc}	22 ±0 ^b	20 ±0 ^{bcd}	18 ±0 ^{de}
		Rebha	1.02 ±0.002 ^{ab}	1.23 ±0.07 ^{cd}	12	62.5 ±0 ^{bcd}	80.35 ±5.35 ^{bc}	27 ±1 ^{bc}	23 ±1 ^{bc}	21 ±1 ^b	19 ±1 ^{bcd}	10 ±0 ^{bcd}
		Nour	1.07 ±0.01 ^{abc}	1.31 ±0.02 ^{cd}	16	58.57 ±1.42 ^{bcd}	77.5 ±2.5 ^{bc}	21.82 ±4.17 ^b	17.84 ±2.15 ^b	17.84 ±2.15 ^b	16.84 ±1.15 ^{bcd}	11.88 ±0.11 ^{bcd}
		Joud	1.11 ±0.005 ^{abc}	1.25 ±0.007 ^{cd}	14	62.85 ±0 ^{bcd}	74.92 ±3.50 ^{bc}	27.74 ±2.25 ^{bc}	20.82 ±3.17 ^{bc}	17.84 ±2.15 ^b	13.86 ±0.13 ^b	9.90 ±0.09 ^{bcd}

WAC: Water Absorption Capacity, OAC: Oil Absorption capacity, MGC: Minimum Gelation Concentration, EA: Emulsion Capacity, ES: Emulsion Stability, FC: Foaming capacity, FS: Foam stability. Mean ± S.D. with different superscripts in a column differ significantly (p < 0.05).

Foaming capacity depends on the ability of proteins to adsorb quickly at the air-water interface during whipping. Foaming capacity and stability generally depend on the interfacial film formed by proteins, which maintains the air bubbles in suspension and slows down the rate of coalescence. The foaming capacities (FC) and foam

stabilities (FS) varied significantly among the flours. FC of flours varied from 4 to 45 ml/100 ml (Table 5). The highest FC (45 ml/100 ml) was observed for lentil flour whereas lowest FC (4 ml/100 ml) was observed for chia flour (Table 5). Legume flours exhibited the highest FC in comparison with cereal and pseudo cereal flours,

especially lentil and fava bean flours. In this line, Kaur and Sandhu [36] showed similar results, indicating that the foams produced by lentil flours were relatively thick with FC between 33.9 and 47.3%. The good foaming ability of lentil flours makes them useful in food systems that require aeration for textural and leavening properties. Foaming stability (FS) is the ability of flours to maintain the whip as long as possible. Foam stability of maize and chia flours were the lowest while lentils presented the most stable foam after 2 hours of storage, followed by fava bean and chickpea. Mortuza, Hannan [90] reported higher foaming capacity for fava bean from Bangladesh of 57.19 % when compared to studied fava bean varieties, and similar value of foam stability (29.78 %) to Badii variety after 2 hours of storage. FC and FS were highly correlated to the protein content of flours (FC: $r=0.503$; FS $r=0.450$ to 0.551 ; $p<0.01$) and slightly correlated to carbohydrates. In this concern, Chandra, Singh [91], and Joshi, Liu [79] stated that the foam capacity of a flour is dependent on the configuration of protein molecules and carbohydrates present in the flour

3.4. Principal Component Analysis

Principal Component Analysis (PCA) was used to better understand the variation among GF flours and to identify correlations between the investigated parameters (Figure 1). Previous studies also used PCA analysis to visualize the variation in the characteristics of various flours [45,78,80,89]. On the score plot, the distance between the positions of flours is proportional to the degree of difference or similarity between them. This analysis exhibited two axes explaining the essential variability. The first and the second PCs described 37.6% and 25.8% of the variance respectively. As shown in figure 1 a), legumes clustered together on the plot, which reflected similarity between those samples, and they were positively influenced by the two first principal components. Instead, pseudo cereal flours are totally different between each other, with chia flour located on the opposite quadrant compared to quinoa flour as represented in figure 1 b). Furthermore, the plot showed similarity between cereal flours as these samples are located closely on the PCA plot; those flours are largely negatively influenced by the second principal component (PC2). The loading plot of the two PCs afforded data about correlations between measured parameters (Figure 1 c). PC1 is well characterized by FC, FS (20, 40, 60 and 120 min), L^* , moisture, CT, WSI, carbohydrates, proteins, Fe, CT, LGC, EC and ES. Legumes differed from most of the other flours for their WSI, FC, FS, Fe, CT, LGC, EC, and ES. Chia differed from red and white quinoa for ash, Zn, DPPH, proteins, WAI, b^* and BD. Di Cairano, Condelli [80] found similar results investigating different properties of GF flours, authors stated that buckwheat flour was differentiated by the other pseudo-cereals, amaranth and quinoa, mainly for the phenolic compounds and OAC. On the other hand, Culetu, Susman [78] found opposite results where pseudo cereal flours were clustered together. Moreover, those two studies [78,80] showed similarities on one hand between cereal flours and on the other between legumes flours.

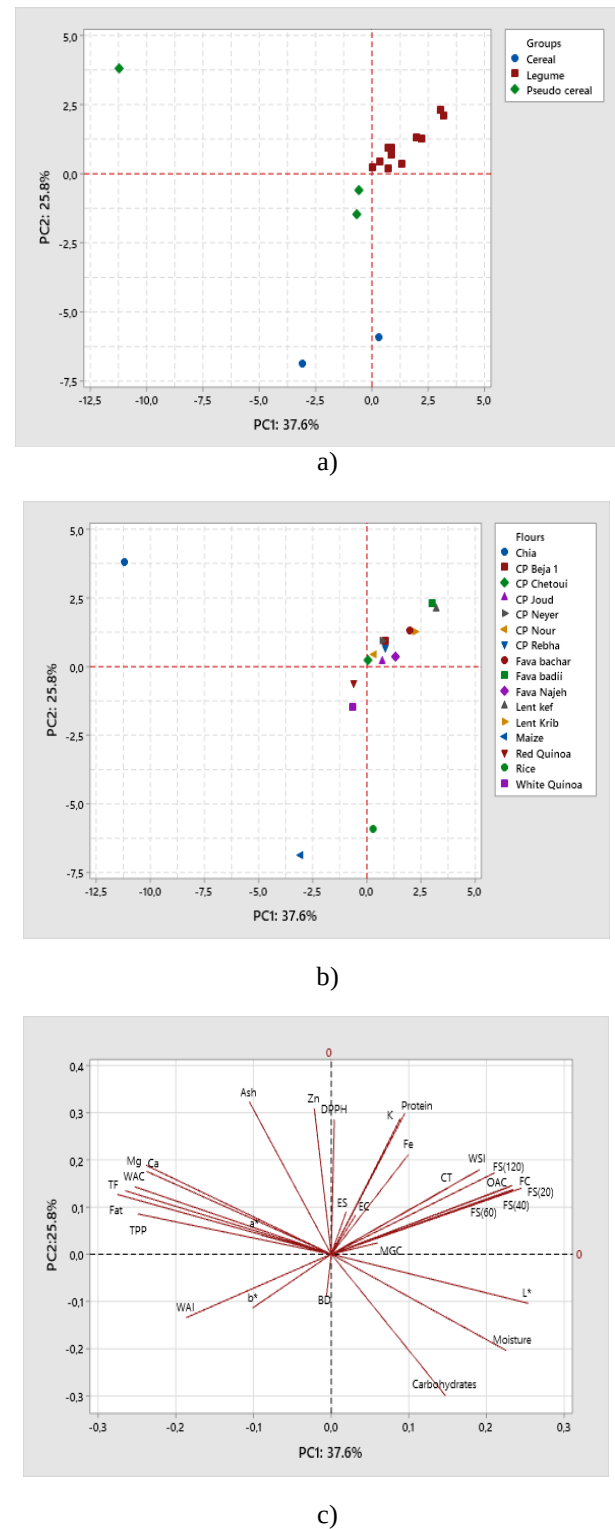


Figure 1. Principal Component Analysis of gluten free cereal, pseudocereal and legume flours. **a)** Score plot of first and second principal component (PC1 and PC2) describing the variation among flours (cereals (●), pseudocereals (◆) and legumes (■)). **b)** Score plot describing the overall variability among the different gluten-free flours (PC: Chickpea, Fava: Fava bean, Lentil: Lentil). **c)** Loading plot of PC1 and PC2 describing the relationship between parameters (BD: bulk density; WAI: Water Absorption Index; WSI: Water Solubility Index; WAC: Water Absorption Capacity; OAC: Oil Absorption Capacity; LGC: Least Gelation Concentration; EC: Emulsion Capacity; ES: Emulsion Stability; FC: Foaming Capacity; FS: Foam Stability (after 20, 40, 60 and 120 min), TPP: Total Poly Phenols; TF: Total Flavonoids; CT: Condensed Tannins)

4. Conclusion

The use of nutrient strengthener flours may contribute to the enhancement of GF bakery product quality. They are introduced into GF formulations in order to mimic the effect of gluten and give highly nutritious GF products, satisfying textural and sensory constraints. The aim of this work was to investigate nutritional, phenolic profile, physical and functional properties of 16 GF flours belonging to local legumes, cereals and pseudo cereals, significant differences could be observed across the tested flours ($p < 0.05$). Cereals showed low contents of protein, mineral (iron, zinc, calcium, potassium, magnesium) and antioxidant activity comparing to legume and pseudo cereal. On the contrary, legume flours exhibited higher protein and lower fat contents; besides, they showed interestingly higher iron, potassium, and zinc levels (except for chia showed no significant difference). Moreover, legume and pseudo cereal had no significant difference concerning the antioxidant activity. Additionally, chickpea had higher EA and ES while lentil followed by fava bean had higher FC and FS with thick foams, fava bean showed the highest OAC which make it potentially useful in flavor retention; lentil presented the highest bulk density which suggests its suitability for use in food preparations since it helps to reduce paste thickness which is an important factor in convalescent and child feeding. The results of the study revealed that legume flours have a great potential to be used in food industry. Going forward, pulses could be a sustainable solution to the challenge of supplying high quality dietary protein to the growing world population. Besides, dietary transitions with legume-based products compared to animal-based equivalents, showed environmental benefits such as being climate friendly and resource efficient. Therefore, it is important to do more research on formulations of cereal products using these flours especially they take into account economic constraints.

Declaration of Competing Interest

The authors declare they have no conflict of interests.

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