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**Research Article**

# CONTRIBUTION TO THE CHARACTERIZATION OF FATTY ACID COMPOSITION OF THE OLIVE OILS EXTRACTED FROM VARIETIES AND LOCAL TYPES COMPARED TO THOSE OBTAINED FROM SOME IMPORTED FOREIGN VARIETIES IN COLLECTION CULTIVATED IN THE GROWING AREA OF OUAZZANE (NORTHERN MOROCCO)

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**ABSTRACT**

The main objective of this study is the characterization and assessment of purity parameters of oils extracted from studied varieties and local types, compared to those of introduced foreign varieties by the analysis and identification of fatty acid profiles (glyceride fraction). The lipidic composition is characterized by the predominance of UFA (C16:1, C18:1; C18:2) and SFA (C16:0, C18:0). Oleic acid ranges from 65% to 78% and is influenced by the variety. The proportion of palmitic acid is higher than the linoleic acid in ¾ of analyzed monovarietal oils. It is too high in foreign varieties than in the Moroccan Picholine types, similar to those recorded in the oleasters types, indigenous traditional and old Moroccan varieties. Most oils are characterized by practically identical and relatively high proportions of stearic acid and linolenic acid presented levels higher than 0,52%. The Moroccan Picholine types G9, G10 and Picholine of Languedoc have exceeded the limit of C18:3 set at 1%. The other minor fatty acids are similar in all studied varieties and local types. The influence of soil characteristics and climate conditions of the zone of origin, the genotype of cultivar and maturation stage of fruit at harvest are preponderant. CPA has distinguished three homogenous groups of varieties. Group1:Dahbia; BM4; BM2; BM3; BLg; G9; M1; BRK; Picual; BB and M6, which has a strong affinity for C18:0,C18:1,C20:0;C20:1 and high ratios of C18:1/C18:2 and MUFA/PUFA. Group2: Picholine of Languedoc related to C18:3. The groupe3: Gordal, BKa, BMM, Ascolana Tenera, BMR, BMK and Manzanille, that is approached by fatty acids: C16:0, C16:1, C17:0; C17:1, C18:2 and low ratios of C18:1/C18:2 and MUFA/PUFA.



## INTRODUCTION

In Morocco, although the olive tree is the main fruit species (65% of the national tree orchard), its varietal structure (genetic) is considered to be commonly limited to a single variety namely Moroccan Picholine that is the most represented in orchard (up to 96%) and very productive, characterized by a high intrinsic adaptability, interesting xerophytic characteristics and socio-economic importance. Today within this dominant and genetically heterogeneous variety, several synonyms cultivars and local types called Moroccan Picholine and whose distribution is restricted to limited and well-defined geographical areas and final destination of most of them is production of oil. Similarly, a fairly large number of these local, old and traditional varieties remain unknown to nowadays despite their large socio-economic interest and their ability to be canned (green and black olives) and to the extraction of oil (Marchenay *et* Lagarde, 1989). These varieties and local types that have not yet been cataloged and that have a potentiality of great values in terms of oil quality would conceal a high biochemical and genetic polymorphism. It is very important to arrive to characterize and identify individually the entire existing local germplasm as a reservoir of the olive genetic resources which are not yet exploited or categorized, where erroneous denominations occur more

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frequently than in the most commonly cultivated varieties (Cicatelli *et al.,* 2013). The approach adopted in this study is to characterize and evaluate the physicochemical performance of the oil obtained from almost majority of these varieties and local types, deemed to be more necessary and interesting, that recently have been developed by an easy and reliable methods to detect the richness of the virgin olive oil in triglycerides that are proven very useful for control of the varietal authenticity (fatty acids, sterols, phenolic compounds, tocopherols) (COI, 1996, 2003, 2009, 2011 *et* 2013; Virginie, 2001; Douzane *et al*, 2010 a *et* b; Ouazzani *et al.*, 2013 et 2014; El Antari, 2013 et 2014).

The oil chemical composition is much more characteristic of the cultivar and depends on several factors such as interaction variety x environment(crop year) (Lacertosa *et al*., 2004; Boskou *et al*., 2006; Allalout *et al.*, 2009; Amaral *et al.,*2010,Esmaeili *et al*., 2012; Cicatelli *et al.,*2013), the quality and health status of olives, degree of its maturity at harvest, cultivation and harvested techniques (Morello *et al*., 2004), geographical or plantation areas and climate (Aparicio *et* Luna, 2002; Piravi *et al.*, 2012), the time and olives storage conditions before producing oil, processing methods, grinding or crushing system, oil extraction and conservation (Boschelle *et al*., 1994; Bruni *et al.*, 1994 a *et* b;. Acar *et al*., 1996; Soulhi, 2000; Virginie, 2001; Abaza *et al*., 2005; Ben Temime, 2004, 2006 *et* 2008; Dhifi *et al*., 2006;Di Giovacchinno,2007 a *et* b; Baccouri *et al.,*2008 a et b; Ben Azzouz *et* Maata, 2008; Diraman, 2010; Terouzi *et al*., 2010; Montano *et al*., 2014). Similarly, the implantation site of olive orchards, geographical areas of production (altitude, latitude, soil or edaphic characteristics, climate), pests, plant protection, tree age, cultural techniques, are agronomic factors that can affect the quality, taste and aromatic characteristics of olive oil produced. Else this consideration have a great importance relatively to which the germplasm is suitable for the production of PDO labeled virgin olive oil (Cimato, 1990; Atouati, 1991; Di Giovacchino, 1991; Acar *et al.,* 1996; Dhifi *et al.,* 2006; Amirante *et al.,* 2006; Baccouri *et al.,* 2008, Amiral *et al.,* 2010; Dag *et al.,* 2011; Ranalli *et al.,* 2007 et 2012; Esmaeili *et al*., 2012; Cicatelli *et al.,* 2013).

However, in published literature, its well established that there is no scientific evidence or comparative studies and scarce information available on characteristics acquired about chemical composition (fatty acids profiles) of virgin olive oil produced of or from these depicted domestic and foreign cultivars grown in their traditional localities of origin (Ouazzane region categorized as Mediterranean climate with hot dry summer and mild to cool wet winters).

***Therefore the aim of this work was to proceed***

In the first time, characterization and evaluation of performances and physicochemical parameters of quality and purity of the olive tree regional patrimonial richness, oils extracted from ancient varieties and local types studied are compared to those obtained from the most important foreign varieties, despoiled and recently introduced by local farmers and which are performant with regard to olive oil content and oil chemical composition.

Secondly, research of important biochemical markers (chemometric index) as a specific and unique fatty acid profile and distinctive signs of origin and quality for oils differentiation, identification and distinction of studied recent varieties and local types.

Thirdly, to provide creation and implementation of the first innovative and defensible labellings in attempt to valorize and enforce the labeled of standard extra virgin olive oil produced at the wide region of olive production of Ouazzane, an area that is evocative and famous, particularly suitable for olive growing, located in northern Morocco, to whom we owe its national fame of table olives and olive oil, as a potential regional products, guarantee of intrinsic quality (taste, savour, aroma, flavour) and identified specific origins, that would be eligibly attributed a labels of origin geographical indications and recognition of a typical designations (PDO, PGI) to promote the marque image and the sign of clear regional identity or specificity around the olive tree patrimony germplasm (Cicatelli *et al*.,2013;Maata, 2014; Ouazzani, 2014; Rahmani, 2014), in order to guaranty and preserve the authenticity, intrinsic quality, excellence, and uniqueness of the virgin olive oil, as food supplement in pharmacies for example (Jakobusic *et al.,* 2015).

## MATERIALS AND METHODS

***Plant Material***

This study has focused on plant material, characterized using morphological descriptors (Kartas *et al*., 2014) and include 5 traditional Moroccan varieties (Bouchouk Laghlide, Bouchouk Rkike, Bakhboukh Beldi, Bouchouika, Dahbia), 6 Moroccan Picholine types (M1, M6, G9, G10, S1, S2), 6 oleasters types (BM2, BM3, BMK, BMM, BMR, BM4) and also involves 5 foreign cultivars (Picholine of Languedoc, Manzanille, Gordal, Picual, Ascolana Tenera) in collection, gathered from high performing farmers with regard to quality of extracted olive oil.

***Analysis methods***

***Olives samples and oil extraction***

References sites and maâsra (with a capacity of 0.9 to 1.2t/day) were selected from implantation site of each variety and local types, to perform the extraction of oil from mature black olives collected on an olive tree. The sites have a water source. Each millstone (maasras) of different sites provides olive oil samples. The harvesting techniques, extraction, and storage determined were completed under acceptable conditions in order to remove the factors capable of altering the olive and degrade fat matter, to obtain the olive oil of optimum quality.

The mature olives used and processed for oil extraction were picked (black stage of maturity) at a human height over the adult trees entire canopy of Moroccan Picholine population variety, indigenous varieties and oleasters types are grown in private groves. Other samples were collected from varieties of foreign origin. Only healthy fruits were harvest (without signs of infection by pests or physical damage) manually picked and were quickly transported to Maasras for their trituration within 3 days after harvest. Olives crushing is performed by a millstone grinder (granite stone roller) driven or drawn by

animal traction. The malaxation (mixing, kneading) operation is realized simultaneously with grinding. The olive paste is then put in concrete settling tanks filled with water, after which, the olive oil supernatant is separated by decantation of the liquid phase (vegetable or waste waters) by density difference, retrieved by hand and then filtered to remove solid impurities. The collected extracted oil is packaged in opaque flasks or brown glass, or glass bottles put in black plastic film (100% occupied volume), stored regularly at ordinary temperature (+4°C) in the dark (refrigeration) until analysis of the purity and varietal authenticity parameters performed in agreed laboratory of chemical analysis and researches of Casablanca in Morocco (COI, 2009 et 2013).

***The glyceride fraction (saponifiable)***

The glyceride fraction (saponifiable) represents between 98% and 99% of the oil total mass. It essentially contains triglycerides, considered as reserve lipids, the main constituents of the cell membrane, accompanied by small quantities of their degradation products, such as monoglycerides, diglycerides and free fatty acids and phospholipids (Sanchez *et al*., 2003).

***Fatty acid analysis (AFNOR: T60-233 et T60-234)***

The oil saponification allows obtaining mixtures of fatty acids. The total fatty acids composition of monovarietal oil were determined by gas chromatography after methyl esterification of the fatty acids. Methyl esters are prepared by esterification of the oil by the action of potassium hydroxide 2N, isooctane in the presence of hydrogen sulfates monohydrated sodium (micro esterification of fat matter).

***Preparation of fatty acids methyl esters (oil esterification)***

Quantity of oil from 0.30 to 0.40 g is put into a capped glass tube, 3 ml of a cold ethanolic solution of potassium hydroxide 2 N and 3 ml of isooctane is added to release and esterify fatty acids by a strong shaking (vortex), and then, 1 g of monohydrated sodium hydrogensulfate is added to the preparation, stirred again (vortex) and left to decant for 2 to 3 hours. Finally, 0.5 ml of the supernatant is removed and 0.5 ml of isooctane is added in a microviale and then is injected in the gaz chromatograph analysis (Agilent technologies, 7890 AGC System).

***Fatty acids***

Fatty acids are constituted by saturated or unsaturated carbon chain comprising a number of carbon atoms (14 to 24) with a predominance of fatty acids of 16 to 18 carbon atoms in the vegetable oils.

The major identified fatty acids are: palmitic (hexadecanoic), stearic, oleic, linoleic, and the minor fatty acids are: palmitoleic (hexadecenoic), margaric (heptadecanoic) margaroleic (heptadecenoic), linolenic, arachidic (ecosanoique) and gadoleic (ecosenoique) (Table n°1). The fatty acid composition is the first chemical criterion used to identify varieties (Sanchez *et al*., 2003, Guerfel *et al.,* 2012, Cicatelli *et al.,*2013), to evaluate adequately the purity of extra virgin olive oil (Zarrouk *et al*., 2009), and distinguish it from different geographical origins and other edible fluid vegetable oils (soybean, rapeseed, sunflower, flax, palm, hazelnut, sesame) (Boggia *et al*., 2005; Rondanini *et al*., 2011, Guerfel *et al.,* 2012).

## RESULTS AND DISCUSSION

From a qualitative point of view, the fatty acid composition of the studied oils, which mainly depends on the olive variety, the genotype x environment interaction and climate of the growing area, indicative of the authenticity of the virgin olive oil (Magliulo *et al.,* 2003; Dabbou *et al.,* 2010; Diraman, 2010; Rondanini *et al.,* 2011;Cicatelli *et al.,*2013; Homapour *et al.,* 2014; Fuentes *et al.,* 2015) is identical and conform to the requirement of the international trades standards (COI, 2013),while a quantitative variation is noted for fatty acids

**Table n°1** Fatty acids composition of olive oils issued from studied varieties and local types (saponifiable) and imposed legal limits(reference values)(Codex, 2003; COI, 2013).

| Varieties local types | C16: 0 | C16: 1 | C17: 0 | C17: 1 | C18: 0 | C18: 1 | C18: 2 | C18: 3 | C20: 0 | C20: 1 | C18:1/ C18:2 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Limits values (%)** | **7.5-20** | **0.3-3.5** | **≤ 0.3** | **≤ 0.3** | **0.5-5** | **55-83** | **3.5-21** | **≤ 1** | **≤ 0.6** | **≤ 0.4** | **3.95-15.71** |
| **BLg** | 10,426 | 0,657 | 0,0305 | 0,0622 | 2,258 | 77,118 | 7,845 | 0,818 | 0,367 | 0,414 | 9,830210325 |
| **BRK** | 11,936 | 0,938 | 0,057 | 0,093 | 2,137 | 74,439 | 8,976 | 0,756 | 0,351 | 0,31 | 8,293114973 |
| **BBeldi** | 10,009 | 0,7002 | 0,0671 | 0,06951 | 3,57661 | 75,05227 | 9,29771 | 0,68569 | 0,34986 | 0,2925 | 8,072124211 |
| **BKa** | 14,85 | 1,40994 | 0,0745 | 0,07357 | 3,07549 | 65,157 | 13,9215 | 0,7798 | 0,42412 | 0,23374 | 4,680314621 |
| **BM2** | 10,394 | 0,62414 | 0,0556 | 0,05845 | 2,3237 | 78,07492 | 6,90765 | 0,92634 | 0,30232 | 0,33325 | 11,30267457 |
| **BM3** | 10,282 | 0,88636 | 0,0497 | 0,10101 | 1,94125 | 77,66061 | 7,48859 | 0,89968 | 0,28639 | 0,40493 | 10,37052503 |
| **BMK** | 12,915 | 1,34 | 0,038 | 0,0658 | 2,372 | 70,5 | 11,162 | 0,978 | 0,328 | 0,298 | 6,316072388 |
| **BMR** | 14,097 | 2,05307 | 0,059 | 0,05802 | 2,0871 | 69,68391 | 10,88797 | 0,70363 | 0,24884 | 0,17954 | 6,400082844 |
| **BMM** | 14,771 | 1,54363 | 0,0475 | 0,09455 | 3,06908 | 66,91003 | 12,39578 | 0,67829 | 0,30474 | 0,18534 | 5,397807157 |
| **BM4** | 9,996 | 1,101 | 0,088 | 0,1008 | 2,302 | 77,479 | 7,519 | 0,828 | 0,307 | 0,275 | 10,30442878 |
| **PM1** | 8,129 | 0,688 | 0,05 | 0,08 | 2,198 | 78,103 | 9,336 | 0,947 | 0,234 | 0,362 | 8,365788346 |
| **PM6** | 10,438 | 0,86624 | 0,0368 | 0,06617 | 4,19134 | 74,34055 | 8,45361 | 0,87622 | 0,49974 | 0,23113 | 8,793941287 |
| **PMS1** | 9,468 | 0,73878 | 0,041 | 0,06883 | 2,51904 | 69,88329 | 15,83505 | 0,84475 | 0,27373 | 0,32773 | 4,41320299 |
| **PMS2** | 9,466 | 0,70825 | 0,043 | 0,0422 | 2,39236 | 70,12468 | 15,89928 | 0,77747 | 0,27352 | 0,27278 | 4,410556956 |
| **PMG9** | 9,25 | 0,676 | 0,05 | 0,07 | 2,45 | 76,57 | 9,43 | 1,009 | 0,296 | 0,303 | 8,119830329 |
| **PMG10** | 10,868 | 0,81349 | 0,0557 | 0,08621 | 2,57984 | 71,44701 | 12,64812 | 1,00105 | 0,2089 | 0,29182 | 5,648824489 |
| **Dahbia** | 11,158 | 1,44552 | 0,0232 | 0,0829 | 1,81914 | 74,03911 | 9,83282 | 0,92299 | 0,31707 | 0,34089 | 7,529794098 |
| **PLanguedoc** | 9,787 | 0,65764 | 0,0804 | 0,10091 | 2,30603 | 72,18235 | 12,97481 | 1,03146 | 0,37497 | 0,50473 | 5,563268364 |
| **Picual** | 13,008 | 1,124 | 0,0936 | 0,11486 | 2,47311 | 78,65259 | 3,30782 | 0,52833 | 0,3934 | 0,30482 | 23,77777207 |
| **Gordal** | 11,829 | 0,94238 | 0,1487 | 0,27082 | 2,79543 | 72,72787 | 9,89402 | 0,65308 | 0,36207 | 0,37693 | 7,350689608 |
| **ATénéra** | 13,166 | 1,25141 | 0,1334 | 0,22301 | 2,85492 | 69,44124 | 11,5637 | 0,58806 | 0,37081 | 0,40771 | 6,005105632 |
| **Manzanille** | 16,606 | 1,336 | 0,13 | 0,216 | 2,204 | 65,933 | 12,317 | 0,791 | 0,307 | 0,154 | 5,353008038 |

analyzed individually. This can be exploited to differentiate between the studied varieties and local types (Rotondi *et al.,* 2004; Baccouri *et al.,* 2008; Douzane *et al.,* 2010; Diraman, 2010*;* El Antari, 2013 et 2014; Ouazzani, 2013 et 2014; Fuentes *et al.,* 2015).

The acidic composition is characterized by the strong predominance of UFA (C16:1, C18:1, C18:2, C18:3) and SFA ones (C16:0, C18:0). Oleic acid exceeds 65.15% to 78.65% and clearly suffers from the varietal influence ???. While a slight superiority of palmitic acid proportions, regarding linoleic acid was observed in 73% of oil samples issued from varieties and local types studied (Table n°1).

In foreign varieties, the proportion of palmitic acid, the most abundant SFA is too high than that recorded in the Moroccan Picholine types, but this rate has remained almost similar to those recorded in the oleasters types and Moroccan indigenous or traditional varieties examined.

The rate of registered palmitoleic acid in oleasters types are superior of 14.43% compared to those obtained in foreign varieties, lower than 14.68% when compared to those encountered in local varieties and inferior to 7.37% than those recorded in Moroccan Picholine types.

Most oils are characterized by almost the same percentage and relatively high stearic acid, and presented linolenic acid content slightly exceeding 0.528% and reached even more than 1%.

This late value is the upper limit recently adopted by the COI (2013). Values exceeding this limit were found in samples taken from Moroccan Picholine types of Mjaara (G9, G10), while only a sample of the Ouazzane area has reached the limit value (Picholine of Languedoc variety). This suggests that the proportion of linolenic acid is significantly influenced by the soil characteristics and climatic conditions in the study area, and closely correlated with the genotype of cultivar and degree of maturity of the olives (Fiorino et Grifi, 1991; Aparicio *et al.,* 2002; Rotondi *et al.,* 2004; Baccouri *et al.,* 2008b; Ibrahim *et al.,* 2014).

The proportions of other minor fatty acids are substantially similar in all varieties and local types considered in this study. the Margaric (heptadecanoic) and margaroleic (heptadecenoic) acids were present in smaller quantities less than 0,27% and those of arachidic (ecosanoique) and gadoleic (ecosenoique) acids were less than 0,5%.

The fatty acid composition is an important parameter of purity and quality (length of shelf life) and classification of olive oils is based on varietal origin and discrimination between morphotypes of Spanish, Italians, Algerians and Turkish specific cultivars, Tunisians oleaster types and tree varieties obtained from cutting (Rotondi *et al.,* 2004; Boggia *et al.,* 2005; Baccouri *et al.,* 2008b; Nicoleta *et al.,* 2010; Douzane *et al.,* 2010aet b; Diraman, 2010; Dabbou *et al.,* 2010; COI, 2009, 2011 et 2013; Guerfel *et al.,* 2012;Fuentes *et al.,* 2015).

Comparison of varieties and local types necessarily involves the comparison of lipid substrate composition, which represents the major fraction of the olive oil. Therefore, in this study, we have the opportunity, through this oil acidic composition to distinguish between tested varieties and local types. Then it is possible to consider the proportions of the different classes of fatty acids as an interesting criterion for establishing differences in fatty acid profiles between cultivars and local types studied.

***Major fatty acids***

***C18: 1: Oleic acid***

The MUFA is the predominant acid in the composition of olive oil and its contents changes due to the diversity of cultivars, the environmental conditions, locality, and growing conditions. It is a recessive genetic marker that needs to be isolated from all the other cultivars (Bruni, 1994a *et* b; Diraman, 2010).

All the different varieties and local types studied are rich in oleic acid; their proportions easily exceed 65% and can discriminate between them. The highest contents are recorded respectively in the local variety Bakhboukh Beldi, Moroccan Picholine type G9, the local variety Bouchouk Laghlide, oleasters types (BM3, BM4, BM2), Moroccan Picholine type M1 and the Picual variety (75.05% - 78.65%) followed by the Moroccan Picholine type S2, oleaster type BMK, Moroccan Picholine type G10, Picholine of Languedoc, Dahbia, Gordal, Moroccan Picholine type M6 and Bouchouk Rkike varieties (70.12%-74.43%), while intermediate proportions (65.15%-69.88%) were noted at Bouchouika and Manzanille varieties, oleaster type BMM. Manzanille and Picual varieties, presented in the NW of Argentina and Turkey oleic acid percentage of 65.3%-74.19% and of 71.9% respectively (Diraman, 2010; Rondanini *et al.*, 2011; Fernandez-Silva *et al*., 2013).

Moroccan Picholine, Picholine of Languedoc and Manzanille varieties grown in the Haouz of Marrakech gave oleic acid percentage of (70% - 73%; 62% - 64% and 64% - 68%, respectively) (El Antari 2003 a *et* b).

The Moroccan Picholine variety recorded in northern Morocco and Turkey a higher oleic acid percentage (74.88%-76.80%; 64.8%-72.8%) (Diraman, 2010, Ibrahim *et al*., 2014; Essiari *et al*.,2014). Moroccan Picholine variety, Koroneiki, Haouzia grown in Settat and Sais regions, presented a very high content of oleic acid (76%; 76.25%-75.77% et74, 7% respectively) and the Arbequine variety presented the lowest content (62.42%-65.48%) (Haddam, 2014; Essiari *et al.*, 2014) decreasing during fruit growth and oil accumulation (Rondanini *et al*., 2011).

In Tunisia, oil extracted from varieties Chetoui, Chemlali (Sfax, Zarzis, Boughrara) Gerboui, Chaibi (56.1%-73.88%);Tounsi, Oueslati, Zarzis (74.5%-74,8%) and oleasters types (71%-78.4%) planted under rainfall conditions, are characterized by very high levels of oleic acid and MUFA, which progressively decreases with the increase of ripeness index (Hannachi *et al*., 2007;Baccouri *et al*., 2008a;Grati Kammoun, 2010; Dabbou *et al*., 2010;Guerfel *et al.,* 2012) and with the high temperatures arising early during lipogenesis (Magliulo *et al*., 2003) marking the different responses in terms of olive oil quality to water stress and the enzymes involved in fatty acid biosynthesis (Rondanini *et al*., 2011). Indeed, this rate increased with the addition of enzymes (pectinase, polygalactoturonase, cellulase, glucanase) to the paste during kneading (Iconomou *et al*., 2010).

Turkish varieties (Gemlik, Kilis, Uslu, Tirilye, Ayvalik) and the Greek variety (Cobrançosa), planted in irrigated or rainfed areas (bour), presented higher percentages of oleic acid (65.7%-83.6%) (Taniglan *et al*., 2007) and (69.7%-73.6%) (Fernandez-Silva *et al*., 2013). In Italy, extra virgin oil of varieties (Leccino, Liguria) has a high oleic acid content (>75%) with low levels of linoleic and palmitic acids, giving it a high oxidative stability and a specific nutritional quality (Dettori *et* Russo, 1993; Fuentes *et al*., 2015) and therefore a high resistance to oxidation (Cinquanta *et al*., 2001; Rondanini *et al*., 2011). However, the oleic acid content of Colombia variety is low (65%) (Boggia *et al*., 2005).

***Palmitic acid C16:0***

The proportions of palmitic (hexadecanoic) acid, the most dominant SFA in olive oil, vary depending on the region and the studied variety and local types. In fact, according to the content of this fatty acid, varieties, and local types can be classified in descending order. In the regions of Mjaara and Ouazzane, the Manzanille and Bouchouika varieties, oleasters types (BMM, BMR), Ascolana Tenera and Picual varieties are richer in palmitic acid, with respective higher levels ranging from 13.0% to 16.6%. The Moroccan Picholine variety has presented in northern Morocco a palmitic acid amount in the range of 11.34% to 17.68% (Ibrahim *et al*., 2014) and 9.37% to 11.56% at Sais region (Essiari *et al*., 2014). In Tunisia, the oleaster types, gave a palmitic acid percentage ranging from 8.7% to 11.9% (Baccouri *et al*., 2008b) making them figeables at low temperatures (Abaza *et al*., 2002). In the Chetoui and varieties Coratina, Itrana, Pendilino, Ottobratica, Picholine, Nociara, the content of palmitic acid gradually decreases as fruit ripened (October to January) (dilution effect) (Bour:12.44% to 9.42%;Irrigated:12.97% to 10.57%) (Poiana *et* Mincione, 2004; Baccouri *et al*., 2008) increases with irrigation of trees (Dettori *et* Russo, 1993; Fernandez-Silva *et al*., 2013) and elevation of temperatures during lipogenesis (Leccino cv) (Magliulo *et al*., 2003). The highest value (18.2%) was recorded in the Chemlali Sfax variety (suckers, cutting) and the lowest in the Coratina foreign variety (11.1%) (Dabbou *et al*., 2010; Guerfel *et al.,* 2012).

In areas of Mzefroune and Masmouda the percentages of this fatty acid, are intermediate and varied from 10% to 12.91%, as in some case of oleaster type BMK, the local variety Bouchouk Rkike, Gordal, Dahbia varieties, Moroccan Picholine types (G10, M6), the local variety Bouchouk Laghlide, the oleaster types (BM2, BM3) and the local variety Bakhboukh Beldi.

Oleaster type BM4, Picholine of Languedoc variety and Moroccan Picholine types (S1, S2, G9, M1), are distinguished from other varieties by their small proportions of palmitic acid ranging from 8.12% to 9.99% (region of Sidi Bousber) (Table n°1). In Settat region the Moroccan Picholine variety presented a palmitic acid content of 10.1% (Haddam, 2014). The Turkish varieties (Gemlik, Kilis, Uslu, Tirilye, Ayvalik) gave palmitic acid percentage of 8.1% to 15.2% (Taniglan *et al.,* 2007).

***Linoleic acid C18:2***

The studied varieties and local types can be classified in ascending order according to the content of this fatty acid, the most dominant PUFA, which is much more susceptible to oxidation than MUFA(C16:1, C17:1, C18:1). The Picual variety presented the lowest percentage (3.30%). Oleaster types (BM2, BM3, BM4) and local varieties Bouchouk Laghlide, Moroccan Picholine type M6, Bouchouk Rkike presented a rate ranging from 6.90% to 8.97%. The highest linoleic acid rates, were observed in the local variety Bakhboukh Beldi, Moroccan Picholine types M1 and G9, the Dahbia, Gordal varieties, oleaster types (BMR, BMK), Manzanille, Ascolana Tenera varieties, oleaster type BMM, Picholine of Languedoc, Bouchouika varieties and Moroccan Picholine types (S1, S2) (9.29% to 15.89%).

The Manzanille variety has given in Turkey a linoleic acid percentage of 7.06% (Diraman, 2010). The Moroccan Picholine variety has given a linoleic acid proportion of 6.83% in Turkey (Diraman, 2010), 9.35% in Settat region (Haddam, 2014), 6.08% to 11.68% in northern Morocco (Ibrahim *et al*., 2014) and 9.38% to 9.4% in Sais region (Essiari *et al*., 2014). In Chetoui and Cobrançosa varieties, linoleic acid reached in irrigated conditions and full maturity stage a high rate of 7.8% to 10.2% (Baccouri *et al*., 2008a; Fernandez-Silva *et al*., 2013). The oleaster types recorded a linoleic acid level of 6.8% to 14.2%. Between the early (late October) and semi-late olives harvesting (early December, January), oleic acid decreases and linoleic acid increases (Poiana et Mincione, 2004; Rotondi *et al*., 2004; Baccouri *et al*., 2008a; Oueslati *et al*., 2009; Fernandez-Silva *et al*., 2013) due to the intense activity of oleate desaturase enzyme in fruit, which transform oleic acid to linoleic acid during the triacylglycerol biogenesis (Gutierrez *et al*., 1999; Baccouri *et al*., 2008a; Hashempour *et al*., 2010). The C18:1/C18:2 ratio, which decreases during the maturation of olives (Poiana *et* Mincione, 2004), can evaluate the maximal oxidative stability and nutritional quality among other Tunisian varieties (Chetoui, Chemlali) (r2:0.71) (Baccouri *et al*., 2008a), affect taste (fruity, bitter, pungency, sapidity) (Bouskou, 1996) and detect the mixture of virgin olive oil blended with refined or cheaper oils (oil seeds, pomace oil) (Oueslati *et al*., 2009). In Tunisia this ratio marked a large variation between cultivars in collection (Arbequina: 4.35; Koroneiki: 13.4; Coratina: 10.85; Chemlali (Boughrara: 4.64; Zarzis: 12.17; Sfax: 3.31) (Dabbou *et al*., 2010) and in Spain, the ratio distinguished between oil of irrigated olive groves and those non irrigated (rainfed) (Salas *et al*., 1997). A strong negative correlation between this ratio and the maturity index was found in the cv Nostrana di Brisighella ($r^2$:0.99) (Rotondi *et al*., 2004) and in Tunisian varieties Chetoui (irrigated $r^2$:0.812; unirrigated $r^2$: 0.949) and Chemlali ($r^2$:0.513) (Baccouri *et al.,* 2008a). Iranian variety Mari has an oleic/linoleic ratio higher than the other varieties (Zard, Raowghani), a percentage of MUFA greater than that of UFA and its oil is so stable and resistant to oxidation than the Zard and Phishomi varieties. Rowghani variety has a high content of PUFA, particularly linoleic acid (Homapour *et al*., 2014). In Spain, the Andalusia varieties (Carrasqueira and Cornicabra) have a low linoleic acid content, a high percentage of oleic acid and high oxidative stability; while the (Moriska, Manzanille, Onil de Bovidella) varieties with low content of oleic acid and high levels of linoleic acid, has low oxidative stability = ($r^2$ : 0.94) (Salvador *et al*., 2001 et 2003; Garcia *et al*., 2009; Oueslati, 2009;Vekiari *et al*., 2010; Fuentes *et al*., 2015). In Greek variety Megaritiki, the presence of the enzymes (pectinase, cellulase, polygalactoturonase, glucanase) during the malaxation of the paste accelerates the oxidation of linoleic acid, thus reducing its percentage, and regenerating an increase

in oleic acid (Ranalli *et al*., 2003; De Faveri *et al*., 2008; Iconomou *et al*., 2010). Turkish varieties (Gemlik, Kilis, Uslu, Tirilye, Ayvalik) presented linoleic acid percentages of 3.5% to 15.5% (Taniglan *et al*., 2007). Chemlali Tunisian variety (tree of cutting) has a higher percentage of PUFA, due to the high content of linoleic acid (C18:2) (Guerfel *et al.,* 2012).

***Stearic acid C18:0***

The stearic acid can be used in varietal distinction (Baccouri *et al*., 2008). According to our results it's evidenced that Bakhboukh Beldi, Bouchouika local varieties, Oleaster type BMM, and Moroccan Picholine type M6, can be distinguished by high levels of this fatty acid (3.07%-4.19%). In opposite, the Dahbia variety and Oleaster type BM3 had the lowest percentage (1.81% and 1.94%). The oils from the Chetoui variety (irrigated and unirrigated), had high stearic acid levels at the last stage of ripening (end of maturity). However, oleaster types gave a stearic content varying from 1.5% to 3.5%. Turkish varieties (Gemlik, Kilis, Usulu, Tirilye, Ayvalik) had a percentage of stearic acid from 2.0% to 5.6% (Taniglan *et al*., 2007). In the Chemlali and Pendilino varieties, the content of this fatty acid decreases with olives maturation process (Baccouri *et al*., 2008a) and at varieties (Coratina, Itrana, Ottobratica, Picholine, Nociara) its exhibit an increasing as ripening proceeded (Poiana *et* Mincione, 2004). In Settat region, Moroccan Picholine, Haouzia, Koroneiki and Arbequine varieties presented a stearic acid content of 2.15%, 2.55%, 2.35% and 1.75% respectively (Haddam, 2014).

concerning the composition of the virgin olive oil and may be of great interest for their beneficial nutritional effect or impact and implication (Diraman, 2010). Another interesting criterion that can distinguish varieties and local types is to consider the proportions of different fatty acids classes, which vary with the varieties and the geographical areas of cultivation (Raina *et al*., 1986; Gouveia, 1997; Abaza *et al*., 2002; Baccouri *et al*., 2008b; Douzane *et al*., 2010a et b; Diraman, 2010; Essiari *et al*., 2014). Thus, the local variety Bouchouika, oleasters types (BMM, BMR, BMK), Moroccan Picholine Type M6, Manzanille, Ascolana Tenera and Gordal foreign varieties are the most rich in SFA (15.14%-19.25%) while Bouchouk Laghlide, Bouchouk Rkike, Bakhboukh Beldi local varieties, oleaster types (BM2, BM3, BM4), Moroccan Picholine type

G9, Picual, and Dahbia varieties have moderate levels in PUFA (10.61%-14.64%) and also a greater lipophilic antioxidant (phenols) content, known for their effective inhibition of lipid oxidation. Which indicate a lower potential susceptibility to inevitable self-oxidation process of oil (Fuentes *et al*., 2015) that starts after its extraction and causes a loss of quality (fruity, bitter, pungent) and leads to a deterioration(rancidity) that always becomes more serious during oil storage (Ben-Hassine *et al*., 2013).

In the Mari Iranian cultivar and varieties Koroneiki, Chemlali Zarzis the UFA were at the highest level, due to the higher ratio of oleic/linoleic acid (C18:1/C18:2).

**Table n°2** The different classes of fatty acids of olive oil issued of studied varieties and local types

| Varieties and local types | Saturated fatty acids (SFA) | Unsaturated fatty acids (UFA) | UFA/SFA | Mono unsaturated fatty acids (MUFA) | Poly unsaturated fatty acids (PUFA) | MUFA/ PUFA |
|---|---|---|---|---|---|---|
| **Bouchouk Laghlide** | 13,08 | 86,91 | 6,64 | 78,25 | 8,663 | 9,03 |
| **Bouchouk Rkike** | 14,48 | 85,51 | 5,91 | 75,78 | 9,732 | 7,79 |
| **Bakhboukh Beldi** | 14,002 | 86,10 | 6,15 | 76,11 | 9,98 | 7,63 |
| **Bouchouika** | 18,42 | 81,58 | 4,43 | 66,87 | 14,70 | 4,55 |
| **Oleaster type BM 2** | 13,08 | 86,92 | 6,65 | 79,09 | 7,83 | 10,10 |
| **Oleaster type BM 3** | 12,56 | 87,44 | 6,96 | 79,05 | 8,39 | 9,42 |
| **Oleaster type BM K** | 15,65 | 84,34 | 5,39 | 72,20 | 12,14 | 5,95 |
| **Oleaster type BM 4** | 12,69 | 87,30 | 6,88 | 78,96 | 8,35 | 9,46 |
| **Oleaster type BM R** | 16,49 | 83,57 | 5,07 | 71,97 | 11,59 | 6,21 |
| **Oleaster type BMM** | 18,19 | 81,81 | 4,50 | 68,73 | 13,07 | 5,26 |
| **Morrocan Picholine type M1** | 10,61 | 89,52 | 8,44 | 79,23 | 10,28 | 7,71 |
| **Morrocan Picholine type M6** | 15,17 | 84,83 | 5,59 | 75,50 | 9,33 | 8,09 |
| **Morrocan Picholine type G9** | 12,05 | 88,06 | 7,31 | 77,62 | 10,44 | 7,44 |
| **Morrocan Picholine type G10** | 13,71 | 86,29 | 6,29 | 72,63 | 13,65 | 5,32 |
| **Morrocan Picholine type S1** | 12,30 | 87,70 | 7,13 | 71,02 | 16,68 | 4,26 |
| **Morrocan Picholine type S2** | 12,18 | 87,82 | 7,21 | 71,15 | 16,68 | 4,27 |
| **Dahbia** | 13,32 | 86,66 | 6,51 | 75,91 | 10,76 | 7,06 |
| **Picholine of Languedoc** | 12,55 | 87,45 | 6,97 | 73,45 | 14,01 | 5,24 |
| **Picual** | 14,64 | 85,38 | 5,83 | 78,89 | 6,49 | 12,15 |
| **Ascolana Tenera** | 16,52 | 83,47 | 5,05 | 71,32 | 12,15 | 5,87 |
| **Gordal** | 15,14 | 84,86 | 5,61 | 74,32 | 10,55 | 7,05 |
| **Manzanille** | 19,25 | 80,75 | 4,20 | 67,64 | 13,11 | 5,16 |

Moroccan Picholine cultivated in northern Morocco presented a rate of 1.04% to 2.06% (Ibrahim *et al*., 2014) and 2.18% to 2.24% at Sais region (Essiari *et al*., 2014). Iranian variety Mari has given the lowest stearic acid content (2.3%) while the Zard variety gave the highest amount (3.05%) (Homapour *et al*., 2014). A higher SFA (palmitic, stearic), in condition of irrigation and MUFA, PUFA and UFA higher under dry conditions (Salas *et al*., 1997), provide better knowledge

Therefore, the percentage of MUFA is greater than PUFA and their olive oil is more stable and resistant to auto-oxidation than other olive oil varieties (Zard, Rowghani, Phishomi) and (Arbequine, Chemlali Sfax),which has the highest oxidative susceptibility for their higher content of PUFA especially linoleic acid (C18:2)(Dabbou *et al*.,2010; Homapour *et al*., 2014).

Furthermore, we consider that both categories of the reconstituted varieties have very similar proportions of MUFA. PUFA (C18:2), unstable, more sensitive to oxidation than MUFA and essential (C16:1, C17:1, C18:1), that cannot be biosynthesized by the human body, are of great biological importance in the metabolism and must, therefore, be supplied by the traditional diet. Olive oil is the most consumed vegetable oils in its raw state or as crudate fat, it has significantly a higher ratio of MUFA/PUFA (4 to10), increase even in non-irrigated conditions (Gomez-Rico *et al.,* 2007), and decrease during the ripeness process of olives or as the harvesting time delayed (Poiana *et* Mincione, 2004;Baccouri *et al*., 2008a et b), it confers to the olive oil a higher oxidative stability and an interesting nutritional quality (Perrin, 1992; Baccouri *et al*., 2008a et b; Diraman, 2010, Fuentes *et al*., 2015). The obtained results provide evidence that oils extracted from studied varieties and local types have significant proportions of UFA that may be strictly related to frosts and very low temperatures recorded in the Ouazzane area (Kiritsakis, 1993).The cold affects the fatty acid transcription of denaturize enzyme during the oil biogenesis (fruit ripening) responsible for the MUFA/PUFA ratio in the olive drupes and that the oil quality responds to the cold hardiness of different olive genotype (Cicatelli *et al*.,2013). They offer levels of UFA, SFA, UFA/SFA ratio which increase throughout the ripening of the olives (Poiana *et* Mincione, 2004) and MUFA, similar to those reported in the literature in oils produced in other neighboring olive-growing areas of the Mediterranean basin (Surinder *et al.,* 1986; Rahmani, 1989; Synouri, 1995; Gouveia, 1997; Sanchez ,1999; Abaza *et al.,* 2002; Poiana *et* Mincione, 2004; Baccouri *et al.,* 2008a et b; Douzane *et al.,* 2010a et b; Diraman, 2010; Rondanini *et al.,* 2011; Guerfel *et al.,* 2012; Haddam *et al.,* 2014; Ibrahim *et al.*, 2014; ELYounsi *et al.,* 2014, Essiari *et al.,* 2014).

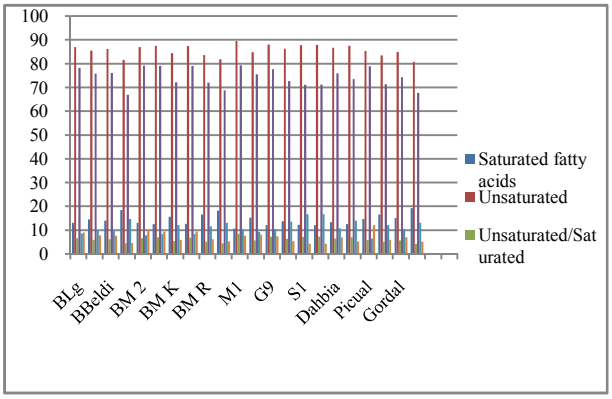



**Figure** n°**1** The different classes of fatty acids issued from studied varieties and local types of olive tree.

***Minor fatty acids***

***C18:3***

The majority of studied varieties and local types have poor oils in linolenic acid C18:3 (<1%), which are more stable, more resistant to oxidation and require little or no chemical hydrogenation (COI, 2011 *et* 2013). Depending on the proportions of this fatty acid, not specific in many olive varieties, two types of varieties are distinguished: The first type, with rates exceeding 1%, its the case of Picholine of Languedoc variety and Moroccan Picholine types (G10, G9) and the second type gathered the other varieties with values less than 1%. Moroccan Picholine, Picholine of Languedoc, Manzanille, M26, Haouzia, Menara, Arbequine, Leccino varieties presented in Marrakech high linolenic acid content (1.04% to 1.28%) (ElAntari *et al*., 2003; ElYounsi *et al*., 2014). In Tunisia, the Picholine of Languedoc variety gave a linolenic acid content of 1.43% (Zarrouk *et al*., 2009). The Moroccan Picholine variety recorded in northern Morocco and Sais region, has a linolenic acid rate of 0.16% to 0.96% (Ibrahim *et al*., 2014; Essiari *et al.*, 2014), this rate is reducing drastically from South to North of Morocco (ElAntari *et al*., 2000 *et* 2003) and during the ripening Italian cultivars (Itrana, Picholine, Cassanese, Pendolino)(October to January)(Poiana *et* Mincione, 2004). It was reported that the percentage of linolenic acid in oils reached 3% in Turkey (Taniglan *et al*., 2007);1,2% to 1.42% in Argentina (Ravetti *et al*., 1999;Rondanini *et al*.,2011);1,1% in Italy (Poiana *et* Mincione, 2004) and 1.5% in New Zealand (Meehan, 2001). The though variation in linolenic acid rate is attributed to seasonal differences of soil water reserves (Romero *et al*., 2003).

Currently, the COI norm (2013) (Table n°1) recently fixed to 1% as a maximum content of this fatty acid, a tracer of adulteration or fraudulent manipulation of virgin olive oils and of falsification of its origin by other oils seeds richer in linolenic acid (sunflower, rapeseed, soybean, palm, flax, sesame) (Olivier, 2003; Haddam *et al*., 2014) and determining its sensory characteristics (quality). It is stated that a lower ratio of C18:2/C18:3 correspond to a higher bitterness (intense better taste, green odor, flavor) (Diraman, 2010).

However, 14% of studied varieties and local types, normally produce proportions of linolenic acid that slightly exceeded the limit of 1%, which limit global market value and criminalizing Moroccan oil to be exported (Maata, 2014; Rahmani, 2014). This suggests wondering about the advisability of the use of such a standard that ignores the reality of upper limits that can reach varieties and local types of olive trees growing under different soil and climatic conditions in the Mediterranean countries. It would be interesting to enhance the ceiling value of the standard to 1.25%, while waiting to lead throughout the Mediterranean countries, the comparative study of olive oils fatty acid composition and to develop the study of the national olive cultivation (INRA, ENA), with the participation of specialized laboratories agreed by the COI (Giampiero, 1999; Codex alimentarius, 2003; El Antari, 2013 et 2014; Ouazzani, 2013 et 2014; Maata, 2014; Rahmani, 2014). It represents the opportunity to evaluate the organoleptic, chemical and technological performances of Moroccan olive oil heritage in different olive basins of the country and its valorization. Preliminary results are encouraging and allowed to characterize oils, identify anomalies and performances and to quantify the variability degree within the dominant local variety (Moroccan Picholine). It is an opportunity to delimit potential cultivation sites for possible technological protection against appellation usurpations, through the labeling and distinctive signs of origin and quality researched and to adopt for local viable and sustainable development in marginal and difficult areas, while preserving local knowledge and biodiversity (El Antari 2013 et 2014; Ouazzani 2013 et 2014; Rahmani, 2014; Maata, 2014; Jakobusic *et al*., 2015).

***Other fatty acids***

The proportions of the other fatty acids are practically almost identical and don't show any particular proportions to evoke. The palmitoleic (hexadecenoic) acid (C16:1), heptadecanoic acid (C17:0) and heptadecenoic (C17:1) exists in all oils of varieties and local types analyzed, with low contents ranging from 0,624% to 2,053% and 0.03% to 0.27% respectively. While the contents of arachidic acid (C20:0) and those of gadoleic acid (C20:1), range between 0.15% and 0.50%. These fatty acids, weakly represented and consistent with those established by (Codex, 2003; COI, 2013) can be used as a criterion for varietal characterization, this is the case of the fatty acids C16:1, C17:1 and C20:0. The local variety Bouchouika, oleaster types (BMK, BMR, BMM, BM4), Dahbia, Picual, Manzanille and Ascolana Tenera varieties, can be distinguished by the fatty acid (C16:1), the foreign varieties (Picholine of Languedoc, Picual, Gordal, Ascolana Tenera, Manzanille) and oleaster type BM3 are characterized by fatty acid (C17:1) and finally, the variety Bouchouika and Moroccan Picholine type M6 by the fatty acid (C20:0).Iranian Rowghani variety presented the highest content (C16:1), comparatively to the other varieties Zard and Mari (Homapour *et al*., 2014). Principal components analysis (PCA) also was applied to lipid compounds groups taken individually and simultaneously, to determine the presence of fatty acids of unification of studied varieties and local types (Alessandri, 1997; Matos *et al*., 2007; Baccouri *et al.,* 2008b; Douzane *et al.,* 2010b). According to this analysis, three homogenous groupings of cultivars and local types aggregated together are distinguished (Figure n°2 et n°3).

**Group 1**: Formed by Dahbia, BM4, BM2, BM3, BLg, G9, M1, BRK, Picual, BB and M6, which has an affinity related to fatty acids C18:0, C18:1, C20:0 and C20:1 and a high ratios of C18:1/C18:2 (7.53 to 23.78) and MUFA/PUFA (7.44 to 12.15).

**Group 2**: Consists of Picholine of Languedoc, G10, S1 and S2, which is distinguished by the fatty acid C18:3.

**Group 3**: Clustered Gordal, BKa, BMM, Ascolana Tenera, BMR, BMK, Manzanille, which are characterized by similar proportions of fatty acid C16:0, C16:1, C17:0, C17:1, C18:2. Groups 2 and 3 are characterized by low ratios of C18:1/C18:2 and MUFA/PUFA ranging from 4.41 to 7.35 and from 4.55 to 7.05 respectively (Poiana et Mincione, 2004; Zarrouk *et al*., 2009).

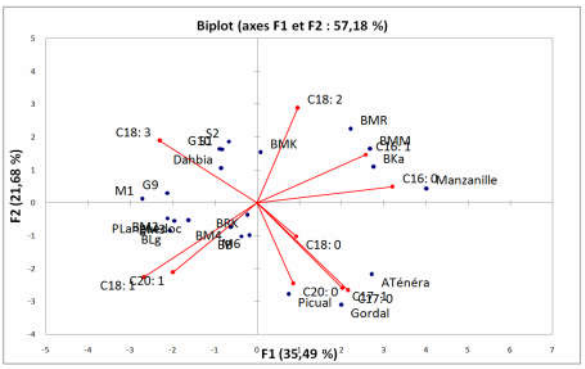



**Figure** n°**2** ACP of the olive oil fatty acids obtained from studied varieties and local types

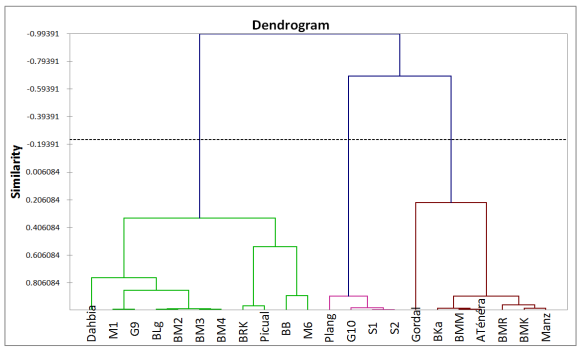



**Figure** n°**3** Dendrogram (CAH) of olive oil fatty acids extracted from studied varieties and local types

For varieties and local types analyzed, the fatty acids proportions observed are complies with the maximum limits specifications adopted by international trade standards, as the purity criteria for extra virgin olive oil and largely determined by the genotype of the cultivar (Rotondi *et al*., 2004; Virginie, 2001; Baccouri *et al*., 2008b; COI, 2009, 2011 *et* 2013,Cicatelli *et al*.,2013), even they are influenced by the soil and climatic conditions of the origin area, cultivation practices and stage of maturity at harvest (Tsimidou *et al*., 1993; Synouri *et al*., 1995; Ranalli *et al*., 1997; Virginie, 2001; Aparicio *et* Luna, 2002; Rotondi *et al*., 1999;2004; Matos *et al*., 2007; Zarrouk *et al*., 2008; Douzane *et al*., 2010a et b; Rondanini *et al*., 2011; Essiari *et al*., 2014). In the Arbequine variety, the elevation of the maximal temperatures of the day by hot air (5°C-10°C) reduces the oleic acid content (2% per °C) in NW Argentina during the biosynthesis and the phase of lipids accumulation (lipogenesis) (Douzane *et al*., 2010a; Rondanini, 2011) and in Turkey the oleic/linoleic ratio is low due to the high content of linoleic acid (Diraman, 2010). However, in oil seeds (soybean, sunflower, flax), high temperatures occurring during seed development, give a higher oleic acid content at the expense of PUFA(Gunstone *et al*., 2007). In Algeria, the oil issued from the Sigoise variety, has a high oleic acid percentage of about 82% (Chemlal and Azeradj, 72%) with 7% to 10% of linoleic acid, 0.4% to 0.8% of linolenic acid and 6% to 14% of palmitic acid (Talantikite *et* Ait Amar, 1988; Douzane *et al*., 2010a et b) and in Tunisia 70.89% (Zarrouk *et al*., 2009). In Italy, the acidic composition of olive oil, extracted from several cultivars (Frasai, Carolea, Coratina, Itrana, Pendilino, Ottobratica, Picholine, Nociara) has the higher oleic acid content. The content of oleic and linoleic acids increases with the maturation of the olives (ripeness index) (October to January), while the ratio C18:1/C18:2 decreases (Fiorino *et* Grifi, 1991; Caselli *et al*., 1993, Poiana et Mincione, 2004) and affect the taste of virgin olive oil used as an integral part of the Mediterranean diet (Bouskou, 1996). The C18:2 is an essential condiment due to their beneficial nutritional and healthful effects recognized in preserving cardiovascular diseases, diabetes, obesity, cancers (Kiritsakis *et al*., 2007). A strong positive correlation was observed between C18:1/C18:2, MUFA/PUFA ratios and the oxidative stability of olive oil extracted from fruits harvested at an earlier date (October) in the Nostrana di Brisighella variety ($r^2$:0.89) (Rotondi *et al*., 2004) and that obtained from the four

varieties Moroccan Picholine, Koroneiki, Haouzia and Arbequine) ($r^2$:0.52) (Haddam *et al*., 2014).

The oil of varieties Moroccan Picholine, Koroneiki and Haouzia, has a low content of PUFA and higher MUFA/PUFA ratio which is more stable (Kamal- Eldin, 2006). However, the oil from the Arbequine variety with a weaker MUFA/PUFA ratio and a higher level of PUFA (C18:2) recorded the low stability (Diraman *et al.,* 2010; Haddam *et al*., 2014).

The level of SFA (stearic acid, palmitic acid) and UFA (linolenic acid) tend to decrease during the evolution of fruit ripening and contribute to the characterization of varieties which affect the acidic profile of virgin olive oil (Stefanoudaki *et al*., 1999, 2001 et 2009; Zarrouk *et al*., 2008).

Variations of stearic acid in the same medium, different between the varieties Frantoio and Carolea, are closely related to the genotype of cultivar (Maestro, 1990; Fiorino *et* Grifi, 1991; Abaza *et al*., 2002). In arid Tunisian areas, oils obtained from indigenous (K17, K20, K21) and introduced varieties (Nabali, Koroneiki, Coratina Arbequine) compared with the widespread varieties (Chemlali Zarzis, Chemlali Sfax) show significant differences between the tested cultivars taking into regard the fatty acid composition.

Moroccan Picholine populations, randomly selected (M1, M6, G9, G10, S1, S2) and most representative of existing groves. The soils are generally marly calcareous and of an altitude of about 172 m, 220 m and 261 m respectively.

In the peripheral region of Ouazzane, the selection of olive trees concerned the cultivar Picual, in high production (on year), conducted in the driest areas (rainfed), and implanted in the Piedmont of Bouhlal mountainous highland (Jebel) (Altitude: 609 m) in three different agroclimatic locations:

- **Site 1** is located in the West, 5 km from the town of Ouazzane, on the left side of the route, connecting the city of Ouazzane to Souk Larbaa on marly-calcareous soil and northern exposure. The altitude is of 217 m.
- **Site 2** is located in the southeast at 15 km of Ouazzane, by taking the route going to the town of Fez on hamri soils (red, rubified), and southern exposure. The altitude is of 240 m.
- **Site 3** is located in the southwest at 11 km of Ouazzane, on calcareous marly soils of hill land and northern exposure. The altitude is of 240 m.

**Table n°3** Fatty acid composition of olive oil, according to the geographical location, altitude and exposure of planted areas

| Varieties and geographical sites | Fatty acids | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | C16:0 | C16:1 | C17:0 | C17:1 | C18:0 | C18:1 | C18:2 | C18:3 | C20:0 | C20:1 |
| **Picual (Ouazzane)** | - | - | - | - | - | - | - | - | - | - |
| Site 1 (North) | 8,65 | 0,59 | 0,13 | 0,22 | 3,76 | 76,77 | 8,48 | 0,64 | 0,43 | 0,33 |
| Site 2 (South) | 11,20 | 0,91 | 0,04 | 0,06 | 3,32 | 77,32 | 5,86 | 0,59 | 0,44 | 0,26 |
| Site 3 (North) | 13,01 | 1,12 | 0,09 | 0,11 | 2,47 | 78,65 | 3,38 | 0,53 | 0,39 | 0,31 |
| **M.Picholine :** | - | - | - | - | - | - | - | - | - | - |
| Mjaara (North) | 9,25 | 0,68 | 0,05 | 0,07 | 2,45 | 76,57 | 9,43 | 1,01 | 0,30 | 0,303 |
| Mjaara (North) | 10,87 | 0,81 | 0,06 | 0,09 | 2,58 | 71,45 | 12,65 | 1,001 | 0,209 | 0,292 |
| S.Bousber(South) | 9,47 | 0,74 | 0,04 | 0,07 | 2,52 | 69,88 | 15,84 | 0,84 | 0,27 | 0,33 |
| S.Bousber (South) | 9,47 | 0,71 | 0,04 | 0,04 | 2,39 | 70,12 | 15,89 | 0,78 | 0,27 | 0,27 |
| Masmouda(South) | 8,13 | 0,69 | 0,05 | 0,08 | 2,20 | 78,10 | 9,34 | 0,95 | 0,23 | 0,36 |
| **Masmouda(South)** | 10,44 | 0,87 | 0,04 | 0,066 | 4,19 | 74,34 | 8,45 | 0,876 | 0,499 | 0,231 |

Local varieties (K17, K20, K21) and varieties (Koroneiki, Coratina, Chemlali Zarzis) have given the highest percentage of oleic acid (60.5%; 76.8%; 75.8%; 73, 9% respectively). The varieties Arbequine and Chemlali Sfax had the lowest rate (Khlif et Trigui, 1990; Humed *et al*., 1991; Dabbou *et al*., 2010). The Gerboui local variety is characterized by a high oleic acid rate (60, 5%) and smallest proportions of palmitic acid and linolenic acid (13.4% and 0.55%) (Mehri Mehri *et* Grati -Kamoun, 2007). The Nabali Jordanian variety has a high proportion of arachidic, stearic and linoleic acid (Humed *et al*., 1991). The Mari Iranian variety shows high UFA content and low SFA/UFA ratio, due to the low content of palmitic and stearic acid (Homapour *et al*., 2014). UFA/SFA and MUFA/PUFA ratios are higher in oil extracted from unirrigated olive groves (bour or rainfed).

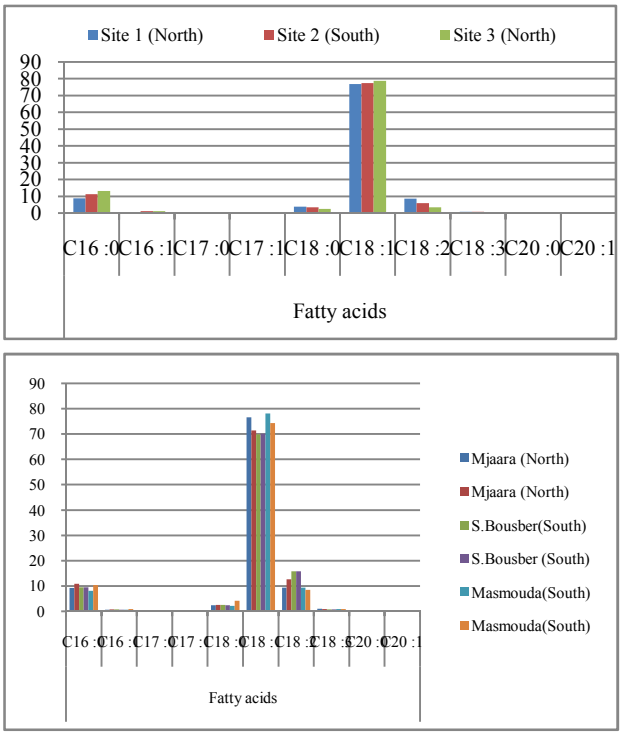
***Influence of the geographical site***

It is important to characterize a cultivar in different area of cultivation, to estimate the limits of its technological potentials researched, in particular the chemical composition of olive oil (Hannachi *et al*., 2007).

The comparative study of fatty acid profiles, was conducted in three referenced geographical sites (Masmouda, Mjaara, Sidi Bousber), and focused on the Picual variety and 6 predominant

For both sampled and targeted varieties, samples were taken at the stage of full black maturity (over ripe fruit). The olives are collected exclusively from trees harvested at a human height over the totality of canopy. The main characteristics of the oils (glyceridic fraction), determined on all samples are represented in Table n°3. They show in accordance with existing literature, some relative variations of oil constituents depending on the cultivar, the geographical location and altitude.

The results obtained after analytical studies, performed on oils collected from crushing all drupes harvested at the black stage of full maturity (maturity sindex:4), showed that the fatty acid composition of some samples is significantly different (Table n°3 et Figures n°4 et n°5). For both varieties tested, the fatty acids composition has exhibited significant variation in the geographical sites considered. It is noted that there is a difference in the fatty acid composition between regions (rainfall, soil type) (Aparicio *et* Luna, 2002), concerning the oleic and linoleic fatty acids. The oleic acid increases with increasing altitude, and when we move from North to the South exposure and decreases with increasing temperature (Rondanini *et al*., 2011).




**Figures n°4 and n°5:** Influence of geographical site on fatty acid composition of the olive oil extracted from the Picual and Moroccan Picholine varieties.

This rate is in the order of 76.76% in the North and reached 78.65% in the South, passing through intermediate values in Mjaara (71.44%) and in Sidi Bousber (70.0%).

The opposite tendency is noted for linoleic acid, which is in the order of 15.867% in the North and decreases to 3.378% in the South, passing through the values 8.896% in Masmouda and 11.03% in Mjaara regions. Whereas, palmitic, palmitoleic and stearic acid, presents significant changes and tend to increase from north to south. In Spanish varieties of Andalusia (Picual, Hojiblanca), fatty acids (C16:0, C17:0, C18:0) decreases with enhancement of altitude (Ferreiro et Aparicio, 1992).

Similar results were obtained relating to the profile of the fatty acid composition and their variation depend on Moroccan growing regions (Zarrouk, 1984). This author has not noted in the Moroccan Picholine significant differences between North and South, in terms of the oleic acid (C18:1) and the linoleic acid (C18:2). Furthermore, we found that proportions of palmitic, palmitoleic, stearic and oleic acid, increase with increasing the altitude, and on the opposite, linoleic acid decreased slightly (Zarrouk, 1984). The varieties Sigoise, Picholine of Languedoc, Arbequine and Coratina recorded a high oleic acid, and low palmitic and linolenic acid content, in the original site, but these varieties are grown in Tunisia (Sfax). These latter have given inverse proportions in these considered fatty acids (Zarrouk *et al.*, 2008). In the NW of Argentina, a negative linear relationship was observed between the oleic and linoleic acid ($r^2$:0.90) and between oleic acid and palmitic acid ($r^2$:0.78).

Thus the varieties with low oleic acid levels (Arbequine, Arauco), showed high levels of linoleic and palmitic acids.

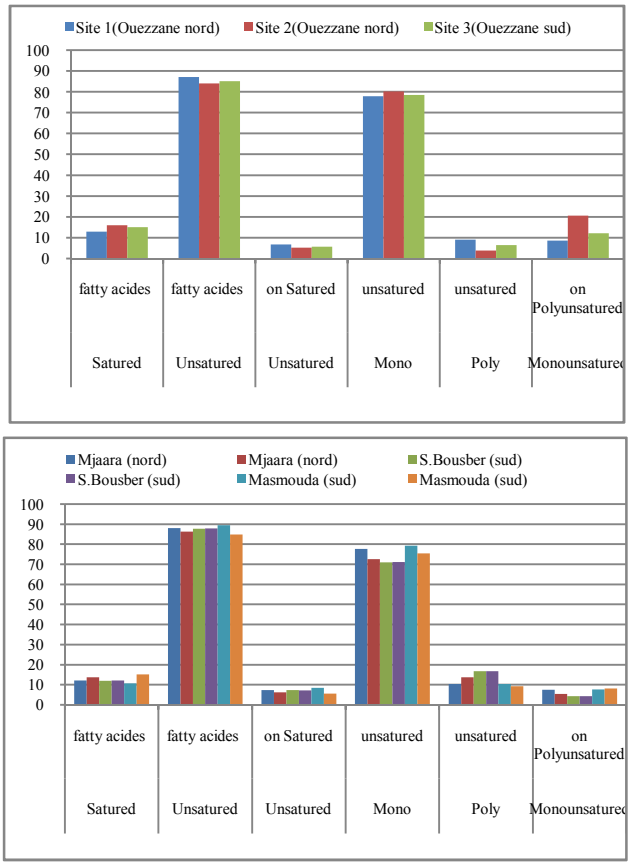
Inversely, varieties with high oleic acid contents (Picual, Changlot, Coratina) had low levels of linoleic and palmitic acid (Diraman, 2010; Rondanini *et al.*, 2011; Homapour *et al.*, 2014).

**Tableau n°4** Composition of saturated and unsaturated fatty acids of olive oil issued from the varieties Picual and Morrocan Picholine according to geographical locations.

| Varieties and geographical sites | Saturated fatty acid (SFA) | Unsaturated fatty acid (UFA) | UFA/ SFA | Mono usaturated (MUFA) | Poly unsaturated (PUFA) | MUFA/ PUFA |
|---|---|---|---|---|---|---|
| **-Picual** | - | - | - | - | - | - |
| Site 1(Ouezzane north) | 12,97 | 87,03 | 6,71 | 77,91 | 9,12 | 8,54 |
| Site 2(Ouezzane north) | 15,97 | 84,10 | 5,266 | 80,19 | 3,91 | 20,51 |
| Site 3(Ouezzane south) | 14,996 | 85,07 | 5,67 | 78,55 | 6,46 | 12,16 |
| **- Moroccan Picholine** | - | - | - | - | - | - |
| Mjaara (north ) | 12,046 | 88,058 | 7,31 | 77,619 | 10,439 | 7,435 |
| Mjaara (north ) | 13,716 | 86,284 | 6,291 | 72,635 | 13,649 | 5,392 |
| S.Bousber (south) | 12,026 | 87,699 | 7,292 | 71,019 | 16,679 | 4,258 |
| S.Bousber (south) | 12,172 | 87,826 | 7,215 | 71,149 | 16,677 | 4,266 |
| Masmouda (south) | 10,614 | 89,522 | 8,434 | 79,232 | 10,29 | 7,699 |
| Masmouda (south) | 15,169 | 84,838 | 5,593 | 75,508 | 9,329 | 8,094 |

The positive effect of the action of oleic acid in olive oil, more resistant to heat, increases the oxidative stability, the antihypertensive activity and reduce the risk of cardiovascular diseases (total cholesterol, LDL levels, triglycerides, blood pressure, oxidative stress, diabetes, arthritis, antiatherogenic, anti-inflammatory, anticancer, antimicrobial effects) (Pinelli *et al.*, 2003; Boggia *et al.*, 2005; Boskou *et al.*, 2006; Covas *et al.*, 2007; Saitta *et al.*, 2009; Cicatelli *et al.*,3013;Moreno *et al.*, 2015), the production of arachidonic acid-derived (prostaglandins), plays a key role in the apparition and development of cancerous tumors (COI, 2011; Psaltopoulou *et al.*, 2004; Jakobusic *et al.*,2015).

Our results reveal that Ouezzane region is very rich in SFA and MUFA and less rich in PUFA. On the contrary, Sidi Bousber region has proven to be very rich in UFA and MUFA and less rich in PUFA. The increment of temperatures in the environment (mesoclimate) causes a reduction of the oils saturated fatty acid content (Ranalli *et al.*, 1999; Kiralan *et al.*, 2009). More the content of PUFA of vegetable oils is high, more their sensitivity to heat is intense and fast (COI, 2011 *et* 2013).




**Figures n°6 and n°7** Composition of saturated and unsaturated fatty acids issued from varieties Picual and Moroccan Picholine according to the geographic location.

The oil of colder areas has a high proportion of oleic acid and higher UFA/SFA ratios (Fiorino *et* Grifi, 1991; Kiritsakis, 1993). In Greek oils, the high stearic (3.2%) and palmitic acid content were used as a factor of varietal distinction and determination of the origin geographical area of olive oil produced (Gigliotti *et al.,* 1993) due to their discriminant efficiency for the characterization of olive varieties (Alessandri, 1993).

## CONCLUSION

Overall, the analyzed cultivars have shown performances in the composition of fatty acids significantly different from the standard, depending on the geographical area and indicating a certain level genetic of variability reflected by the important effect of the varietal factor related to the genotype on the chemical composition of virgin olive oil that discriminate it from other analyzed oils. The peculiarities reside in the diversity of its organoleptic characteristics representatives of the different terroirs, associating to each region a specific mono-cultivar (only one variety) in research of authenticity (Cicatelli *et al.*, 2013).

This character allows from the one part to select a largest number of varieties and local types, greatly adapted to the condition of their cultivation areas, able to give high yield oil with good quality and improved fatty acid composition which is more abundant in oleic acid and MUFA and rich in natural antioxidants such as (polyphenols, tocopherols, fat-soluble vitamins E, K). From the other hand, it can be useful for the creation of future olive orchards and introduced in the national collection. Thus the Bouchouk Laghlide, Picual varieties, Oleaster types (BM2, BM3, BM4), Moroccan Picholine type M1, gave a high oleic acid (77.11%-78.65%) and MUFA rates (78.25%-79.23%), while Bouchouk Rkike, Bakhboukh Beldi, BMK, BMR, M6, G9, G10, S1, S2, Dahbia, Picholine of Languedoc, Ascolana Tenera, Gordal, presented significant percentage of oleic acid (69.68%-76,57%) and MUFA (71.02%-77.62%). The Bouchouika, BMM and Manzanille varieties had moderate values of oleic acid (65.15%-66.91%) and MUFA (66.87%-68.73%).

This has allowed in recent years to define a geographical indication of origin (extra virgin olive oil of Ouazzane) (Rahmani, 2014) and reinforces to think about the geographical indications (Masmoudia oil, Ouazzania oil, Mezgueldia oil, Settia oil) as likely to valorize the regional products, basic food as a main source of energy and essential unsaturated fatty acids for the human body, symbol of nobleness and emblematical in this region and a healthy lifestyle to save for future generations, through education, training, information and promotion (Hannachi *et al*., 2007; Lavee, 2015). It has suggested that the evaluation of other chemical composition (pigments, tocopherols, phenols..) and organoleptic qualities of Ouazzanian olive oils seemed necessary (Homapour *et al.,* 2014).

Consumer greater interest in the Mediterranean typical diet (placed on UNESCO list of Intangible Cultural Heritage of Humanity), therapeutic and nutritional virtuousness and beneficial promoting properties recognized on human health , related to the organs of the olive tree (leaves, fruit, oil), have created a renewed interest in the cultivation of the olive tree, resulting in new extensions of plantations with various and interesting varieties, but little exploited, a renovated and recent technology, a cuvees of special or monovarietal exclusive olive oils, which correspond to a recent request of informed consumers (Lavee, 2015; Moreno *et al.,* 2015; Montero, 2015), that putted the olive oil sector in a competitiveness environment where only the quality, notoriety and the ultra premium distinction of olive oil are paramount. The current challenge for profitable olive cultivation and of a high or excellent quality, requires the evaluation of consumer demand for olive oil in the traditional market and on the creation of interesting opportunities in emerging and potential local markets, with an integrated production-marketing approach to give more visibility to promote the consumption and of competitive olive oil quality, to which the consumer is fiercely attached (Cicatelli *et al.,* 2013; Eric, 2015; Jakobusic *et al.,* 2015).

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