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Chemical Composition, Nutritional, and Antioxidant Activity of Two Quercus Species Acorns Growing in Morocco

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Abstract: Interest in acorns as a functional food is growing, but the absence of full chemical characterization studies seriously hinders their further development as a functional food. Herein, the nutritional composition, starch properties, and antioxidant activities of acorns collected from *Quercus rotundifolia L and Quercus faginea L were investigated. The moisture content, crude fat, protein, total ash, and carbohydrates contents* of acorn were 11.18-12.95%, 5.37-11.52%, 3.12-3.56%, 2.13-2.42%, and 83.66-88.59%, respectively. Oleic (58.1%, 63.8%), Linoleic (18.6%,23.1%), and palmitic (13.1%,13.4%) were the most predominant fatty acids. The β-sitosterol was the main component in each variety, consisting of 83.5–89% of total detected sterols. Additionally, acorns were an excellent source of minerals (K, Fe, Ca, P, and Mn). The antioxidant activity of 80% ethanol extracts was assessed. Their DPPH and ABTS+ free-radical-scavenging capacity varied from 22,06 ± 1.12 to 30,53 ± 0,55 µmol TE / g and from 18,96 ± 0,84 to 25,20 ± 0,15 µmol TE/ g, respectively. The amylose content varied from 27.9% to 30.5%. The swelling power of starches varied from 12.54 % to 19.25%, whereas the solubility index was in the range of 4.85–7.66%. These findings suggest that *Q. rotundifolia* and *Q. faginea* could be used as an interesting sources of raw material for industrial applications.

Keywords: Quercus spp; nutrient composition; starch acorn; antioxidant activity.

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1. Introduction

Consumer awareness of the link between eating and health has boosted the demand for healthy food and functional food [1]. Numerous factors involve changes in consumer demand, including health problems; the demand for convenience; changes in distribution systems and prices; and changes in demographic characteristics (such as population aging) [2]. The market for functional food is increasing at an annual rate of 15-20% [3]. With the global expansion of the functional food market, researchers are striving to provide natural food components with preventive and beneficial effects on human health [4].

Acorn is the fruit of the oak (Quercus) tree, originally belonging to the Fagaceae family. For many years, acorns have been used as animal feed and consumed by humans in many countries worldwide. Acorns are a high-nutritional-value food: starch (31-51%), proteins (2–8 %), fat (0.7–12%), Vitamin A, and essential amino acids, presenting a higher nutritional value

than cereals [5], besides being an important source of macro minerals (such as K, Mg, and Ca), and microelements (such as Fer, Zn, and Mn) [6]. Furthermore, acorns have a high content of monounsaturated fatty acids, particularly oleic acid (63%), as well as essential fatty acids such as linoleic (ω-6) (20%) and linolenic (ω-3) (2-4%) fatty acids, which are important for consumer health [7]. Furthermore, acorns contain various biologically active compounds, such as tocopherols, sterols, aliphatic alcohols, phenolic acids, triterpenoids, flavonoids, and tannins [8]. Acorn, as functional food, contains nutrients and many potential health benefits, such as anti-inflammatory, antioxidant, antibacterial, neuroprotection, antitumor, and antidiabetic effects [8–10]. In light of the above-mentioned health effects, acorn has been used to produce various food products such as hot beverages, cookies, bread, pasta, biscuits, and cakes [11]. Nevertheless, adding acorns to the human diet is still limited [10].

In Morocco, Quercus spp represents around 40% of the Moroccan forest capital (about 1814000 ha). However, no studies have been done on the chemical composition of Moroccan Quercus acorns. Therefore, the aim of the current study was to determine, for the first time, the nutritional composition, antioxidant activities, and starch characteristics of two Quercus species (*Q. rotundifolia* and *Q. faginea* acorns).

2. Materials and Methods

2.1. Collection of plant material.

Quercus rotundifolia Lam (Q.R) and *Quercus faginea* Lam (Q.F) acorns were harvested from different locations in Morrocco (Boulmane and Tetouan). After collecting the samples, They were instantly sent to the lab, scrubbed, and straightened in order to remove any fruits that were damaged. The collected samples were separated from the shell and dried at 45 °C for 72 h. The dried samples were then milled into powder and stored for analysis.

2.2. Proximate composition.

The ash and moisture content was calculated according to (AOAC, 2000). Total lipids were determined by the Soxhlet method [12]. The protein content was measured by the Kjeldahl appareil (AOAC, 2000). Total carbohydrates were calculated by subtracting 100 g from the total amount of protein, ash, and crude fat. And total energy was determined by the following formula:

$$Energy\ (Kcal) = 3.75 \times (g\ carbohydrate) + 9 \times (g\ fat) + 4 \times (g\ protein) \qquad \text{Eq 1}$$

2.3. Fatty acids.

Fatty acid methyl esters (FAME) were prepared in accordance with BDS EN ISO 12966-2:2017. The analysis was performed by a chromatograph Clarus 580 GC_G12086, equipped with an N2 PFloW. CP-Wax 52CB column for FAME (30 m × 0.25 mm i.d, 0.25 µm film thickness;) was used. The carrier gas was helium at a flow rate of 1.5 ml/min. The oven temperature programming was as follows: 100 °C for 2 minutes followed by an increase of 6 °C/min to 240°C. The temperature of the injector and detector was 260°C and 280°C, respectively; a split ratio 1:80 was used. The amounts of FA were calculated as a percentage of total FA recovery as a mean value and standard deviation (SD) according to BDS EN ISO 12966-4:2015.

2.4. Mineral contents.

The mineral constituent (Ca, Mn, Mg, P, Fe, Cu, Zn, Se, As, Pb) were determined using inductively coupled plasma-optical emission spectrometry (ICP-OES). K was evaluated by atomic absorbance spectroscopy. A sample of 1 g was ashed at 125 °C using a 1:3 mixture of nitric acid and hydrochloric acid. When the mixture had reached room temperature, it was screened and adapted to a volume of 50 mL with Ultrapure water.

2.5. Preparation of hydro-alcoholic extracts.

In brief, 10 g of sample were extracted with 100 mL of ethanol: water (80/20 v/v) at 40◦C for 60 min. The extracts were then filtered and kept at 25°C till they were tested for antioxidant activity and polyphenol quantification.

2.5.1. Total phenolic content.

The quantity of total phenolics in acorns was assessed in accordance with [13]. Briefly, 0.1 ml of extract was combined with 500 µl of Folin-Ciocalteu reagent and 400 µl of sodium carbonate solution (7.5%, m/v). The combination was then incubated for 15 min at 45 C, followed by 30 min at room temperature. The total phenolic content was calculated using gallic acid as a standard and the absorbance of the mixture at 765 nm.

2.5.2. Total flavonoid content.

The total flavonoid of the extract was assessed using the method mentioned (Didi, 2020). Catechin was used as the standard, and the total flavonoid content was expressed as mg of catechin equivalents (CE)/g of extract.

2.5.3. Determination of antioxidant activity.

The antioxidant activity was assessed using ABTS (2.20-azinobis (3-ethylbenzothiazoline-6-sulphonic acid). Briefly, 50 µL of each extract was mixed with 950 µL of ABTS reagent. The decrease in absorbance was assessed at 734 nm. The results were presented as µmol Trolox equivalents (TE) per g of dry matter [14].

The antioxidant ability of the extracts was also measured by the 2.2 diphenyl-1-picrylhydrazyl (DPPH) method. Briefly, 50 µL of each sample was added to 950 µL of DPPH solution. After 30 min in the dark, the decrease of absorbance was determined at 515 nm read.

2.6. Quantification of chlorophylls and carotenoids.

The chlorophylls (a and b), lycopene, and β-carotene were analyzed using the method described by [12]. In brief, 1g of samples was mixed with 20 mL acetone/hexane (8:12, v/v) and vortexed for 40 minutes. After that, the absorbance of the mixture was assessed at 453, 505, 645, and 663 nm. The chlorophyll and carotenoid contents were evaluated using the equations below:

$$Chlorophyll\ a\ (\frac{mg}{100mL}) = 0.999A663 - 0.0989A645 \quad \text{Eq 2}$$

$$Chlorophyll\ b\ \left(\frac{mg}{100mL}\right) = -0.328A663 + 1.77A645 \quad \text{Eq 3}$$

$$Lycopene\left(\frac{mg}{100mL}\right) = -0.0458A663 + 0.204A645 + 0.372A505 - 0.0806A453 \quad \text{Eq 4}$$

$$\beta - carotene\left(\frac{mg}{100mL}\right) = 0.216A663 - 1.22A645 - 0.304A505 + 0.452A453 \quad \text{Eq 5}$$

The results of the samples were presented as mg/100 g of dried sample.

2.7. Starch preparation.

Starch was isolated, according to [15]. Acorn flour was drenched in 0.3 % NaOH for 2 hours. A 180-mesh sieve was used to screen the dispersion. After allowing the starch milk to settle, the supernatant was discarded, and the precipitate was rinsed with distilled water and left to dry at 40 °C for 24 h.

2.7.1. Amylose content.

A colorimetric method (based on amylose-iodine complex formation) was applied to determine the amylose content of starch [16]. Approximately 0.1 g of acorn starch, 1 mL of ethanol (95%), and 9 mL of NaOH (1N) were combined and boiled for 15 min before adding distilled water up to 100 mL. 5 ml of the mixture was mixed with 1 mL of acetic acid solution (1 N) and 2 mL of iodine solution. In addition, the mixture was diluted to 100 ml with distilled water and incubated for 20 minutes at room temperature, and the absorbance at 620 nm was measured. The amylose content of each sample was calculated using the following formula:

$$Amylose\ content\ (\%) = 3.06 \times A\ (Absorbance\ value) \times 20 \quad \text{Eq 6}$$

2.7.2. Swelling power and solubility.

The method reported by [16] was used to measure the swelling power (Sp) and solubility (S) of acorn starches. Briefly, 2% Starch mixes (starch/distilled water) were boiled for 30 minutes in a water bath at 60, 70, 80, and 90 °C with constant stirring. In addition, the samples were centrifuged for 10 minutes at 3000g after being heated. The mixture was discarded and left to dry at 105 °C, and the precipitate was assessed. The swelling power (SP, g/g) and solubility (S, %) were assessed using the fowling formula :

$$Sp\left(\frac{g}{g}\right) = \frac{Sediment\ weight}{masse\ of\ dry\ starch} \times (100 - S) \quad \text{Eq 7}$$

$$S(\%) = \left(\frac{Masse\ of\ dried\ supernatant}{masse\ of\ dry\ starch\ weight}\right) \times 100 \quad \text{Eq 8}$$

2.7.3. X-ray diffraction.

The isolated starches were analyzed applying an X-ray diffractometer (D8-Advance, Bruker., Germany) set to 40 kV, 100 mA, 0.01 (slit) in the 2-region of 5-80.

2.7.4. Scanning electron microscopy (SEM).

The morphology of the isolated starches was investigated with scanning electron microscopy SEM (JEOL JSM-35, Tokyo, Japan).

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3. Results and Discussion

3.1. Acorn flours.

Table 1 shows the chemical composition of acorn flours. This table shows that the two species' ash, fat, and carbohydrate content differed significantly ($p< 0.05$). The result showed that the moisture content of acorn flours ranged from 11.18% to 12.95%. This is consistent with prior findings for other acorn species flours [17]. Moisture content is an important parameter in flour storage, as levels greater than 13g/100g may cause caking and/or a rise in the microbial growth rate. Hence, the low moisture content is appropriate for long flour storage and thus suitable for industrial processing. Total ash content was considerably higher for all acorn flours (2.1 to 2.4 g/100 g). The ash content was consistent with previous findings on the ash content of different acorn species. [12,17]. The higher ash content of acorn flour indicates that they have the ability to improve the dietary intake of minerals when blended with low-ash flour (such as wheat). The fat content in acorn flours varied significantly ($P<0.05$), (Table 1) and ranged from 5.3 to 11.5%. When compared to previous studies, the fat content of (Q.F) and (Q.R) flour was found to be relatively high (2-8.44%, respectively)[12] [17]. It was remarkable that acorn flour had a much higher fat content than that oat, buckwheat, wheat, or whole rye flour [5]. Protein content ranged from 3.12-3.56. Compared to previous data, the protein content observed in this study was relatively low [10, 18]. In general, acorn flours exhibit low protein content when compared with conventional cereal flour [5]. Nevertheless, the proteins in acorn flour are gluten-free. Hence, acorn flour is an alternative to gluten-free flour used in producing bread and pastry products[5]. In addition, acorn flours are rich in carbohydrates (83–88 %). According to the literature, starch is the most prevalent carbohydrate, with a share ranging from 31.4 to 49.0 %, depending on the isolation method used in flour production[11]. Finally, (Q.R) acorn flour had the highest energy value (425 15 kcal/100g), possibly due to its higher fat content, while (Q.F) acorn flour had 394 kcal/100g.

Table 1. Proximate composition of acorn and acorn starch.

Chemical compositions (%)	**Q.R**		**Q.F**	
	Acorn powder	**Acorn starch**	**Acorn powder**	**Acorn starch**
Moisture	11.18 ± 0.15	14.45± 0.07	12.95± 0.27	15.60± 0.1
Crude Ash	2.139 ± 0.8	0. 217± 0.28	2.471± 0.7	0.387± 0.45
Crude protein	3.124 ± 0.95	1.05 ± 0.24	3.56 ± 0.65	1.11± 0.53
Crude Fat	11.52 ± 1.58	0.41± 0.17	5.37 ± 0.85	0.39 ± 0.08
Carbohydrate	83.217± 2.14	98.23± 0.65	88.599 ± 1.45	98.21± 0.77
Energy (Kcal/100g)	425.11 ± 4.25	-	394.81 ±3.75	-
Amylose content		27.9 ± 1.45		30.5 ± 0.87

3.2. Acorn starch.

Table 1 shows the chemical composition of starches extracted from acorn flours. The moisture content is within the acceptable range (14.45–16.0%) for native starches (<20%).[18]. The ash content ranges from 0.21 to 0.38 %, well within the accepted level for industrial starch (0.5%) [16]. The obtained values did not differ significantly ($p > 0.05$). The lipid content ranges from 0.39 to 0.41 %. These findings are similar to those of corn starch (1.24 %) and Ramon starch (1.27%) [19]. The obtained starch reveals high values of amylose content (27-29), which is regarded as superior to other sources of starch such as sago (26%), Potato (20%), Cassava (17%), wheat (26%), sweet potato (18%), and maize starches (28%). Differences in

amylose contents are mainly ascribed to environmental factors, genotypic differences, climatic conditions, starch extraction methods, and harvest time [20].

3.3. Fatty acids composition.

The fatty acid profiles of acorn oil are presented in Table 2. As shown, the most abundant fatty acids of (Q.R) and (Q.F) were oleic (63.8, 58.1), followed by linoleic (18.6, 23,6%) and palmitic (13.3, 13.4%), respectively. Stearic acid (C18:0) level was low in both species, with a similar range (2.2% and 1.7%). The quantity of α-linolenic acid (α-C18:3) differed significantly ($p \leq 0.05$) between *Q. rotundifolia* (1 %) and *Q. faginea* (2.2%). Furthermore, minor quantities of myristic (C14:0), palmitoleic (C16:1), heptadecanoic, (17:0) heptadecenoic (17:1) arachidic (C20:0), and eicosenoic (C20:1) acids were detected (below 0.5%). These findings align with the values that were reported by [21–23]. The minor difference in fatty acid levels noticed within the same species could be explained by various factors, including oak acorn maturity, differences in oil processing, environmental conditions, or different harvest dates [24,25]. As previously stated, the main monounsaturated fatty acid (MUFA) found in acorn oils is oleic acid. Numerous studies have clearly demonstrated the health benefits of this fatty acid, particularly in reducing cardiovascular disease risk and preventing diabetes mellitus T2DM, besides contributing to improving hemostasis, glucose metabolism, and endothelial dysfunction [26].

The ratio PUFAs /SFAs was shown to be a useful indicator to evaluate the quality of fat. Hence, the higher this ratio, the better effect. In the current study, the PUFAs /SAFs index ranged from 1.24-1.63, which was higher than the recommendations (0.45) [3]. In addition to the PUFAs/SFAs ratio, the AI, TI, and HH index were also used in this study to judge the FAs' nutritional quality. The indices of AI and TI are linked to anti-atherogenic and pro- and anti-thrombogenic FAs[27]. High AI and TI values may promote platelet aggregation and, as a result, the formation of thrombus and atheroma in the cardiovascular system [28]. FAs with lower AI and TI provide better nutritional quality, and their consumption may help to prevent cardiovascular disorders. The HH index assesses the effect of FA composition on cholesterol metabolism [29]. In opposition to AI and TI, higher HH values are considered more beneficial for the human body. The AI and TI of acorn species in the present investigation were 0.15-0.16 and 0.17-0.22, respectively. The HH ratios ranged between 6.17-6.36. These ratios were similar to those typically detected in linseed, sesame, and olive oils [30]. Therefore, acorn oil may be considered a good source of essential fatty acids for human consumption.

Table 2. Fatty acid composition of *Q.rotundifolia* and *Q.faginea* oils (%).

Fatty acid	Q.R	Q.F
Myristic (C14:0)	-	0.1 ±0.1
Palmitic (16:0)	13.1 ±0.01	13.4 ±0.00
Palmitoleic acid (16:1)	0.1 ±0.01	0.2 ±0.03
Heptadecanoic acid (17:0)	0.1 ±0.02	0.1 ±0.1
Heptadecenoic acid (17:1)	0.1 ±0.01	0.1 ±0.01
Stearic acid (18:0)	2.2 ±0.1	1.7 ±0.07
Oleic acid (18:1)	63.8 ±0.1	58.1 ±0.2
Linoleic acid (18:2)	18.6 ±0.01	23.1 ±0.03
Linolenic acid (18:3)	1 ±0.01	2.2 ±.0.02
Arachidic acid (20:0)	0.3 ±0.2	0.2 ±0.09
Eicosenoic (C20:1)	0.5 ±0.01	0.5 ±0.01
ΣSFA	15.7	15.5
Σ MUFA	64.5	58.9

Fatty acid	Q.R	Q.F
Σ PUFA	19.6	25.3
Σ PUFAs / ΣSFA	1.24	1.63
AI	0.15	0.16
TI	0.22	0.17
HH	6.36	6.17

PUFA: polyunsaturated fatty acids; MUFA: monounsaturated fatty acids; SFA: saturated fatty acids;

$$AI{:}\,Atherogenic\ index = \frac{(C12{:}0 + C14{:}0 + C16{:}0)}{(\omega - 3PUFAs + \omega - 6PUFAs + MUFAs)}$$

$$TI{:}\,Thrombogenic\ index = \frac{(C14{:}0 + C16{:}0 + C18{:}0)}{(0{,}5\omega - 6PUFAs + \omega - \frac{3PUFAs}{\omega - 6PUFAs})}$$

$$HH{:}\frac{Hypocholesterolemic}{Hyperchloesterolemic} = \frac{(cis - C18{:}1 + \sum PUFA)}{(C12{:}0 + C14{:}0 + C16{:}0)}$$

3.4. Phytosterol composition.

Ten phytosterols were detected in acorn oils (Table 3). The predominant sterol in both species was β-sitosterol (89.59-91.9%), followed by campesterol (3.2–4.12%) and stigmasterol (2.3–3.56%). The remaining sterols (Cholesterol, clerosterol, Δ5-avenasterol, Δ5,24–stigmastadienol, Δ7-avenasterol, and Δ7-stigmastenol) were below 2.0% in both species Table 3. In terms of sterol composition, there were no significant differences between acorn species. β-sitosterol was found to be as the predominant sterol produced from such acorn species as *Q. rotundifolia* L (88.21%) and Q. Suber L(83.52%)[31], Q. ilex and Q. suber (87.74–92.53%) [32] Q. calliprinos (78.7%)and Q. robur (64.3%), Q. rubra (68.1%) ; Q. ilex and Q. coccifera [33]. This indicates that the phytosterol composition weakly depends on species and geographical locations. This sterol is also abundant in olive oil, sesame oil, soybean oil, and sunflower oil [34,35]. The current research demonstrated that acorn oils have a high nutrient value because phytosterols are known to provide various biological benefits. They can act as anticarcinogenic, like protection against colon, breast, and prostate cancers [36]. Furthermore, several studies have documented the ability of these compounds to lower serum low-density lipoprotein levels (LDL). Moreover, phytosterols are also known to have antioxidative, antibacterial, anti-inflammatory, anti-atherosclerotic, and ant-ulcerative properties in humans [37].

Table 3. Sterol composition (%) in Q.rotundifolia and Q.faginea oils (g/100g).

Sterol	Q.R	Q.F
Cholesterol	0.3± 0.02	0.19±0.1
Campesterol	3.2± 0.11	4.12±0.09
Campestanol	0.05±0.01	0.1±0.01
Stigmasterol	2.3±0.08	3.56±0.22
Clerosterol	0.47± 0.01	0.91±0.18
β-sitosterol	91.9±2.11	88.95± 1.45
Δ5-avenasterol	1.27±0.01	1.41±0.03
Δ5,24-stigmastadienol	0.21±0.02	0.17±0.01
Δ7-stigmastenol	0.2±0.01	0.37±0.01
Δ7-avenasterol	0.1±0.01	0.22±0.01

3.5. Total polyphenol, flavonoid, chlorophyll, carotenoid content, and antioxidant activity.

Phenolic compounds in plants are a type of secondary metabolite that originate from the pentose phosphate, shikimate, and phenylpropanoid pathways [38]. These compounds had different biological activities such as anti-microbial, anti-aging, antioxidant, anti-inflammatory, anti-atherogenic, anti-allergenic, and cardioprotective [39]. Hence, the total phenolic and flavonoid contents of the acorns were evaluated. The total polyphenol contents were found to be 98.3 and 212.5 mg GAE/g of dry extract for (Q.F) and (Q.R), respectively. The total flavonoid contents were 6.2 and 10.3 mg /g of dry extract for (Q.F) and (Q.R), respectively. The phenolic contents differed significantly ($p< 0.05$) between acorn species. These amounts were higher than those found for acorns of Q. fageina[21] and for *Q. rotundifolia* acorns [40]. The TFCs also were higher than those found for other acorn species from an eastern Algeria forest (*Q. ilex*, *Q. suber*, and *Q. coccifera* (63.3-119.4 mg GAE·100 g−1)[41]but lower than those reported for Q.ilex (18.54 mg/g)[42]. The contents of chlorophyll (a,b), β-carotene, and lycopene were also determined. As shown in Table 4, the Chla content ranged from 0.057 to 0.121mg/; Chlb content varied from 0.010 to 0.014 mg/g; β-carotene varied from 0.132 to 0.542 mg/g; the lycopene content ranged from 0.068 to 0.098 mg/g. The total Chlb, lycopene, and β-carotene content between the acorn species were statistically similar ($p \geq 0.05$) except chlb ($p < 0.05$). The content of chlorophyll a in (Q.F) and (Q.R) was significantly higher than chlorophyll b. This was in line with the findings of [12,43], which found that the chlorophyll a and b in acorn species range from 0.016 to 5 mg g-1 and 0.003 to 0.0017 mg g-1, respectively. The lycopene content of (Q.F) was lower than in previously reported data[12].

The antioxidant capacities of the extracts were analyzed using the free radical scavenging capacity (DPPH) and the radical cation scavenging capacity (ABTS) (Table 4). Both experiments showed that acorns from *Q. rotundifolia* had the highest antioxidant activity (Table 4). These variations could be attributed to differences in nutritional composition and phytochemical content. In addition, numerous studies have established that high amounts of phenolic compounds in acorn extracts are associated with significant antioxidant activity [6] [44]. Furthermore, several researchers have revealed a significant positive correlation between antioxidant activity and the total phenolic content of fruits and vegetables [45, 46]. Comparing the antioxidant activity of the acorn species used in these studies is difficult because the different antioxidant capacity assays have not been standardized [21]. Our results revealed that acorn extracts might be a rich and novel source of natural antioxidants and might be used as a food additive.

Table 4. Total polyphenol, flavonoid, Chlorophylls, carotenoids, and antioxidant profiles of acorn species.

Species	TPC (mg GAE/g)	TFC(mg CE/g)	DPPH (µmol TE/g)	ABTS (µmol TE/g)	Chla (mg/g)	Chlb (mg/g)	Lycopene (mg/g)	β-carotene (mg/g)
Q.R	212.5 ± 2.15	10.3 ± 0.25	30,53 ± 0,55	25,20 ± 0,15	0.057 ± 3.45	0.014± 0.58	0.068 ± 2.14	0.532 ± 0.78
Q.F	98.3 ± 1.15	6.2 ± 0.86	22,06 ± 1.12	18,96 ± 0,84	0.121 ± 2.01	0.010 ±1.02	0.091± 1.17	0.786 ± 0.95

3.6. Mineral content.

Table 5 shows the mineral content of acorn flour. K, P, Ca, and Mg were clearly the most abundant elements, with values within the ranges 673–930, 90.1-125, 48.4-60.2, and 76.5-90.2 (mg/100g), respectively. The mineral contents of both species differ significantly (p

<0.05). Rababah et al., 2008 described several acorn flours in the aspect of mineral content. Their results for selected minerals in *Q. ithaburensis* and *Q. calliprinosare* acorns are low than those presented in our study. A similar amount of K (773 mg of K/100 g) was also found in acorn flour[48] [15] also observed similar contents for Ca and Fe and higher values for K (2088mg/100g) and Mg (102.9mg/100g) from *Quercus glandulifera* Bl. acorns. Furthermore, when compared to conventional flours, acorn flour contains a considerable amount of iron, manganese, and copper[5].

Considering the recommended mineral intake (USDA, 2013), the content of K (673-930 mg/100 g), Fe (14.30-20.75 mg/100 g), Mn(2.61-7.86 mg/100 g), and Cu (0.27-0.81 mg/100 g) seemed to be significant for adult males and females. Therefore, the use of acorn flours in food products may contribute significantly to meeting these dietary requirements.

Table 5. The mineral content of acorn flour.

Mineral (mg/100g)	*Q.R*	*Q.F*	Adult DIRs (male, female)
K	930 ± 43	673 ± 28	(4.7,4.7) g/day
P	125.03± 12	90.12 ±23	(500,700) mg/day
Ca	60.21 ± 8	48.45 ±15	(1.0,1.0) g/day
Mg	76.51 ± 3	90.23 ± 7	(420.320)mg/day
Fe	14.30 ± 1.25	20.57 ±4.58	(8,18) mg/day
Zn	1.38 ± 0.6	3.51 ± 1.54	(7.0,4.9) mg/day
Mn	2.61 ± 0.45	7.86 ± 1.28	(2.3.1.8)mg/day
Cu	0.81 ±0.1	0.27 ± 0.74	(900.900)ug /day
As	Nd	Nd	
Se	Nd	Nd	
Pb	Nd	Nd	

3.7. Acorn starch characteristics.

3.7.1. Swelling power and solubility.

Table 6. Solubility and swelling power of isolated acorn starches.

	Swelling power g/g				Solubility %			
	Temperature °C				Temperature °C			
	60	**70**	**80**	**90**	**60**	**70**	**80**	**90**
QR	8,59 ± 0.05	11,43± 0.12	12.83± 0.35	19.25±0.45	1.02± 0.1	2.45±0.2	4.17±0.25	7.66± 0.24
QF	5.69± 0.12	7.76±0.55	10.73±0.7	12.54± 0.81	0.78± 0.08	2.18± 0.08	2.77± 0.49	4.85± 0.63

The swelling and solubility index offer information on interactions among starch chains and water molecules in the crystalline and amorphous regions. The swelling power and solubility of the isolated acorn starches in the range from 60 to 90 °C are presented in Table 6. The swelling power of starch samples increased significantly ($P \leq 0.05$) as the temperature was raised from 50 to 90 °C. The swelling powers in the QF and QR starches were 12.54 g/g and 19.25 g/g at 90 °C, respectively (Table 6). The starches isolated differed significantly ($p < 0.05$) for swelling power. Previous studies showed that the swelling power of acorn starches varied from 13 to 14 g/g at 90 °C [49]. SP values are low to other starch sources (such as sweet potato starch (35-40.41 g/g) [50], but comparable to chestnut starches (13.6-17.3 g/g) [51]. The limited swelling of the starch (12.5-19.2 g/g) may be ascribed to the presence of high levels of cross-linking [49]. Furthermore, [52] attributed the limited swelling to C-type starches, typical of legume starches. Furthermore, increased amounts of phenolic compounds may contribute to reduced swelling power. According to Li et al. 2017, these compounds may possibly interrelate with the structure of starch, resulting in a rise in gelatinization temperature and, as a result, a

reduction in swelling power[53]. The solubility index of starches increased remarkably with the rising temperature from 60 to 90 °C, which could be attributed to more leaching of amylose with the elevation in the temperature. The solubility of acorn starches ranged from 4.8 to 7.6% at 90°C for (QF) and (QR), respectively. Solubility differed significantly between the starches ($P \leq 0.05$). The solubility of acorn starches was comparable to starch extracted from acorns of *Q. ilex* subsp. ballota (4-14%) [16] and Quercus fabri Hance (4.8-15.22%) [54] but higher than those of *Q. rotundifolia* and *Q. suber* (1.3–1.5% at 90 °C) [49]. In general, the variations in solubility and swelling power are mainly attributed to the presence of non-starch compounds (e.g., lipids and ash), starch isolation methods, amylose content, and viscosity patterns [20].

3.7.2. X-ray diffraction.

The starch X-ray diffraction pattern was C type (Figure 1), which is a combination of A- and B-type unit cells, with strong diffraction peaks at around 17.34°, 17.69°, 22.54° and 23.04° (2θ) and a few small peaks at around 15° 2θ[55]. The C-type X-ray diffraction pattern for the starch of acorns *Q. rotundifolia* and Q. suber was also reported by Correia et al. (2013). The relative crystallinity of acorn starches ranged from 28.03% to 30.88%, which were much more than that of *Q. glandulifera* Bl acorn starch from China (23.5%) [15] and lower than those of *Q. rotundifolia* and *Q. suber* acorns starches (43.1–46.6%)[49]. Generally, the variation in the degree of crystallinity and diffraction pattern of starch granules was mostly impacted by genotype, agronomic, and cultivation conditions [11].

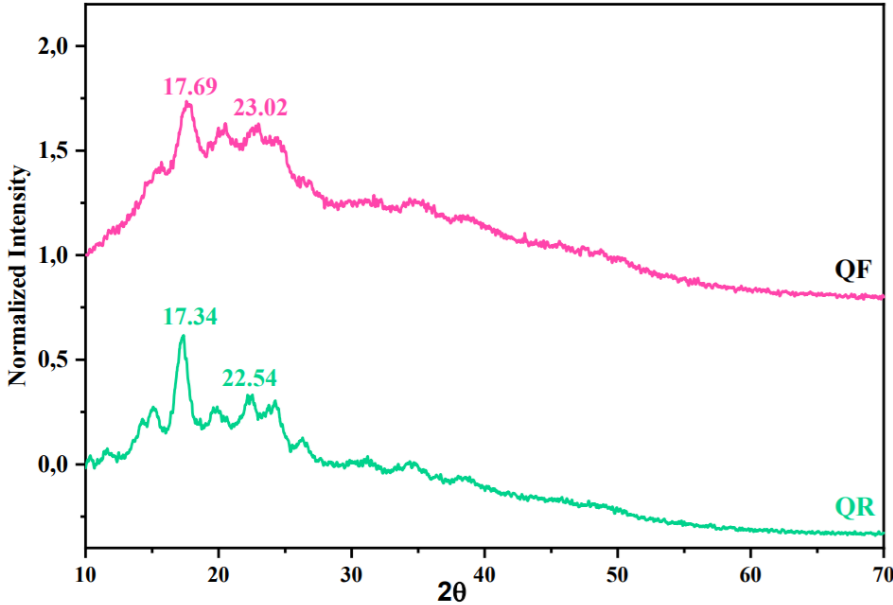

Figure 1. X-ray diffraction patterns of the isolated acorn starches.

3.7.3. Morphological characteristics.

The morphology of acorn starch granules is displayed in Figure 2. The surface of starch granules is smooth without scratches, and the presence of holes has not been observed. The granules of acorn starches (QF) ranged from 3.1 to 14.4 m and had ovoid, round, triangular, and hemispherical shapes. Acorn starches (Q.R) had a round, elongated and ovoid shape, with dimensions ranging from 3.7 to 15.5 μm. The size of acorn starch granules is larger than rice, oat, and quinoa starches and lower than wheat, maize, potato, and barely starches [56]. It has been described that the size of acorn starch granules of different species varied from 2.5 to

126.2 µm [11]. The observed difference may be related to botanical origin as well as changes in starch extraction methodology and size measurement techniques. According to the literature, granule size and shape are strongly related to physicochemical characteristics, such as crystallinity, solubility, and swelling power [11].

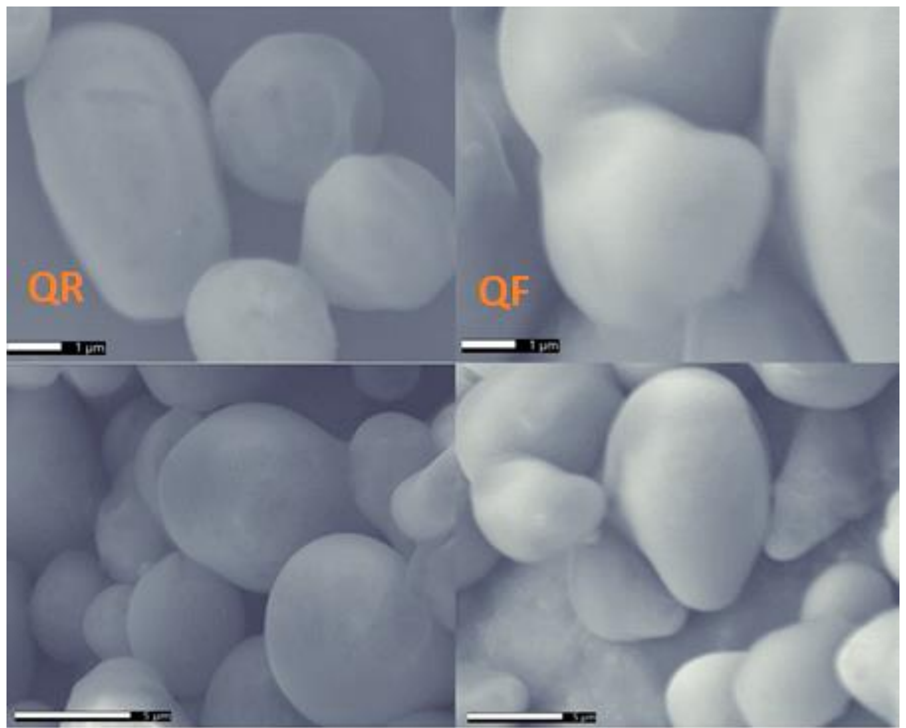

Figure 2. SEM images of isolated acorn starches.

4. Conclusions

The composition and nutritional value of Moroccan *Quercus acorns* were investigated. Results showed that acorn oils were generally characterized as high oleic acid and also linoleic acid. In addition, β-sitosterol was the main phytosterol that was presented in acorn oils (88.95-91.9%). The results also suggest that acorn flours are excellent sources of essential minerals (such as P, Ca, Mg, Mn, Fe, and Zn), chlorophylls, beta-carotene, and carbohydrates. Furthermore, acorns possessed rich sources of potentially functional components (such as flavonoids and phenolics) and showed higher antioxidant capacity. Additionally, acorn starches' physicochemical and functional properties are very interesting, indicating that acorn starch is adequate for some specific applications. Therefore, these findings provide scientific proof of the presence of functional components in acorn flours, indicating that these Moroccan acorns could be used in food, nutraceuticals, and pharmaceutical industries.

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Conflicts of Interest

The authors declare that they have no conflict of interest.

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