Bulgarian Journal of Agricultural Science, 24 (No 4) 2018, 719–726

Influence of fermentation and germination treatments on physicochemical and functional properties of acorn flour

Menasra Amina[1], Fahloul Djamel[1*], and Haddad Djamel[2]

[1]*Food Sciences Laboratory (LSA), Department of Food Technology, University of Hadj Lakhdar Batna, Batna 05000, Algeria*
[2]*Laboratory (LESEI), Faculty of Engineering, University of Batna, Batna 05000, Algeria*
**E-mail:* dfahloul2@yahoo.com

Abstract

Amina, M., Djamel , F. & Djamel , H. (2018). Influence of fermentation and germination treatments on physicochemical and functional properties of acorn flour. *Bulgarian Journal of Agricultural Science, 24*(4), 719-726.

Acorn nuts (*Quercus ilex* L.) were treated by fermentation and germination in order to determine the effect on flour properties. Results showed that fermentation and germination treatments enhance functional properties of acorn flour which are very important in food preparation. Also, these treatments improve nutritional value of flour, increasing the mineral content (2-2.06%), decrease starch (23.8-31.15%) and phenolic content "antinutritional group" (0.52-0.55%). This research suggests that fermentation and germination treatments may help to enhance acorn flour quality.

Keywords: acorn; fermentation; germination; flour; properties
Abbreviations: M*:* Moisture; DM*:* Dry Matter; A*:* Ash; OM*:* Organic Matter; S*:* Starch; TTA*:* Total Titratable Acidity; TPC*:* Total Phenolic Content; BD*:* Bulk Density; WHC*:* Water Holding Capacity; OHC*:* Oil Holding Capacity; EA*:* Emulsifiying Activity; FC*:* Foaming Capacity; SP*:* Swelling Power; NAF*:* Natural Acorn Flour; FAF*:* Fermented Acorn Flour; GAF*:* Germinated Acorn Flour

Introduction

Quercus ilex L., also called holly oak or evergreen oak, is a common Mediterranean, medium-size, evergreen tree which is widely distributed along the Balkan peninsulas and the Mediterranean region (Karioti et al., 2010). The acorn is an edible oval fruit of oak trees (Ghaderi-Ghahfarrokhi et al., 2017).

Acorns are considered as a good sources of fibers, vitamins (mostly A and E), mineral elements, unsaturated fatty acids (Vinha et al., 2016) and biologically active compounds such gallic acid, and ellagic acid (Bahmani et al., 2015).

Plant proteins have been reported to have limiting amino acids and it is necessary to combine these plant proteins in proportions that improve the protein intake of consumers. Many processes are available for improving the nutritional quality of plant foods. These methods include traditional methods such as cooking, soaking, dewatering, fermentation, germination, smoking, salting, curing, etc. (Okpala and Okoli, 2012).

Germination is a natural biological process (Sangronis and Machado, 2007). Germination has been reported to induce an increase in free limiting amino acids and available vitamins with modified functional properties of seed components (Okpala and Okoli, 2012).

Fermentation improves amino acid composition and vitamin content, increases protein and starch availability and lowers levels of antinutrients (Okpala and Okoli, 2012).

Despite their botanical availability, acorns are not currently widely used as common nuts. Also, the available information on nutrients and chemical composition of acorns is far from being exhaustive and more research should be carried out to achieve

a comprehensive characterization of this raw material, hence boosting its potential applications (Vinha et al., 2016). Furthermore, no work has been done on the effect of fermentation and germination treatments on acorn (*Quercus ilex* L.) flour quality. So, the present study was carried out to evaluate functional and physicochemical properties of natural and treated acorn flours by fermentation and germination processes.

Material and Methods

Acorns were collected during the month of October 2016 in Ichamoul region of Batna, Algeria. All reagents and chemicals used in the experimental work were of analytical grade and were purchased from Sigma Co. (St. Louis, MO, USA). All materials were stored at 4°C until testing (Correia et al., 2009).

Physical characteristics of acorn

Acorn samples were characterized physically by determining the number of nuts in 1 kg, the mass of 1000 nuts and the ratio of the shell and the nut (Rakic et al., 2006). Length and maximum diameter were measured for 100 randomly selected nuts using digital calipers with a sensitivity of 0.01 mm (Fos'hat et al., 2011; Galvan et al., 2011).

Fermentation and germination treatments

Treated acorns nuts by fermentation and germination processes were prepared according to Khattab and Arntfield (2009) and Sangronis and Machado (2007), respectively as described in Fig. 1.

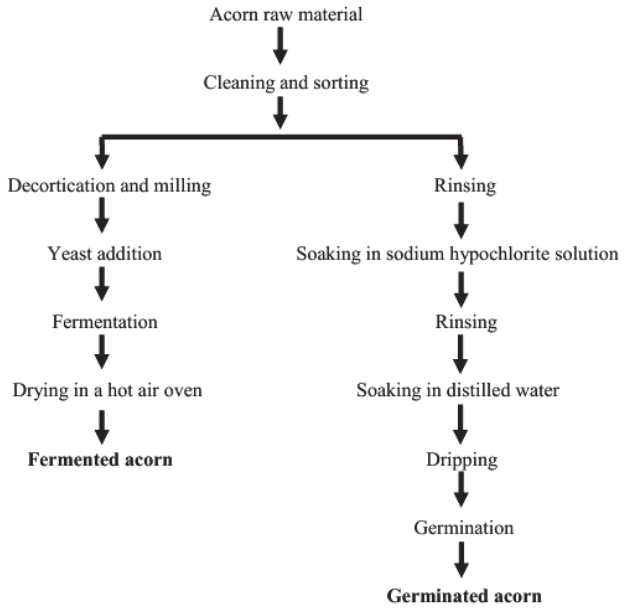

Fig. 1. Fermentation and germination treatments

Acorn flours production

Natural and treated acorns were dried at temperature of 50±5°C for 16-18 h, ground in laboratory mill (Sangronis and Machado, 2007) and sieved (one millimeter) to obtain a fine homogeneous flour. Samples were stored at 4°C until analysis (Galvan et al., 2011).

Physicochemical properties of acorn flours

Moisture content in acorn flours was determined according to AACC Method 44-19.01 (AACC, 2012). Flours were characterized for pH and ash according to AACC Methods 02-52 and 08-01, respectively (AACC, 2000). Total titratable acidity (TTA) was determined by a Sodium hydroxide (NaOH) titration according to AOAC (2005). Total starch content was determined by polarimetric method (Correia et al., 2009).

Total phenolic content (TPC) in acorn flours was determined by Folin-Ciocalteu assay (Ghaderi-Ghahfarrokhi et al., 2017) using the Shimadzu Ultraviolet-Visible (UV-Vis) spectrophotometer T60U (Tejerina et al., 2011). Total phenolic compounds were quantified using a gallic acid standard curve ranging 2.55 µg/mL (Tejerina et al., 2011). The results were calculated with regard to the dry matter (Rakic et al., 2006).

Color of flours was assessed using a colorimeter (CR-10, Konica Minolta Sensing Inc., Osaka, Japan). A white tile (L* = 97.46; a* = -0.02; b* = 1.72) was used as reference. Total color difference (TCD*) as defined by Equation (1) was also calculated (Correia et al., 2009):

$$TCD^{*} = \sqrt{(L^{*} - L_{0}^{*})^{2} + (a^{*} - a_{0}^{*})^{2} + (b^{*} - b_{0}^{*})^{2}} \quad (1)$$

Functional properties of acorn flours

Bulk density was determined as described by Chinma et al. (2009). Water and oil holding capacities were defined as determined by Zouari et al. (2016). Emulsifying activity and foaming capacity were determined as described by Elkhalifa et al. (2010). Swelling power was defined as described by Adebowale and Maliki (2011).

Statistical analysis

Statistical analysis was performed using SPSS software version 20.0 (SPSS Inc., Chicago, IL, USA). Significant differences between the detected parameters were compared by means of Duncan's multiple comparison test at the 95% confidence level ($p \leq 0.05$) (Aponte et al., 2014). The results for acorn flours properties were compared using principal component analysis (PCA). Comparison was performed by STATISTICA software, version 10.0 (StatSoft, France) (Aponte et al., 2014).

Results and Discussion

Physical properties of acorn

Physical properties of acorn are presented in Table 1. Length of acorn nut was found to be 39.99 mm. This value is similar to those (22.81-40.35 mm) reported by Galvan et al. (2011) for holm oak (*Quercus ilex* subsp. *ballota*). Maximum diameter of acorn nut was found to be 17.9 mm. This value is lower than (18.21 mm) mentioned by Fos'hat et al. (2011) for acorn (*Quercus suber* L.). Acorn nut mass was found to be 4.16 g. This value is lower than (7.22 g) reported by Rakic et al. (2006) for acorn (*Quercus robur*) and similar to those (2.41-6.12 g) found by Galvan et al. (2011) for holm oak. Cantos et al. (2003) mentioned that the weight of fifteen acorns (*Quercus ilex*) is 178.78 g. The number of acorn nuts in one kilogram was found to be 240.38. Rakic et al. (2006) reported the number of acorn (*Quercus robur*) in one kilogram equal to 138.91. This result is acceptable due to the difference between acorns masses. Shell of acorn was found to be 16.82%. This value is higher than (14.33%) found by Rakic et al. (2006) for acorn (*Quercus robur*). Nut ratio of acorn was found to be 83.17%. This value is lower than (85.66%) mentioned by Rakic et al. (2006) for acorn (*Quercus robur*).

According to Rakic et al. (2006), physical characteristics of the starting material are important for several reasons, foremost the nut collecting procedure, including the mechanical separation of the shell from the nut, drying and crushing or milling.

Table 1
Physical characteristics of acorn (*Quercus ilex* L.)

Parameters	Acorn nut
Length (mm)	39.99 ± 0.01^{a}
Maximum diameter (mm)	17.99 ± 0.4^{b}
Number of nuts in 1kg	240.38 ± 0.12^{c}
Mass of 1000 nuts (g)	4.16 ± 0.02^{d}
Shell (%)	16.82 ± 0.03^{e}
Nut (%)	83.17 ± 0.02^{f}

Superscript values with different letters indicate significant difference ($P \leq 0.05$) analyzed by Duncan's multiple range test.

Physicochemical properties of acorn flours

The effect of fermentation and germination treatments on physicochemical properties of acorn flours is presented in Table 2. Moisture content of acorn flours was found to lie in the acceptable limits. Values lie within the limits that enable safe storage. The level of moisture of natural acorn flour (10.37%) is higher than found by Rakic et al. (2006) for acorn *Quercus robur* flour (7.89%) and reported by Li et al. (2015) for *Quercus glandulifera* Bl flour (7.55%), respectively.

Moisture content increases by germination (10.48%) and fermentation (10.50%) treatments. Similarly, Chinma et al. (2009) and Gernah et al. (2011) found an increase in moisture of germinated tigernut (*Cyperus esculentus*) and fermented maize (*Zea mays*) flours, respectively. High moisture content in fermented acorn flour can be attributed to the addition of water to the acorn prior to fermentation (Ojokoh and Bello, 2014). The increase in moisture content in germinated acorn flour might be due to its low dry matter content (Chinma et al., 2009).

Ash content in natural acorn flour (1.85%) is similar to those (1.34-2.02%) reported by Galvan et al. (2011) for holm oak (*Quercus ilex* subsp. *ballota*). Found ash values by Rakic et al. (2006), Hegazy et al. (2014) and Li et al. (2015) for

Table 2
Physicochemical properties of acorn flours

Parameters	Natural acorn flour	Fermented acorn flour	Germinated acorn flour
Moisture (%)	10.37 ± 0.015^{aA}	10.50 ± 0.015^{aBC}	10.48 ± 0.015^{aCB}
Dry matter (%)	89.63 ± 0.015^{bA}	89.50 ± 0.015^{bBC}	89.52 ± 0.015^{bCB}
Ash (%)	1.85 ± 0.015^{cA}	2.06 ± 0.015^{cB}	2.00 ± 0.01^{cC}
Organic matter (%)	98.15 ± 0.015^{dA}	97.94 ± 0.015^{dB}	98 ± 0.01^{dC}
pH	6.05 ± 0.011^{eA}	3.93 ± 0.011^{eB}	5.83 ± 0.02^{eC}
TTA (%)	0.15 ± 0.015^{fAC}	0.23 ± 0.005^{fB}	0.17 ± 0.015^{fCA}
Starch (%)	34.2 ± 0.608^{gA}	31.15 ± 0.020^{gB}	23.8 ± 0.2^{jC}
TPC (%)	0.61 ± 0.005^{hjA}	0.55 ± 0.005^{hB}	0.52 ± 0.01^{hC}
L*	72 ± 0.1^{iAB}	67.03 ± 0.25^{iBA}	67.3 ± 0.3^{iC}
c*	27.36 ± 0.25^{jhAB}	26.46 ± 0.35^{jBA}	26.09 ± 0.27^{jC}
h°	78.1 ± 0.2^{kAC}	78.13 ± 0.25^{kB}	80.46 ± 0.18^{kCA}
TCD*	35.95 ± 0.19^{lA}	39.5 ± 0.085^{lB}	38.70 ± 0.03^{lC}

TTA: total titratable acidity, TPC: total phenolic content, L^*: lightness, c^*: chromaticity, h°: hue angle, TCD^*: total color difference. Superscript values with different letters (lower-case in same column or upper-case in the same line) indicate significant difference ($P \leq 0.05$) analyzed by Duncan's multiple range test.

acorns *Quercus robur*, *Quercus glandulifera* Bl and chestnut *Castanea sativa* Mill. are 2.07%, 0.03% and 2.44% , respectively.

An increase in ash content was observed with germination and fermentation treatments (Table 2) and there were significant differences ($P \leq 0.05$) in ash content values among flour samples. Idris et al. (2005) and Gernah et al. (2011) observed an increase in mineral content with increased time of germinated sorghum and fermented maize flours, respectively. The increase in ash level during fermentation could be as a result of incomplete utilization of minerals by fermenting organisms during metabolism (Ojokoh and Bello, 2014).

pH value decreased (3.93) and total titratable acidity (TTA) increased (0.23) under fermentation treatment influence. These results could be due to carbohydrates degradation resulting in acidification of fermented acorn flour (Bilgiçli et al., 2006; Ojokoh and Bello, 2014). These observations are in agreement with earlier studies by Ojokoh and Bello (2014) for fermented millet (*Pennisetum glaucum)* and soybean (*Glycine max)* blend flours. pH decreased slightly (5.83) in germinated acorn flour. Similary, Gernah et al. (2011) found a decrease in pH of germinated maize (*Zea mays*) flour. This change in pH during germination could be a result of hydrolysis of some complex organic molecules like lipids and proteins (Gernah et al., 2011).

Starch content of flours varied from 23.8% to 34.2% (Table 2) and there were significant differences ($P \leq 0.05$) in starch content values among flour samples. Starch value in natural acorn flour (34.2%) is higher than (33.5%) mentioned by Correia et al. (2009) for *Quercus rotundifolia* flour. Starch content decrease by germination (23.8%) and fermentation (31.15%) treatments. Chinma et al. (2009) and Gernah et al. (2011) found a decrease in starch content of germinated tigernut (*Cyperus esculentus*) and fermented maize (*Zea mays*) flours, respectively. The decrease in starch by germination may be attributed to an increase in alpha-amylase activity which breaks down complex carbohydrates into simpler and more absorbable sugars used during germination (Chinma et al., 2009). Also, metabolic activities for growth reduce carbohydrate level during fermentation. The decrease may also be attributed to the conversion of carbohydrate to glucose and by fermenting microorganism as energy source (Ojokoh and Bello, 2014).

Total phenolic content in acorn flours ranged from 0.52 to 0.61% (Table 2) and there were significant differences ($P \leq 0.05$) in total phenolic content values among flour samples. Phenolic content in natural acorn flour (0.61%) is lower than (0.62%) found by Belarbi (2003) and that (0.631%) reported by ElMahi et al. (2016) for acorn (*Quercus ilex*) flour but higher than (3.778 times ten power three micrograms per gram) mentioned by Hegazy et al. (2014) for chestnut (*Castanea sativa* Mill.) flour. Total phenolic content found by Ghaderi-Ghahfarrokhi et al. (2017) for acorns (*Quercus branti*) and (*Quercus castaneifolia*) flours is 4.48 and 9.61 grams per one hundred grams, respectively. Diversity in quantities of polyphenols and other phytochemicals present in plant foods may be due to variety fruit, plant genetics, sunlight, soil composition, season, region of cultivation, stage of maturity and post harvest maturity (Ghaderi-Ghahfarrokhi et al., 2017). Acorn contains considerable amounts of tannin and other phenolic substances. They are classified in category of antinutritional group along with other components such as phytats, lectins, enzyme inhibitors, saponins, etc. (Ghaderi-Ghahfarrokhi et al., 2017). All treatments conducted in this work caused a significant decrease in total phenolic content of acorn flours. The highest reduction in total phenolic content was caused by germination (0.52%) followed by fermentation (0.55%) treatment. Ghavidel and Prakash (2007) and Ojokoh and Bello (2014) reported a reduction in antinutrients content in germinated legume seeds and fermented millet (*Pennisetum glaucum)* and soybean (*Glycine max)* blend flours, respectively.

Lightness (L*) of natural acorn flour (72) is lower than (75.2) reported by Correia et al. (2009) and that (78.83) found by Hegazy et al. (2014) for acorn (*Quercus suber*) and chestnut (*Castanea sativa* Mill.) flours, respectively. Lightness (L*) value of acorn flour decreased by fermentation (67.03) and germination (67.3) treatments. These observations are in agreement with earlier studies by Hallén et al. (2004) who reported a reduction in lightness of enriched wheat flour by fermented and germinated cowpea flours. Lightness (L*), chromaticity (c*), hue angle (h°) and total color difference (TCD*) values of acorn flours (Table 2) were similar to those found by Correia et al. (2009) for acorn (*Quercus suber*) flours.

Functional properties of acorn flours

Table 3 shows functional properties of natural and treated acorn flours. Bulk density of acorn flour (0.64 g/cm^3) decreased by fermentation treatment (0.59 g/cm^3). Similary, Adebowale and Maliki (2011) found a decrease in bulk density of fermented pigeon pea *(Cajanus cajan)* seed flour. The low bulk density could be attributed to the relatively lower protein content (Oppong et al., 2015). Germination treatment decreased the bulk density of acorn flour from 0.64-0.48 g/cm^3. Similar observations of lowered bulk density by germination treatment was reported by Chinma et al. (2009) and Elkhalifa and Bernhardt (2010) for tigernut and sorghum

Table 3
Functional properties of acorn flours

Parameters	Natural acorn flour	Fermented acorn flour	Germinated acorn flour
BD (g/cm^3)	0.64 ± 0.015^{aA}	0.58 ± 0.015^{aB}	0.48 ± 0.005^{aC}
WHC (g/g)	1.03 ± 0.03^{bAB}	1.05 ± 0.02^{bBA}	1.10 ± 0.01^{bC}
OHC (g/g)	0.76 ± 0.015^{cAB}	0.83 ± 0.035^{cBAC}	0.9 ± 0.1^{cCB}
EA (%)	41.28 ± 0.02^{dA}	41.72 ± 0.02^{dB}	42.09 ± 0.005^{dC}
FC (%)	7.63 ± 0.03^{eA}	6.83 ± 0.02^{eB}	8.09 ± 0.005^{eC}
SP (%)	6.7 ± 0.1^{fAB}	6.59 ± 0.01^{fBA}	7.01 ± 0.015^{fC}

BD: bulk density, WHC: water holding capacity, OHC: oil holding capacity, EA: emulsifying activity, FC: foaming capacity, SP: swelling power. Superscript values with different letters (lower-case in same column or upper-case in the same line) indicate significant difference ($P \leq 0.05$) analyzed by Duncan's multiple range test.

flours, respectively. According to Ocheme et al. (2015), the reduction in bulk density observed in germinated flour may be due to the breakdown of complex compounds such as starch and proteins as a result of the modification occurring during germination.

Water absorption capacity of germinated acorn flour (1.10 grams per gram) was higher than natural acorn flour (1.03 grams per gram). These observations are in agreement with earlier studies by Ocheme et al. (2015) who found an increase in water holding capacity of germinated sorghum flour. The increase in water absorption capacity by germination treatment could be attributed to the change in the quality of protein during germination and also the breakdown of polysaccharide molecules; hence the sites of interaction with water and holding water would be increased (Elkhalifa and Bernhardt, 2010). Water absorption capacity of acorn flour increased by fermentation treatment (1.05 grams per gram). Adebowale and Maliki (2011) found an increase in water holding capacity of fermented pigeon pea (*Cajanus cajan*) seed flour.

The increase in oil absorption capacity was recorded in germinated acorn flour (0.9 grams per gram). Similary, Chinma et al. (2009) reported an increase in oil absorption capacity of germinated tigernut (*Cyperus esculentus*) flours. The increase in oil absorption capacity could be attributed to the change in the quality of protein upon germination and also its capacity to hold fat globules as the amount of lipophilic protein increases. Furthermore, the decrease in fat content of germinated acorn flour might have resulted in its ability to absorb more oil in its structure (Chinma et al., 2009).

Emulsion capacity of different acorn flours ranged between 41.28% and 42.09% (Table 3) and there were significant differences ($P \leq 0.05$) in emulsion capacity values among flour samples. The increase observed in the emulsion capacity of germinated acorn flour (42.09%) might be attributed to dissociation and partial unfolding of polypeptides that expose the hydrophobic sites of amino acids, which aids hydrophobic association of the peptide chains with the lipid droplets (Elkhalifa and Bernhardt, 2010). These observations are in agreement with earlier studies by Chinma et al. (2009) and Elkhalifa and Bernhardt (2010) who observed an increase in emulsifying activity of germinated tigernut (*Cyperus esculentus*) and sorghum flours, respectively.

Foam capacity of acorn flour increased by germination (8.09%) and decreased by fermentation treatment (6.83%) and there were significant differences ($P \leq 0.05$) in foam capacity values among flour samples. Similary, Chinma et al. (2009) found an increase in foaming capacity of germinated tigernut (*Cyperus esculentus*) flour and Adebowale and Maliki (2011) reported a decrease in foam capacity of fermented pigeon pea (*Cajanus cajan*) seed flour. During germination, the amount of soluble proteins increased, resulting in improved foam capacity. Germination may have caused surface denaturation of proteins and reduced the surface tension of the molecules, which gave good foamability (Elkhalifa and Bernhardt, 2010).

Swelling capacity of acorn flours varied between 6.59% and 7.01% with germinated acorn flour showing the higher value. Swelling capacity for acorn flour (six point seven percent) is lower than (10.04%) reported by Correia and Beirao-da-Costa (2011) for acorn (*Quercus rotundifolia*) flour. Difference may be justified by variation in the botanical source (Correia et al., 2009). Swelling capacity of acorn flour decreased with fermentation treatment from 6.7% to 6.59%. These observations are in agreement with earlier studies by Adebowale and Maliki (2011) who found a decrease in swelling power of fermented pigeon pea (*Cajanus cajan*) seed flour. Swelling capacity of acorn flour increased by germination treatment from 6.7% to 7.01%. Ocheme et al. (2015) reported an increase in swelling capacity of sorghum flour as a result of germination. The increase in swelling capacity was probably due to an increase in soluble solids brought by the breakdown of lipid, fiber and larger amount of amylase-lipid complex in flour that could inhibit the swelling of starch granules (Ocheme et al., 2015).

Principal component analysis (PCA) of acorn flours properties

Relationships between acorns flours properties were obtained using factorial principal component analysis (PCA). The original data set was renormalized by an autoscaling transformation (data not shown) and different parameters were analyzed by a multivariate approach (Popovic et al., 2013). The two first principal components (PCs) were sufficient to explain the maximum variation in all original data. Figure 2 shows plots of loadings (Fig. 2A) and scores (Fig. 2B) obtained from PCs, where the first two principal components (PC1 and PC2) accounted for 93.67% of the total variance of the data. In particular, PC1 explained 55.64% of the variation of the data, while PC2 explained 38.03% (Aponte et al., 2014).

For Fig. 2A, in the unit circle, parameters (lightness (L*), water holding capacity (WHC), emulsifying activity (EA), oil holding capacity (OHC), total phenolic content (TPC), bulk density (BD), starch (S) and total colour difference (TCD*)) are well presented in the first axis than the others (swelling power (SP), chromaticity (c*), foaming capacity (FC), pH, organic matter (OM), dry matter (DM), moisture (M), ash (A), total titratable acidity (TTA) and hue angle (h°)) in the second axis. Fig. 2A divided the preceding parameters very well in the following way:

Axis 1, for PC1:

Lightness (L*), water holding capacity (WHC), emulsifiying activity (EA) and oil holding capacity (OHC), are strongly negatively correlated with total phenolic content (TPC), bulk density (BD), starch (S) and total color difference (TCD*). These variables contribute strongly to the formation of axis 1;

Lightness (L*), water holding capacity (WHC), emulsifying activity (EA) and oil holding capacity (OHC) have a great effect on PC1 than total phenolic content (TPC), bulk density (BD), starch (S) and total color difference (TCD*), because they were positively correlated by PC1 and any increase in these variables produces an increase in PC1. On the other hand, total phenolic content (TPC), bulk density (BD), starch (S) and total color difference (TCD*) were negatively correlated by PC1;

Emulsifying activity (EA) and oil holding capacity (OHC) parameters are positioned closely due to the significant positive correlations among them (Popovic et al., 2013). According to Zouari et al. (2016), flour proteins act as surface active agents and stabilize the emulsion by performing electrostatic repulsion on oil droplet surface.

Axis 2, for PC2:

Swelling power (SP), chromaticity (c*), foaming capacity (FC), pH, organic matter (OM) and dry matter (DM) are strongly negatively correlated with moisture (M), ash (A), total titratable acidity (TTA) and hue angle (h°). These variables contribute strongly to the formation of axis 2 ;

Swelling power (SP), chromaticity (c*), foaming capacity (FC), pH, organic matter (OM) and dry matter

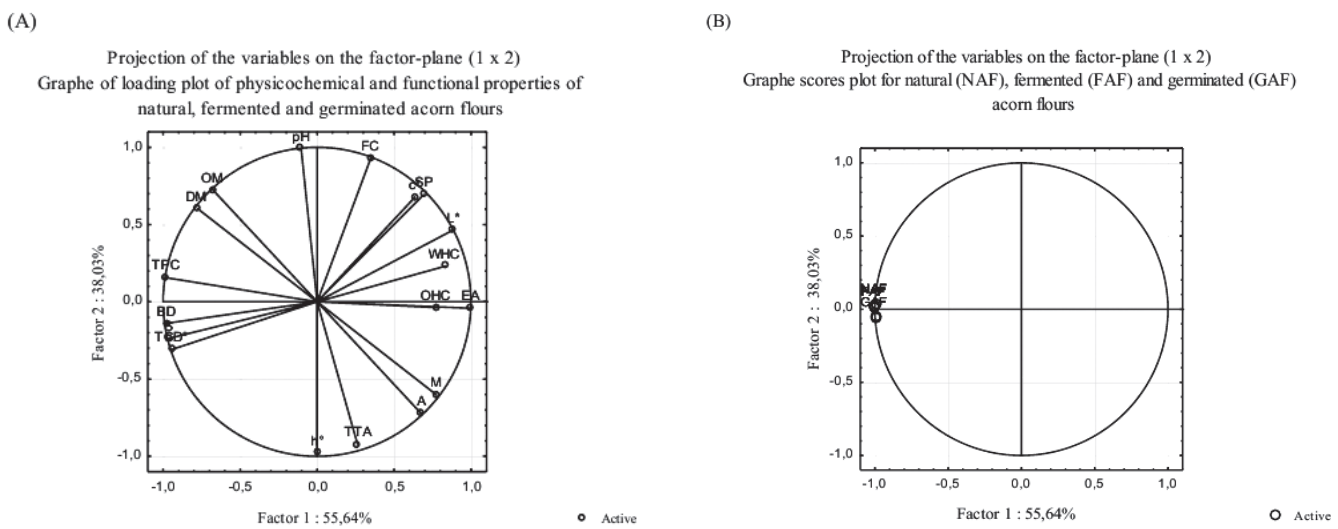

Fig. 2. Graph of loading plot of physicochemical and functional properties (A) and scores plot (B) for acorn flours

M: moisture, DM: dry matter, A: ash, OM: organic matter, S: starch, pH, TTA: total titratable acidity, TPC: total phenolic content, L*: lightness, c*: chromaticity, h°: hue angle, TCD*: total color difference, BD: bulk density, WHC: water holding capacity, OHC: oil holding capacity, EA: emulsifying activity, FC: foaming capacity, SP: swelling power, NAF: natural acorn flour, FAF: fermented acorn flour, GAF: germinated acorn flour. Parameters with close interdependence and correlation are close to each other and vice versa. Flours samples that are close to each other possess similar properties statuses.

(DM) have a great effect on PC2 than moisture (M), ash (A), total titratable acidity (TTA) and hue angle (h°), because they were positively correlated by PC2 and any increase in these variables produces an increase in PC2. On the other hand moisture (M), ash (A), total titratable acidity (TTA) and hue angle (h°) were negatively correlated by PC2;

Opposite direction of ash (A), moisture (M) and total titratable acidity (TTA) on one side and organic matter (OM), dry matter (DM) and pH on another side, indicates that ash (A), moisture (M) and total titratable acidity (TTA) are the major contributors of organic matter (OM), dry matter (DM) and pH, respectively (Popovic et al., 2013).

For Fig. 2B, the scores distribution allowed for clustering of the samples into three groups (natural acorn flour (NAF), fermented acorn flour (FAF) and germinated acorn flour (GAF)). The difference between these groups is based on PC1 (total phenolic content (TPC), bulk density (BD), and starch (S)) (Popovic et al., 2013). All three groups (natural acorn flour (NAF), fermented acorn flour (FAF) and germinated acorn flour (GAF)) were negatively scored on PC1 (Aponte et al., 2014). Natural acorn flour (NAF) and fermented acorn flour (FAF) showing positive scores on PC2 but germinated acorn flour (GAF) entirely located in the negative part. As expected, fermented acorn flour (FAF) was located more proximate to natural acorn flour (NAF) rather than germinated acorn flour (GAF). Therefore, fermented acorn flour (FAF) and natural acorn flour (NAF) possesses similar properties statuses (Popovic et al., 2013). In particular, natural acorn flour (NAF) proved to be strongly characterized by total phenolic content (TPC) (Aponte et al., 2014). Total phenolic content in natural acorn flour (NAF) is higher than fermented (FAF) and germinated (GAF) acorn flours. Results of principal component analysis (PCA) revealed the influence of fermentation and germination treatments on the physicochemical and functional properties of acorn flours (Aponte et al., 2014).

Conclusions

The research showed that fermentation and germination treatments improve functional properties (water absorption capacity: 1.05-1.10 grams per gram, emulsifying activity: 41.72-42.09% and oil holding capacity (0.83-0.9 grams per gram), increase mineral content (2-2.06%) but decrease starch (31.15-23.8%) and phenols content (0.55-0.52%) of acorn flour. The obtained results suggest that the incorporation of natural and treated acorn flours may help enhancing nutritional value in food formulations.

Acknowledgement

The authors would like to acknowledge technicians of Food Science Laboratory (LSA) for their assistance throughout the preparation of this article.

References

AACC (American Association of Cereal Chemistry) (2000). Official Methods of Analysis, 9th edition, USA, St. Paul., Minnesota.

AACC (American Association of Cereal Chemistry) (2012). Official Methods of Analysis, 11th edition, USA, St. Paul., Minnesota.

Adebowale, O. J., & Maliki, K. (2011). Effect of fermentation period on the chemical composition and functional properties of Pigeon pea (Cajanus cajan) seed flour. *International Food Research Journal*, *18*(4), 1329-1333.

AOAC (Association of Official Analytical Chemists) (2005). Official Methods of Analysis, 18th edition, USA, Washington, DC.

Aponte, M., Boscaino, F., Sorrentino, A., Coppola, R., Masi, P., & Romano, A. (2014). Effects of fermentation and rye flour on microstructure and volatile compounds of chestnut flour based sourdoughs. *LWT-Food Science and Technology*, *58*(2), 387-395.

Bahmani, M., Forouzan, S. H., Fazeli-Moghadam, E., Rafieian-Kopaei, M., Adineh, A., & Saberianpour, S. H. (2015). Oak (Quercus branti): an overview. *J Chem Pharm Res*, *7*(1), 634-9.

Belarbi, M. (2003). *Study of the nutritional and antinutritional compounds of three species of oak and their nutritional efficiency in the Wistar rat*. Doctoral dissertation, Abou Bekr Belkaid University*, Tlemcen*, Algeria.

Bilgiçli, N., Elgün, A., Herken, E. N., Ertaş, N., & İbanoglu, Ş. (2006). Effect of wheat germ/bran addition on the chemical, nutritional and sensory quality of tarhana, a fermented wheat flour-yoghurt product. *Journal of Food Engineering*, *77*(3), 680-686.

Cantos, E., Espín, J. C., López-Bote, C., de la Hoz, L., Ordóñez, J. A., & Tomás-Barberán, F. A. (2003). Phenolic compounds and fatty acids from acorns (Quercus spp.), the main dietary constituent of free-ranged Iberian pigs. *Journal of Agricultural and Food Chemistry*, *51*(21), 6248-6255.

Chinma, C. E., Adewuyi, O., & Abu, J. O. (2009). Effect of germination on the chemical, functional and pasting properties of flour from brown and yellow varieties of tigernut (*Cyperus esculentus*). *Food Research International*, *42*(8), 1004-1009.

Correia, P. R., Leitão, A. E., & Beirão-da-Costa, M. L. (2009). Effect of drying temperatures on chemical and morphological properties of acorn flours. *International Journal of Food Science & Technology*, *44*(9), 1729-1736.

Correia, P. R., & Beirão-da-Costa, M. L. (2011). Effect of drying temperatures on starch-related functional and thermal properties of acorn flours. *Journal of Food Science*, *76*(2), E196-E202.

Elkhalifa, A. E. O., & Bernhardt, R. (2010). Influence of grain germination on functional properties of sorghum flour. *Food Chemistry*, *121*(2), 387-392.

ElMahi, F. Z., Benali, M., DIF, M., Bouazza, S., & Rih, A. (2016). First phytochemical analysis of the anti-nutritional aspect of holm oak acorn (Quercus Ilex L) of Tessala (Algeria NW) before and after cooking. *Advances in Environmental Biology*, *10*(1), 259-264.

Fos' hat, M., Etemad, V., Gharibzahedi, S. M. T., & Ghahderijani, M. (2011). Physical, mechanical and aerodynamic properties of acorn (Quercus suber L.) as potentials for development of processing machines. *Australian Journal of Crop Science*, *5*(4), 473-478.

Galván, J. V., Novo, J. J. J., Cabrera, A. G., Ariza, D., García-Olmo, J., & Cerrillo, R. M. N. (2012). Population variability based on the morphometry and chemical composition of the acorn in holm oak (Quercus ilex subsp. ballota [Desf.] Samp.). *European Journal of Forest Research*, *131*(4), 893-904.

Gernah, D. I., Ariahu, C. C., & Ingbian, E. K. (2011). Effects of malting and lactic fermentation on some chemical and functional properties of maize (Zea mays). *American Journal of food technology*, *6*(5), 404-412.

Ghavidel, R. A., & Prakash, J. (2007). The impact of germination and dehulling on nutrients, antinutrients, in vitro iron and calcium bioavailability and in vitro starch and protein digestibility of some legume seeds. *LWT-Food Science and Technology*, *40*(7), 1292-1299.

Ghaderi-Ghahfarrokhi, M., Sadeghi-Mahoonak, A. R., Alami, M., & Mousavi Khanegah, A. (2017). Effect of processing treatments on polyphenol removal from kernel of two Iranian acorns varieties. *International Food Research Journal*, *24*(1), 86-93.

Hallén, E., İbanoğlu, Ş., & Ainsworth, P. (2004). Effect of fermented/germinated cowpea flour addition on the rheological and baking properties of wheat flour. *Journal of Food Engineering*, *63*(2), 177-184.

Hegazy, N., Kamil, M., Hussein, A., & Bareh, G. (2014). Chemical and technological properties of improved Biscuit by Chestnut Flour. *International Journal of Food and Nutritional Sciences*, *3*(6), 7-15.

Idris, W. H., Abderahman, S. M., El Maki, H. B., Babiker, E. E., & El Tinay, A. H. (2005). Effect of germination, fermentation and cooking on phytic acid and tannin contents and Hcl-extractability of minerals of sorghum (*Sorghum biocolor*) cultivars. *Journal of Food Technology*, *3*(3), 410-416.

Karioti, A., Bilia, A. R., & Skaltsa, H. (2010). Quercus ilex L.: A rich source of polyacylated flavonoid glucosides. *Food Chemistry*, *123*(1), 131-142.

Khattab, R. Y., & Arntfield, S. D. (2009). Nutritional quality of legume seeds as affected by some physical treatments 2. Antinutritional factors. *LWT-Food Science and Technology*, *42*(6), 1113-1118.

Ocheme, O. B., Adedeji, O. E., Lawal, G., & Zakari, U. M. (2015). Effect of germination on functional properties and degree of starch gelatinization of sorghum flour. *Journal of Food Research*, *4*(2), 159.

Ojokoh, A., & Bello, B. (2014). Effect of fermentation on nutrient and anti-nutrient composition of millet (*Pennisetum glaucum*) and soyabean (*Glycine max*) blend flours. *Journal of Life Sciences*, *8*(8), 668-675.

Okpala, L. C., & Okoli, E. C. (2014). Development of cookies made with cocoyam, fermented sorghum and germinated pigeon pea flour blends using response surface methodology. *Journal of Food Science and Technology*, *51*(10), 2671-2677.

Oppong, D., Arthur, E., Kwadwo, S. O., Badu, E., & Sakyi, P. (2015). Proximate composition and some functional properties of soft wheat flour. *International Journal of Innovative Research in Science, Engineering and Technology*, *4*(2), 2347-6710.

Popović, B. M., Štajner, D., Ždero, R., Orlović, S., & Galić, Z. (2013). Antioxidant characterization of oak extracts combining spectrophotometric assays and chemometrics. *The Scientific World Journal*, 1-8.

Rakić, S., Povrenović, D., Tešević, V., Simić, M., & Maletić, R. (2006). Oak acorn, polyphenols and antioxidant activity in functional food. *Journal of Food Engineering*, *74*(3), 416-423.

Sangronis, E., & Machado, C. J. (2007). Influence of germination on the nutritional quality of *Phaseolus vulgaris* and *Cajanus cajan*. *LWT-Food Science and Technology*, *40*(1), 116-120.

Li, S., Zhou, Y., Liu, M., Zhang, Y., & Cao, S. (2015). Nutrient composition and starch characteristics of *Quercus glandulifera* Bl. seeds from China. *Food Chemistry*, *185*, 371-376.

Tejerina, D., García-Torres, S., de Vaca, M. C., Vázquez, F. M., & Cava, R. (2011). Acorns (*Quercus rotundifolia* Lam.) and grass as natural sources of antioxidants and fatty acids in the "montanera" feeding of Iberian pig: Intra-and inter-annual variations. *Food Chemistry*, *124*(3), 997-1004.

Vinha, A. F., Barreira, J. C., Costa, A. S., & Oliveira, M. B. P. (2016). A new age for Quercus spp. fruits: Review on nutritional and phytochemical composition and related biological activities of acorns. *Comprehensive Reviews in Food Science and Food Safety*, *15*(6), 947-981.

Zouari, R., Besbes, S., Ellouze-Chaabouni, S., & Ghribi-Aydi, D. (2016). Cookies from composite wheat–sesame peels flours: dough quality and effect of Bacillus subtilis SPB1 biosurfactant addition. *Food Chemistry*, *194*, 758-769.

Received: November 12, 2017; *reviewed:* December 21, 2017; *accepted:* April 11, 2018