

TRADITIONAL ALGERIAN FERMENTED FOOD: FIRST DATA ON NUTRITIONAL CHARACTERISTICS OF WHEAT (*TRITICUM DURUM*) FERMENTED IN UNDERGROUND SILOS MATMOR (MASCARA, ALGERIA) COMPARED TO UNFERMENTED WHEAT

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Abstract. Wheat grain is a staple food that contains numerous compounds beneficial to nutrition and health. Fermentation is of importance to improving the nutritional attributes of cereal grains for human consumption. With the aim of identifying the influence of the fermentation by the traditional storage "Matmora" during 9 years, on the biochemical components of the wheat, we tried to make a comparative study between the properties of both samples of *durum* wheat fermented wheat "El-Hammoum", unfermented wheat and to estimate their nutritional values and to find that the fermented durum wheat possesses antihyperglycemic activity and a less allergenic effect and which would be recommended for the people having an allergy and/or intolerance of the gluten.

Fermented wheat "El-Hammoum", an Algerian food contains a considerable amount of water, proteins, is a good source of lipids, fiber, mineral content and polyphenols, and low content of pH, test Weight, Falling Number, and gluten. Moreover, the results of the electrophoresis on SDS-PAGE gel, showed a total degradation of proteins gliadins and gluténins after sequential extraction of every protein. The decrease in gliadin, gluten protein fraction, implicated in celiac disease, would be beneficial for these patients. Also good for health; it is used as a remedy by diabetics because showed good in vitro antihyperglycemic activity; because has low contents of total sugars and starch linked to its fermentation. These findings also provide evidence that "El-Hammoum" may be useful for the treatment of complications associated with Diabetes and raise the possibility of a new application as a complementary therapy associated with hypoglycaemic drugs.

In conclusion, fermentation of cereals in underground silos "Matmora" can be a way to produce food grains enriched with health promoting compounds and enhanced functional attributes.

Keywords: Fermented wheat "el hammoum", unfermented wheat, traditional storage, nutritional analysis, SDS PAGE, antihyperglycemic activity.

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1. Introduction

Fermentation of grain is a natural, biological processing technique and traditional method that can be used to improve the nutritional, functional, and sensory properties of cereals grains along with increased micronutrients (Hefni & Witthoft, 2011). For the increase of grains nutritive value, germination for several days or fermentation is necessary (Rakcejeva *et al.*, 2014). The minimal environmental conditions needed for

grain to fermentation are optimum humidity, availability of oxygen, an adequate temperature, and time for the different metabolic processes (Sangronis & Machado, 2007). The fermentation of grains realizes a significant growth of nutritional value by increasing the bioavailability of nutritional compounds, vitamins, bio-elements, and other biologically active substances due to the partial hydrolysis of starch, proteins, hemicelluloses, and even celluloses. Fermentation triggers the enzymatic activity in grain, and dormant hydrolytic enzymes are activated, breaking down starch, fibers, and proteins and leading to an increase in the number of digestible compounds along with improvement of functional properties without any chemical modification. During the fermentation endohydrolase enzymes (α and β -amylases), proteolytic enzymes, and catalyze are activated in cereal grains, which enhances the starch degradation, a complex biochemical process and thus lead to the conversion of insoluble granules to soluble starch and dextrin (El Hag *et al.*, 2002; Dirar, 1992; Karovicova & Kohajdova, 2007).

In Algeria, the storage of durum wheat (*Triticum durum*) performs in underground silos "Matmor" in some rural areas. Will generate a spontaneous fermentation of wheat grains stored in contact with the wall "Matmora", this fermentation can last 2-10 years. To give birth to a new stable fermented product called "El-Hammoum" which comes from "Hmoum" meaning black referring to the black color of fermented wheat is appreciated for its properties, organoleptic, nutritional, and healthy.

Several authors reported that the spontaneous fermentation of starch products, such as cereals, is of lactic or acetic acid type (Viéra-Dalodé *et al.*, 2007). "El-Hammoum" became a local product, especially in rural areas where it is consumed in the form of couscous, an Algerian food that was considered a food with medicinal properties in the prevention and treatment of many intestinal pathological and physiological complications. Functional properties of fermented wheat "El-Hammoum" proteins make its flour suitable to be used as a supplement to replace toxic protein sources (e.g. unfermented wheat). Hence, fermented wheat "El-Hammoum" flour can substitute gluten-rich cereals in the diet of people suffering from celiac disease.

In Algeria, an ethnopharmacological survey showed a particularly interesting use of "El-Hammoum" by diabetics. Limited information is available regarding the chemical, physical, and functional properties of fermented wheat "El-Hammoum" fermented during the traditional storage "Matmora"

The objectives of this research were to identify the fermented wheat "El-Hammoum" and the characterization of its physicochemical, nutritional quality, functional properties, and health benefits.

2. Material and methods

Plant material

The fermented wheat "El-Hammoum" used in this study, was obtained from July 2008 harvest and was stored until May 2017 in a "Matmora" located in the region of Feraguig (Mascara, Algeria) with latitude 35.5331, 35° 31′ 59″ North, and longitude 0.155139, 0° 9′ 19″ East (Figure 1).; hermetically sealed at the opening with clay in order to isolate the content from outside air and pests, and whose walls are lined with wheat straw. The fermented wheat "El-Hammoum" (FWH) was retrieved from the walls, unfermented wheat (UFW), in the center, served as a control sample (Figure 2).

All samples were transferred to the laboratory under refrigeration and stored at 4°C until their analysis.

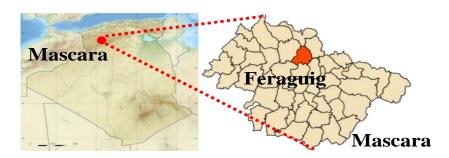


Fig 1. Map of the origin of "Matmora" "El-Hammoum" a region of Feraguig (Mascara, Algeria)



Fig 2. Picture of unfermented wheat (E) fermented wheat (D), the opening of the "Matmora"(C) and sampling area (A, B) (Original picture)

Biochemical Analysis

1. Insoluble fiber by the method Weende (%) NF V 03-040.

2. Total Proteins were evaluated by the Kjeldahl method (NF V 03-050) from the total nitrogen content multiplied by the conversion factor of 5.7.

3. Total lipids were obtained by extraction with hexane at 140°C (NF 03-713), using an extractor (SOXLHET). The missile phase (solvent and lipid) was isolated, the solvent was evaporated, and the lipid fraction was recovered too, be weighed.

Physicochemical Analysis

1. Humidity was carried out in a drying-room at a temperature between 130 and 133°C Until a constant mass was obtained (NF V 03-707).

2. Ashes were mineralized at 900°C until complete combustion of organic matter (NFV03-720).

3. The moisture content (%) (ISO 712)

4. PH measurement was performed using a pH-meter.

5. The color was measured using a spectrocolorimeter (CHROMA METRE CR-410 MINOLTA).

Technological composition

1. Wet gluten content, dry gluten content was determined using Glutomatic Perten Instruments (Perten Instruments AB, Sweden) according to (%) ISO 21415-2

2. The falling number or full-time Hagberg was determined according to (NF ISO 3093) using Falling Number 1500 (Perten Instruments).

3. Test weight (TW), determined using the Nilema-litre, according to standard (NF V 03-719).

Minerals Analysis

Mineral composition (Fe) and (Zn) were determined using atomic emission spectroscopy (Varian 220), potassium (K+) was measured using a flame spectrophotometer. According to the described method (Pinta, 1971).

Protein characterization using SDS-PAGE for the fermented and unfermented wheat

Using the sequential extraction procedure of protein of whole grain flour, following combined protocols proposed by (Laemmli, 1970) modified by (Payne *et al.* 1980; Singh, 1991). Electrophoresis was performed using SDS-PAGE (sodium dodecyl sulfate-polyacrylamide gel electrophoresis) following (Payne et al., 1980; Singh, 1991). Protein markers of different molecular weights from a Sigma-Aldrich kit (MM: 26.60 and 180.00 Da).

Phytochemical characterization Preparation of methanolic extracts

Wheat seed extracted with 80% methanol and filtered after 48 hours. The residue was re-extracted with the addition of 80% methanol, and after 24 h, it was filtered again. Combined filtrates were concentrated on a rotary evaporator at 45°C for methanol elimination, and the extracts were kept under refrigerated conditions until use (Romani *et al.*, 2006).

Determination of total phenolic, flavonoid concentration

Total phenolics in crude extract of fermented wheat "El-Hammoum", was determined by the folin–ciocalteu method (Singleton *et al.*, 1999) using gallic acid as standard. The total flavonoid content in the extract was determined spectrophotometrically based on flavonoid aluminum complication (Topçu *et al.*, 2007).

In vitro antihyperglycemic activity (a-Glucosidase Inhibition)

 α - glucosidase enzyme inhibition assays were carried out on 96-well microplates following the method described by (Zhang *et al.*, 2014) with minor modifications. Briefly, the reaction mixture contains α -glucosidase (15 µL, 1.0 unit/mL in 100 ml phosphate buffer, pH 6.8) mixed with different concentrations of the extract of "El-Hammoum" (2000 µg – 31.25 µg) and 40 µl of phosphate buffer in a 96-well plate. After incubation at 37 °C for 10 min, 45 µL of p-NPG solution (2ml in 0.1 ml phosphate buffer, pH 6.8) was added to start the reaction. Acarbose was used as standard. Enzymatic activity was monitored by measuring the p-nitrophenol released from PNPglycoside at 405 nm every 45 sec for 3 min. Results were expressed as a concentration of samples resulting in 50% inhibition of enzyme activity (IC50). The inhibition percentage was calculated according to the following formula:

%Inhibition = [A control – A sample]/A sample x 100

Data analysis

All results were reported as mean, and SD Statistical significance was determined by one-way analysis of variance (ANOVA) using R stat. Significant differences between means were determined by the Turkey method, and differences at the level P <0.05were considered being significant. The concentration giving 50% inhibition (IC50) was calculated with Microsoft excel 2013.

3. Results and discussion

The morphological aspect of fermented wheat

The very pronounced mold smell that came from the fermented wheat "El-Hammoum" sample can be explained by the production of volatile and aromatic compounds during fermentation. The brown color observed (Figure 3) can be explained by the non-enzymatic browning, as has been observed in other studies on food fermentation (Faithong & Benjakul, 2012). A particular taste acid was also perceived.



Fig 3. Picture of fermented and unfermented wheat (Original picture)

Biochemical analysis

Proteins

Total proteins are often hydrolyzed in polypeptides and amino acids. They are used in microbial metabolism during the fermentation (Karovičová & Kohajdova, 2007). This explains the low value obtained in our sample of fermented wheat (12, 76±0,05%), compared with unfermented wheat (15,4±0,1%). Protein content decreased (Table 1, Figure 4) very highly significantly during fermentation (P< 0, 001). This might be due to that part of the protein was utilized for lactic acid bacteria growth and development.

The fermented wheat is within the standards (10 à 15%) for optimal use (Jeantet *et al.*, 2007). The fermentation of grains realizes a significant growth of nutritional value by increasing the bioavailability of nutritional compounds, vitamins, bio-elements, and other biologically active substances due to the partial hydrolysis of starch, proteins, hemicelluloses, and even celluloses (Karovičová & Kohajdova, 2007; Bilgiçli & Ibanoğlu, 2007).

Fat content

The value of total lipid in the fermented wheat "El-Hammoum" is $(1,37\pm0,07\%)$. It lies within the range of values (1-3%) obtained by (Godon, 1991; Kouakou *et al.*, 2013). The lipid content in our sample was higher than that observed in the unfermented wheat $(1,08\pm0,05\%)$. But no significant difference between fermented wheat "El-Hammoum" and unfermented wheat has been observed. Manifestly, fermented wheat is a good source of lipids, are excellent sources of energy (Table 1, Figure 4).

In flour from the wheat, some lipids are associated with starch granules (1.0%), but others are not (1.4%). Starch lipids are made up of 9% non-polar lipids, 5%

glycolipids, and 86% phospholipids (Delcour *et al.*, 2010). Starch lipids are contained within starch granules as inclusion complexes and are located between amylase and monoacyl lipids, such as lysophosphatidylcholines (Feillet, 2000).

After fermentation starch, was degraded to soluble sugars, and the; lipids associated with starch granules is degraded to free fatty acids and triglycerides whith activity of lactic acid bacteria and amylolytic bacteria and lipase activity (Collins *et al.*, 2003; Feillet, 2000); which could explain the increase in fat content in fermented wheat compared to unfermented wheat. This hydrolysis is accelerated by an increase in temperature and humidity (Bothast, 1978; Feillet, 2000; Rajakumari *et al.*, 2008).

Cellulose content

The fermented wheat "El Hammoum" cellulose is $(0,10 \pm 0,07 \%)$ low compared to that of unfermented wheat $(0,23 \pm 0,0003\%)$ % with significant difference (p<0,05). These differences may be linked to fermentation. Many authors observed a decrease in fiber content which they attributed to enzymatic solubilization of part of the seed fiber during seed fermentation (Brou *et al.*, 2013). Fermentation triggers the enzymatic activity in grain, and dormant hydrolytic enzymes are activated, breaking down starch, fibers, hemicelluloses, and even celluloses and proteins and leading to an increase in the number of digestible compounds along with improvement of functional properties without any chemical modification (Karovicova & Kohajdova, 2007) (Table 1, Figure 4).

Physicochemical Analysis

Moisture Content

The fermented wheat taken directly from the "Matmora" had a moisture content of 40%. For our sample, the fermented wheat was dried, and its; moisture was (14,96±0,14%), these result is high than unfermented wheat (12,26±0,05%) with very highly significant difference (p<0,001); but It is within the standards for optimal use (FAO, 1996; Fredot, 2006). These results are also high than those reported by (Bekhouche *et al.*, 2013) 12,99%, but this value is similar to that found by (Dubois, 1996) \leq 16%. According to Bartali (Bartali Afie & Persoons, 1989), fermented wheat that is near the walls of the Matmora has high water content (Table 1, Figure 5).

Ash content

The ash content recorded in the fermented wheat "El-Hammoum" is low $(1,66\pm 0,01\%)$ with a very highly significant difference than (P< 0. 001); that of unfermented wheat $(2,32\pm 0,03\%)$, (table 01, and figure 05) but comparable to the value ($\leq 2.1\%$) of Codex Alimentarius (FAO, 1996). similar findings have been reported in other studies (Bekhouche *et al.*, 2013).

Fermentation enhances hydrolytic enzyme activity and modifies the grain structure and components (Traore *et al.*, 2004). Reduction in mineral substances could be due to interaction of minerals with environmental conditions (increase humidity, increase in CO_2 , an increase in temperature and a long time for storage 9 years, and with the different metabolic processes during the storage in "Matmora". Miranda et al., (2010) indicate that the reduction in mineral substances results in the diffusion of these micronutrients into intercellular spaces, especially at high temperatures.

PH and Titratable acidity

The fermented wheat "El-Hammoum" has a pH lower $(5,23\pm0,005\%)$ compared to unfermented wheat (6, 31±0, 02 %) with a very highly significant difference (P<0.001). Acidity in unfermented wheat showed a load normally 0, 16±0,02%. However, in the fermented wheat "El Hammoum" acidity increases during the fermentation to reach a maximum value $(0,39\pm0,01\%)$ so, fermented wheat "El Hammoum" is more acidic than unfermented wheat, with a very highly significant difference (P<0.001).

Acidity may increase during the fermentation time due to the liberation of organic acids such as lactic acid or acetic acid from microbial metabolism reactions (Faid *et al.*, 1994). The lactic acid bacteria consume sugar and carbohydrates and convert it into lactic acid which lowers the pH (Mokhtari *et al.*, 2016). It has been well documented that high acidification rates are usually accompanied by fast and high growth rates of lactic acid bacteria in fermented wheat during the fermentation (Faid *et al.*, 1994; Guyot, 2010) (Table 1, Figure 5).

Technological Analysis

Gluten

The level of gluten present in our sample fermented wheat "El-Hammoum" is hydrolysis; this may partly explain the depolymerization of gluten witch, is the cause of the non-formation of a paste during gluten content determination. The modification of gluten in the fermented wheat "El Hammoum" would be linked to a very highly significant (P<0.001) degradation with could be explained by the hydrolysis of gluten by microbial enzymes or amylases present in grain during fermentation (Bauer *et al.*, 2010, Bornet, 1993).

Fermentation induces the synthesis of hydrolytic enzymes, and during fermentation protein, and starch was degraded to soluble sugars and amino acids, respectively (Thiele *et al.*, 2004; Schober *et al.*, 2002). Similar results were reported in germinated products (Shastry and John, 1991). These results should be confirmed by other analyses. This could be useful to consider the protein characterization (glutenin and gliadin) of fermented wheat "El Hammoum" by electrophoresis on SDS-PAGE Gel (Table 1, Figure 6).

Wheat Falling Number Test weight (TW)

The fermented wheat has a low falling number $(151,5\pm0,07s)$ with a very highly significant difference (P< 0,001), which means a very high amylase activity (Topping, 2007). However, excessive presence of α -amylase causes a decrease of starch content in the fermented wheat "El Hammoum" therefore a decrease in total sugar content. It could result from starch hydrolysis to monosaccharide and soluble sugars, which were used as an energy source during grain fermentation and lactic acid bacteria growth. The growth of lactic acid bacteria results from the production of organic acids (World Health Organization) which resulted in the decrease of the pH (Kohajdova & Karovicova, 2007) at a (5,23\pm0,005%) observed in the fermented wheat "El Hammoum".

The modification of starch in the fermented wheat "El Hammoum" results in a low glycemic load after "Hammoum" consumption, hence the interest of diabetics for this local product. In addition, high amylase activity associated with the degradation of gluten affects the rheological characteristics of pasta, they have as a consequence the deterioration of breadmaking quality, and this explains the Hammoum consumption only in the form of couscous.

Test weight (TW)

Also, the test weight of fermented wheat "El Hammoum" have undergone reductions of $(67,31\pm0,013 \text{ (kg/hl)})$ with very highly significant difference (P< 0,001), than those of the unfermented wheat $(76,95\pm0,089(\text{kg/hl}))$. These results are in accordance with (Tester & Morrison, 1990). This decrease is due partly to the loss of dry matter and on the other hand the increase of the water content (Dexter & Edwards, 1998) (Table 1, Figure 6).

Minerals Analysis

Minerals content: iron (Fe), potassium (K), and zinc (Zn), were estimated in fermented Wheat "El-Hammoum" and unfermented wheat the finding is reported in the same table 1 figure 07. The results illustrated that the iron (Fe) $(98,33\pm0,57\%)$ and zinc (Zn) $(42,33\pm0,57\%)$, is higher in fermented wheat "El-Hammoum", than potassium (K) $(3,31\pm0,21\%)$.

Unfermented wheat also has high results in potassium (K) $(3,84\pm0,29\%)$. A very highly significant difference between in fermented wheat "El-Hammoum" and unfermented wheat has been observed for the iron (Fe), and zinc (Zn) (p<0.001) and significant difference between fermented wheat "El-Hammoum" and unfermented wheat has been observed for the potassium (K) (p<0.05).

Previous works have described the mineral content of unfermented wheat (AOAC, 1990). However, to our knowledge, this is the first report of mineral contents of fermented wheat "El-Hammoum". This difference between the two can be explained by many factors such as milling conditions and the quantity of existing (Arzani & Ashraf, 2017). The evidence reports that minerals and trace elements play an important role in the regulation of certain body functions. Whereas most microelements (Fe and Zn) play a vital role as a structural part in many enzymes. Also, microelements have key roles in the formation of the erythrocyte cells (Fe), and macro minerals such as K have a high potential to control blood pressure. The minerals are also involved in immune (Zn) systems (Arzani & Ashraf, 2017).

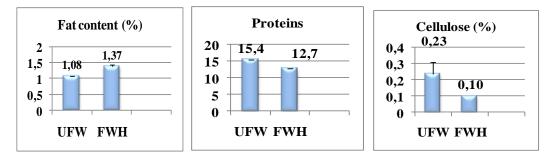


Fig. 4. Biochemical analysis of fermented wheat El-Hammoum and unfermented wheat

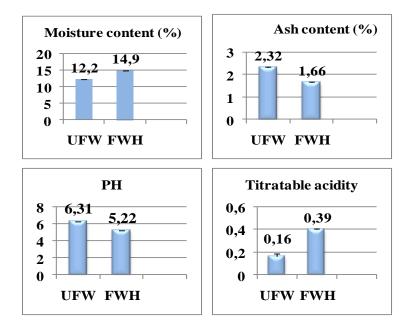


Fig. 5. Physicochemical analysis of fermented wheat "El-Hammoum" and unfermented wheat

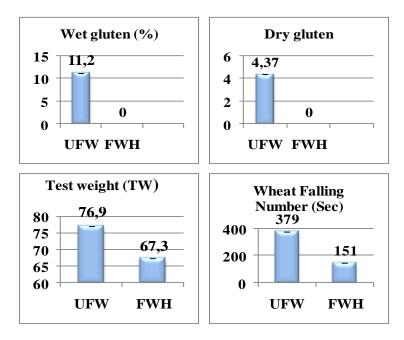


Fig. 6. Technological analysis of fermented wheat "El-Hammoum" and unfermented wheat

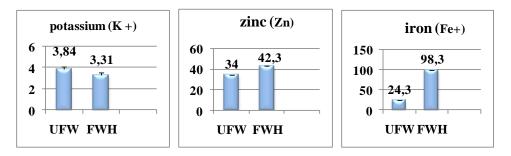


Fig 7. Minerals analysis of fermented wheat "El-Hammoum" and unfermented wheat

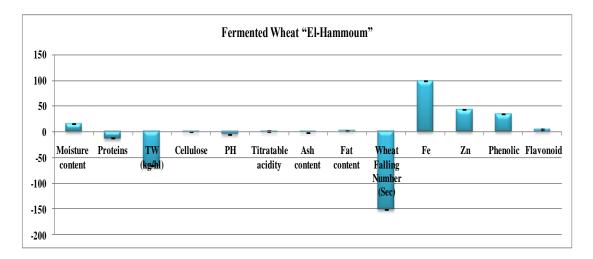


Fig 8. Variations of different parameters of fermented wheat "El-Hammoum"

Table 1. Proximate composition of fermented wheat "El-Hammoum" and unfermented wheat

Parameters	Unfermented Wheat Average ± SD	Fermented Wheat Average ± SD	P-Value
Moisture content (%)	12,26±0,05	14,96±0,14 ***	< 0.001
Ash content (%)	$2,32 \pm 0,03$	1,66± 0,01 ***	0.002
РН	6,31±0,02	5,23±0,005 ***	< 0.001
Wheat Falling Number (Sec)	379±1,41	151,5±0,07***	< 0.001
Wet gluten (%)	11,26±0,04	-	< 0.001
Dry gluten	4,37±0,04	-	< 0.001
TW (kg/hl)	76,95±0,089	67,31±0,013 ***	< 0.001
Proteins (%)	$15,4\pm 0,1$	12,76±0,05***	< 0.001
Fat content (%)	1,08±0,05	$1,37\pm0,07$	0.05
Titratable acidity	0,16±0,02	0,39±0,01 ***	< 0.001
Cellulose (%)	0,23±0,0003	$0,10{\pm}0,07$ *	0.03
Fe	42±0,00	98,33±0,57 ^{***}	< 0.001
K+	3,84±0,29	$3,31\pm0,21^*$	0.01
Zn	34±0,00	42, 33 ±0, 57 ***	< 0.001
CIE L [*]	$61,7 \pm 0,33$	$50,01\pm1,40^{***}$	0.002
CIE a [*]	0,85±0,042	$1,45\pm0,06^{***}$	< 0.001
CIE b [*]	0,76±0,02	20,21±0,15 ***	< 0.001
Phenolic	18,42±0,02	33,57±0,032***	< 0.001
flavonoid	2,42±0,002	3,9±0,012***	< 0.001

The observed data express the values of the mean \pm SD (n = 3). CIE Commission International l'Eclairage. L* (lightness), a* (redness), and b* (yellowness), Phenolic: (µg EAG/ml), flavonoid: (µg QE/ml). P< 0. 001 Very highly significant difference, p< 0.01 highly significant difference, p<0.05 Significant difference

Protein characterization using SDS-PAGE for the fermented and unfermented wheat

The results of electrophoresis on SDS-PAGE Gel it's revealed a hydrolysis total of glutenin and gliadin fractions of fermented wheat "El-Hammoum". SDS-PAGE electrophoresis of fermented wheat show polypeptide sequences of gliadins and

glutenins (electrophoretic bands) quite distinct and different from that of unfermented wheat compared to the control (Commercial peptide kit).

Fermentation induces the synthesis of hydrolytic enzymes (Thiele *et al.*, 2004; Schober *et al.*, 2002). The result could be explained by the partial hydrolysis of gluten by microbial enzymes or amylases present in grain during fermentation (Bauer *et al.*, 2010). Similar results were reported in germinated products (Shastry & John, 1991). Our results showed that the fermented wheat "El-Hammoum" is considered as gluten-free foods. These fermentation products give new therapeutic hopes for celiac disease. These results should be confirmed by other studies, like allergenicity, Antigénicité (Figure 9, 10).

The content of the various parameters studied for fermented wheat "El Hammoum" are within the limits of the values published by many authors, only the gluten content obtained seemed to be affected; this may be due to the long-term storage of wheat in the "Matmora".

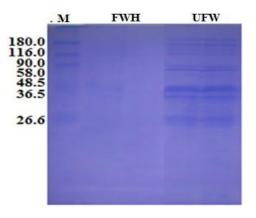


Fig. 9. Electrophoresis diagrams of glutenin

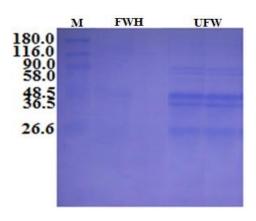


Fig 10. Electrophoresis diagrams gel of gliadin

M: Kit protein α_2 Macroglobulin from equin serum180 kDa, B-Galactosidase from E.coli 116.00 kDa, Lactoferrin from human milk 90.00 kDa, Pyruvate Kinase from rabbit muscle 58.00 kDa, Fumarase from porcine heart 48.50 kDa, Lactic Dehydrogenase from rabbit muscle 36.50 kDa, Triosephosphate isomerase from rabbit muscle 26.60

Phytochemical characterization Preparation of methanolic extracts

Methanolic Extract of the unfermented wheat has a liquid appearance, yellow color with pleasant odors. But extract of the fermented wheat "El-Hammoum" has a dark color and a strong odor, with a vicious aspect.

Determination of total phenolic, flavonoid concentration

The results showed a very highly significant increase in the total polyphenol content and flavonoid in the fermented wheat compared to unfermented, $(33,57\pm0,032 \mu g EAG/ml)$ $(3,9\pm0,012\mu g QE/ml)$, respectively, followed by unfermented wheat $(18,42\pm0,02\mu g EAG/ml)$ $(2,42\pm0,002 \mu g QE/ml)$ respectively. Similar results were reported in fermentation products (Jung *et al.*, 2010) and germinated products (Ibrahim et al.; 2000; Oloyo, 2004). This increase in polyphenol content during fermentation is possible only with certain *Lactobacillus strains* (Eom *et al.*, 2011) on the one hand and on the other hand, it is due to the presence of certain types of phenolic compounds, such as isoflavones, particularly the aglycone subgroup, which increase significantly during the lactic acid fermentation (Hubert *et al.*, 2008).

This explains our results because the fermented wheat underwent lactic fermentation firstly and secondly, what is known to contain aglycones in its polyphenol profile (McCallum, 1989). The changes in the flavonoid content during fermentation could be explained by various types of key enzymes or cofactors being synthesized, which leads to the production of the flavonoids. The fermentation is induced by rehydration of the seed hang the storage in Matmour, which increases both respiration and metabolic activity thus allowing the mobilization of primary and secondary metabolites (Dykes & Rooney, 2007) Moreover, the richness of El-Hammoum in polyphenols gives it positive effects on health; protection against cardiovascular disease, type II diabetes, obesity, anticarcinogenic action, fight against cellular aging (Serpen *et al.*, 2008; Duthie & Brown, 1994) (Table 1, Figure 11 and 12).

In vitro antihyperglycemic activity Alpha -Glucosidase inhibitory activities

In a rural environment, the traditional use of some foods or medicinal plants to treat diseases is still used today. In Algeria, an ethnopharmacological survey showed a particularly interesting use of fermented wheat "El Hammoum" by diabetics. In this study, we tested the ability of fermented wheat "El Hammoum", to inhibit α -glucosidase activity in-vitro compared with fermented wheat and that of Acarbose used as a standard. Fermented wheat "El Hammoum" exhibited strong very highly significant inhibitory activity against a-glucosidase with (IC50: 383,18 ±0,93 µg/ml), which is comparable with the unfermented wheat (IC50: 1469,13±0,85 µg/ml). These results were low compared to the standard Acarbose (IC50: 275,43± 1,59 µg/ ml).

Excessive presence of α -amylase causes a decrease of starch content in the fermented wheat, therefore decrease in total sugar content (Chandrashekar & Desikachar, 1984; Di Cagno *et al.*, 2013), results in a low glycemic load after El-Hammoum consumption, hence the interest of diabetics for this local product. Polyphenolic compounds of wheat may interfere with the activity of digestive enzymes in the brush border of the small intestine, could slow the liberation of D-glucose from oligosaccharide and disaccharides, resulting in delayed glucose absorption and decreasing postprandial glucose levels (Zhao *et al.*, 2014) (Figure 13 and Table 2).

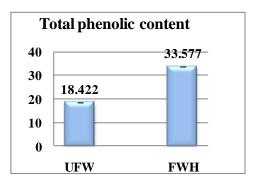


Fig 11. Total phenolic compound content (µg EAG/ml) of fermented wheat "El-Hammoum" and unfermented wheat

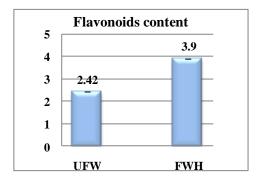


Fig 12. Total flavonoids compound content (μ g QE/ml) of fermented wheat "El-Hammoum" and unfermented wheat

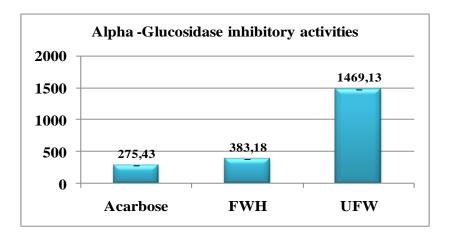


Fig 13. Alpha -Glucosidase inhibitory activities of fermented wheat "El-Hammoum" and unfermented wheat. IC50 values expressed in μg/ml of the samples studied sort in ascending order

Compound	Alpha glucosidase inhibitory activities (IC50 μg/ml) Average ± SD
FWH	383,18 ±0,93 ***
UFW	1469,13±0,85
Acarbose	275,43±1,59

Table 2. Alpha -Glucosidase inhibitory activities of fermented wheat "El-Hammoum" and unfermented wheat

* IC50 values expressed are means \pm SD of three parallel measurements. P< 0, 001 very highly significant difference

4. Conclusions

The fermentation of wheat in the "Matmora" tends to disappear due to the settlement of rural populations in urban areas. According to the present results, it can be concluded that significant variations found between fermented wheat "El-Hammoum" and unfermented wheat for the different components studied assert the influence of the fermentation to stored wheat in underground pits "Matmora". A wide range of these compounds is affected by fermentation, such as gluten is degraded, others, like antioxidants and total phenolics, are increased. This new finding will beneficial and could be a good alternative to people having an allergy or intolerance the gluten, but these results should be confirmed by other studies like allergenicity, antigénicité. Also "El-Hammoum" would be a very interesting perspective on the diet for diabetics because showed well in vitro antihyperglycemic activity.

Fermented wheat flour can also be used as a functional agent in fabricated foods such as pasta, couscous goods in the future food processing industry.

In conclusion, the natural fermentation in underground silos "Matmora" can improve the functional properties of stocked grains, and it would be possible to design new food and to produce a natural healthy product.

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References

- AOAC. (1990). Official Methods of Analysis: Changes in Official Methods of Analysis Made at the Annual Meeting. Supplement 15.
- Arzani, A, Ashraf, M. (2017). Cultivated ancient wheats (Triticum spp.): a potential source of health-beneficial food products. *Comprehensive Reviews in Food Science and Food Safety*, 16(3), 477-488.
- Bartali, E.H., Afie, S., Persoons, E. (1989). Stockage des céréales dans des entrepôts souterrains." Céréales en régions chaudes: conservation et transformation. Aupelf J. Libbey Eurotext, 27-38.
- Bauer, W.J., Badoud, R., Löliger, J., Etournaud, A. (2010). Science et technologie des aliments: Principes de chimie des constituants et de technologie des procédés (éd. 1ère). Italie: PPUR Presses Polytechniques.

- Bekhouche, F., Merabti, R., Bailly, J.D. (2013). "Lemzeiet": Traditional couscous manufacture from fermented wheat (Algeria): process and technological and nutritional quality. *African Journal of Food Science and Technology*, *4*(8), 167-175.
- Bilgiçli, N., İbanoğlu, Ş. (2007). Effect of wheat germ and wheat bran on the fermentation activity, phytic acid content and colour of tarhana, a wheat flour–yoghurt mixture. *Journal of Food Engineering*, 78(2), 681-686.
- Bothast, R.J. (1978). Fungal deterioration and related phenomena in cereals, legumes and oilseeds. *Post-harvest Biology and Botechnology*, 210-243.
- Brou, K., N'da-Kouassi, A., Kouadio, J., Guehi, T., N'guess, K., Gnakri, D. (2013). Biochemical characterization and functional properties of weaning food made from cereals (millet, maize) and legumes (beans, soybeans). J. Food Chem. Nutr., 1, 22-32.
- Chandrashekar, A., Desikachar, H.S. (1984). Studies on the hydration of starches, flour and semolina from different cereal grains. *Journal of Food Science and Technology*, 21(1), 12-14.
- Cilla, I., Martínez, L., Beltrán, J.A., Roncalés, P. (2006). Dry-cured ham quality and acceptability as affected by the preservation system used for retail sale. *Meat Science*, 73(4), 581-589.
- Collins, Y.F., Mcsweeney, P.L., Wilkinson, M.G. (2003). Lipolysis and free fatty acid catabolism in cheese: a review of current knowledge. *International Dairy Journal*, 13(11), 841-866.
- Delcour, J.A., Hoseney, R.C. (2010). Principles of Cereal Science and Technology. 3rd ed. St Paul, MN, USA: American Association of Cereal Chemists.
- Dexter, J.E., Edwards, N.M. (1998). Incidence des facteurs de classement fréquemment détectés sur l'aptitude technologique du blé dur. Association of Operative Millers-Bulletin Commission canadienne des grains, Laboratoire de recherches sur les grains, Winnipeg (Manitoba) R3C 3G8 Contribution, (M231), 35.
- Di Cagno, R., Coda, R., De Angelis, M., Gobbetti, M. (2013). Exploitation of vegetables and fruits through lactic acid fermentation. *Food Microbiology*, *33*(1), 1-10.
- Dirar, H.A. (1992). Sudan's fermented food heritage Board on Science and Technology for International Development, National Academy Press, Washington 27-34. In *Applications* of *Biotechnology to Traditional Fermented Foods* (pp. 27-34). Washington : Board on Science and Technology for International Development, National Academy Press.
- Dubois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.T., Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, 28(3), 350-356.
- Duthie, G.G., Brown, K.M. (1994). Reducing the risk of cardiovascular disease in Functional foods: Designer foods, Pharmafoods, Nutraceuticals, I. Goldberg, and Editor. Chapman & Hall: New York. NY, USA, 19-38.
- Dykes L, Rooney LW. 2007. Phenolic compounds in cereal grains and their health benefits . Cereal foods world , 52 (3): 105-111.
- El Hag, M.E., El Tinay, A.H., Yousif, N.E. (2002). Effect of fermentation and dehulling on starch, total polyphenols, phytic acid content and in vitro protein digestibility of pearl millet. *Food Chemistry*, 77, 193-196.
- Eom, S-H., Kang, Y.M., Park, J.H., Yu, D.U., Jeong, E.T., Lee, M.S., Kim, Y.M. (2011). Enhancement of polyphenol content and antioxidant activity of brown alga Eisenia bicyclis extract by microbial fermentation. *Fish Aquatic Sciences*, 14(3), 192-197.
- Faid, M., Boraam, F., Zyani, I., Larpent, J.P. (1994). Characterization of sourdough bread ferments made in the laboratory by traditional methods. *Zeitschrift für Lebensmittel-Untersuchung und Forschung*, 198(4), 287-291.
- FAO. (1996). Codex Alimentarius : 7-Céréales, légumes secs, légumineuses, produits dérivés et protéines végétales, FAO, 2ème édition, Rome, 164.
- Faithong, N., Benjakul, S. (2012). Changes in antioxidant activities and physicochemical properties of Kapi, a fermented shrimp paste, during fermentation. J. Foof Sciences, Technology.

- Fredot, E. (2006). Connaissance des aliments. Bases alimentaires et nutritionnelles de la diététique, Tec. Doc. Lavoisier, Paris, 397.
- Godon, B. (1991). Les constituants des céréales: nature, propriétés et teneurs, Biotransformation des produits céréaliers, I.N.R.A./A.P.R.I.A, Tec. Doc. Lavoisier, Paris: 679.
- Gourchala, F., Hobamahoro, A.F., Mihoub, F., Henchiri, C. (2014). Effect of natural fermentation on the nutritional quality of" El Hammoum" durum wheat (triticum durum) fermented product of the Algerian country. *International Journal of Bio-Technology and Research*, 4(4), 9-18.
- Guyot, J.P. (2010). Fermented Cereal Products. In *Fermented Foods and Beverages of the World*. Tamang Et Kailasapathy (Eds). Ed. Taylor & Francis Group, Floride, p: 247-261.
- Hefni, M., Witthöft, C.M. (2011). Increasing the folate content in Egyptian baladi bread using germinated wheat flour. *LWT Food Sci. Technol.*, 44, 706-712.
- Hubert, J., Berger, M., Nepveu, F., Paul, F., Dayde, J. (2008). Effects of fermentation on the phytochemical composition and antioxidant properties of soy germ. *Food Chemistry*, 109, 709-721.
- Ibrahim, S.S., Habiba, R.A., Shatta, A.A., Embaby, H.E. (2000). Effect of soaking, germination, cooking and fermentation on antinutritional factors in cowpeas. *Nahrung*, 46(2), 92-95.
- Jeantet, R., Croguennec, T., Schuck, P., Brule, G. (2007). Science des aliments (2 éme Edition ed.). Paris: Tec & Doc-Lavoisier.
- Jung, J., Shin, J.H., Yoo, S.K. (2010). Changes of Antioxidant Activity and Phenolic Compounds during Fermentation by Lactic Acid Bacteria. *FASEB Journal*, 830.2.
- Karovičová, Z.K.J., Kohajdova, J. (2007). Fermentation of cereals for specific purpose. *Journal* of Food and Nutrition Research, 46(2), 51-57.
- Kouakou, B., N'Da-Kouassi, A., M Halbin, K.J., Tagro, G., N'Guessan, K.F., & Gnakri, D. (2013). Biochemical characterization and functional properties of weaning food made from cereals (millet, maize) and legumes (beans, soybeans). *Journal of Food Chemistry* and Nutrition, 1(1), 22-32.
- Laemmli, U.K. (1970). Cleavage of structural proteins during the assembly of the head of bacteriophage T4 . *Nature*, 227(5259), 680.
- Martínez, L., Djenane, D., Cilla, I., Beltrán, J.A., Roncalés, P. (2005). Effect of different concentrations of carbon dioxide and low concentration of carbon monoxide on the shelf-life of fresh pork sausages packaged in modified atmosphere. *Meat Science*, *71*(3), 563-570.
- McCallum, J.A. (1989). Biochemistry of phenolic compounds in wheat grain (Triticum aestivum L.). Thesis in plant physiology. New Zealand: University of Canterbury.
- Miranda, M., Vega-Gálvez, A., López, J. (2010). Impact of airdrying temperature on nutritional properties, total phenolic content and antioxidant capacity of quinoa seeds (Chenopodium quinoa Willd.). *Ind Crop Prod.*, 3, 258–263.
- Mokhtari, S., Kheroua, O., Saidi, D. (2016). Isolation and Identification of Lactic Acid Bacteria from Algerian durum wheat (Triticum durum) natural fermented in underground silos Matmora" El-Hammoum" and their antimicrobial activity again pathogenic germs. *Journal of Nutrition and Health Sciences*, 3(4), 1-12.
- Oloyo, R.A. (2004). Chemical and nutritional quality changes in germinating seeds of Cajanus cajan L. *Food Chemistry*, 85(4), 497-502.
- Paredes Lopez, O., Mora-Escobedo, R. (1989). Germination of amaranth seeds: effects on nutrient composition and color. *Journal of Food Science*, 54(3), 761-762.
- Payne, P.I., Law, C.N., Mudd, E.E. (1980). Control by homoeologous group 1 chromosomes of the high-molecular-weight subunits of glutenin, a major protein of wheat endosperm. *Theoretical and Applied Genetics*, 58(3-4), 113-120.
- Pinta, N. (1971). Spectrométrie d'absorption atomique. Tome 2. Application à l'analyse chimique. Masson et Cie. Paris.
- Rajakumari, S., Grillitsch, K., Daum, G. (2008). Synthesis and turnover of non-polar lipids in yeast. *Progress in Lipid Research*, 47(3), 157-171.

- Rakcejeva, T., Zagorska, J., Zvezdina, E. (2014). Gassy Ozone Effect on Quality Parameters of Flaxes Made from Biologically Activated Whole Wheat Grains. *Int. J. Biol. Vet. Agric. Food Engg.*, 8, 378-81.
- Romani, A., Pinelli, P., Cantini, C., Cimato, A., Heimler, D. (2006). Characterization of Violetto di Toscana, a typical Italian variety of artichoke (Cynara scolymus L.). Food Chemistry, 95(2), 221-225.
- Sangronis, E., Machado, C. (2007). Influence of germination on the nutritional quality of Phaseolus vulgaris and Cajanus cajan. *LWT-Food Sci Technol.*, 40, 116-120.
- Schober, T.J., Clarke, C.I., Kuhn, M. (2002). Characterization of functional properties of gluten proteins in spelt cultivars using rheological and quality factor measurements. *Cereal Chemistry*, 79(3), 408-417.
- Serpen, A., Gökmen, V., Pellegrini, N., Fogliano, V. (2008). Direct measurement of the total antioxidant capacity of cereal products. *Journal Cereal Sciences*, 48, 816-820.
- Shastry, M., John, E. (1991). Biochemical change and in-vitro protein digestability of endospern of germinating Dolichos lablab. *J. Sci. Food Agri.*, 55, 529-538.
- Singh, S.P., Nodari, R., & Gepts, P. (1991). Genetic diversity in cultivated common bean: I. Allozymes. *Crop Science*, *31*(1), 19-23.
- Singleton, V.L., Orthofer, R., Lamuela-Raventós, R.M. (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent. *Methods in Enzymology*, 299, 152-178.
- Tester, R.F., & Morrison, W.R. (1990). Swelling and gelatinization of cereal starches. I. Effects of amylopectin, amylose, and lipids. *Cereal Chem.*, 67(6), 551-557.
- Thiele, C., Grassl, S., Gänzle, M. (2004). Gluten hydrolysis and depolymerization during sourdough fermentation. *Journal of Agricultural and Food Chemistry*, 52(5), 1307-1314.
- Topping, D. (2007). Cereal complex carbohydrates and their contribution to human health. *Journal of Cereal Science*, 46(3), 220-229.
- Topçu, G., Ay, M., Bilici, A., Sarıkürkcü, C., Öztürk, M., Ulubelen, A. (2007). A new flavone from antioxidant extracts of Pistacia terebinthus. *Food Chemistry*, *103*(3), 816-822.
- Traore, T.M., Icard-Verniere, C., Traore, A.S., Treches, S. (2004). Changes in nutrient composition, phytate and cyanide contents and x-amylase activity during cereal malting in small production units in Ouagadougou (Burkina Faso). *J Food Chem.*, 88, 105–114.
- Viéra-Dalodé, G., Jespersen, L., Hounhouigan, J., Moller, P.L., Nago, C.M., Jakobsen, M. (2007). Lactic acid bacteria and yeasts associated with gowé production from sorghum in Bénin. *Journal Applied Microbiology*, 103(2), 342-349.
- World Health Organization (WHO). 1996. Fermentation: Assessment and research. Report of a Joint FAO/WHO Workshop on fermentation as a household technology to improve food safety: 79.
- Zhang, J., Zhao, S., Yin, P., Yan, L., Shi, L., Ma, C. (2014). α-Glucosidase inhibitory activity of polyphenols from the burs of Castanea mollissima Blume. *Molecules*, *19*(6), 8373-8386.
- Zhao, Q., Hu, X., Guo, Q., Cui, S., Xian, Y., Gao, X. (2014). Physicochemical properties and regulatory effects on db/db diabetic mice of β-glucans extracted from oat, wheat and barley. *Food Hydrocolloids*, 37, 60-68.