

# Research Journal of Pharmaceutical, Biological and Chemical Sciences

## Effect of artisanal conservation methods on the nutritional and sensory analysis of Moroccan Picholine.

Berrada W<sup>1,2\*</sup>, Errachidi F<sup>1,2</sup>, Alaoui Belrhiti A<sup>2</sup>, Dahmanis<sup>1,4</sup>, Lemjalladi<sup>1,3</sup>, Ghadraoui L<sup>2</sup>, Taouda H<sup>1,5</sup>, and Chabir R<sup>1</sup>.

<sup>1</sup>Laboratory of Physiopathology and Nutrition, Faculty of Medicine and Pharmacy, Sidi Mohamed Ben Abdelah University, BP 1893 Km 2200 Road of SidiHrazem, Fez, Morocco.

<sup>2</sup>Laboratory of Functional Ecology and Environment, faculty of sciences and technology, Sidi Mohamed Ben Abdellah University, po.box 2202 – route d'Imouzzer, Fez, Morocco.

<sup>3</sup>Laboratory of Applied Organic Chemistry Faculty of Science and Technology, Sidi Mohamed Ben Abdallah University, Fez, Morocco.

<sup>4</sup>Laboratory of Biological Tests, Food and Nutritional Transition Team, Faculty of Sciences, IbnTofail University, Kenitra.

<sup>5</sup>Laboratory of Bioactive Molecules, Faculty of Sciences and Techniques Saiss, Fez, B.P. 2202 Imouzzer Road, Fes, Morocco.

### ABSTRACT

This research shows the impact of artisanal conservation methods on nutritional and sensory qualities of some varieties of Moroccan table olives. We have selected and prepared 12 samples of Moroccan picholine variety belonging to three groups (green olives, red olives and black olives). Biochemical properties and sensory analysis were evaluated by principal component analysis (PCA). The results revealed that black olives had the highest levels of phenolic compounds (0,3812g/100 g) for (sample 2) vitamin C (0,0335g /100g) for (sample 4). Whereas, the green olives contain the strongest antioxidant activity (AA= 1 mg/ml) for (sample 1), vitamin C (0,0387g /100g) for (sample 2) and total sugars (avg= 2,475%). However, the lowest levels of vitamin C and antioxidant activity were noted in red olives. Principal components analysis has led deduce that there is a strong correlation between biochemical and sensory analysis. Although antioxidant activity show the existence of mathematical models between phenolic compounds concentration and antioxidant activity. Showed linear model for black olives, linear and exponential models for green olives and logarithmic model for red olives.

**Keywords:** sensory analysis, biochemical analysis, mathematical models, Moroccan Picholine and PCA.

*\*Corresponding author*

## INTRODUCTION

In Morocco olive production is carried out over a total area of 879,000 hectares, with an average harvest of 1.8 tons /hectares in 2017 with a 50 % increase over the previous season, which recorded only 1.2 T/ha [1].The cultivation has traditionally been based on planting a single cultivar so-called “Moroccan Picholine” occupying almost 90% [2].This autochthonous cultivar shows good adaptability to a wide range of Moroccan pedoclimatic conditions, and exhibits interesting agronomic traits, oil quality, and composition characteristics [3].We distinguish two conservation activities of olives: modern and traditional activity. The first conservation activity is provided by 68 industrial units with an overall capacity of approximately 131,500 T / year. They are located mainly in Marrakech and Fez cities. As regards traditional activity is a practice Widespread in Morocco, is difficult to identify and does not constitute a structured sector in identifiable units. But she is directly integrated into the retail trade and therefore serves primarily as a source of supply for the local market [4].Compared to other fruit, table olive, is distinguished by a characteristic aroma and flavor. These sensory characteristics, together with high nutritional value are the main features that have resulted in the increase of table olives consumption in recent years [5].Volatile compounds analysis can be used as an indicator to check table olives quality [6]. Fruit maturity, processing methods, geographic location and other parameters influence biochemical table olives composition [7]. The Moroccan Picholine is a source of valuable nutrients and bioactives and therapeutic interest [8]. .Olive fruit contains appreciable concentration (1–3 g/100g) of fresh pulp weight, of hydrophilic (phenolic acids, flavonoids and secoiridoids) and lipophilic (cresols) phenolic compounds that are known to possess multiple biological activities such as antioxidant, anti-carcinogenic, anti-inflammatory, antimicrobial, antihypertensive, anti-dyslipidemic, cardiogenic, laxative, and antiplatelet [9].Other important compounds present in olive fruit are pectin, organic acids, and pigments. Virgin olive oil (VOO), extracted mechanically from the fruit (10-25g/100g), and is also very popular for its nutritive and health-promoting potential, especially against cardiovascular disorders due to the presence of high levels of monounsaturated fatty acids[10 ; 11].Other valuable minor components present in olive fruit such as phytosterols, tocopherols, carotenoids (0.6–2.4mg/100g), chlorophylls (1.8–13.5mg/100g) and squaleneT [12].The objectives of the present study are the improvement table olives by traditional making. The investigation of chemical composition and sensory profile of artisanal-style (black olives, red olives and green olives). Moreover, we try to assess the presence of correlating biochemical analysis with the sensory profile through multivariate analyses (PCA).

## MATERIAL AND METHODS

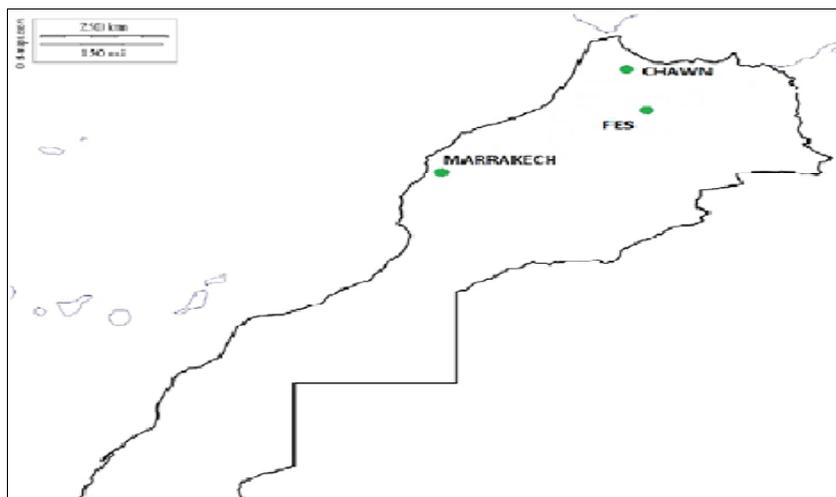
### Plant material

The present work was carried out on monovarietal olive fruits ‘oleaeuropaea’ from the Moroccan variety ‘Moroccan picholine’. The 12 samples of olives were collected and grouped to three pools (green table olive, red table olive and black table olive) and they were obtained from Fez, Marrakech and Chawn(table 1) between November and January 2017 (Figure 1). The olive fruits were hand-picked and 2 kg of olives were collected from the same trees in different stage of maturity. Only healthy fruits without any kind of infection or physical damage were selected. After harvesting, the olive fruits were immediately transported to the laboratory in cool bags.

**Table 1: Characteristics of the sampling sites**

sites	Longitude	laltitude	Bioclimatic stage	Pluviometry
Marrakech (BO)	7°59.5964	31°380496	Semi-arid	250 mm
Fez (GO)	5°0,0168	34°033130	Semi-arid	536 mm
Chefchawn (RO)	5°27449	35°17180	Humid	880 mm

BO: black olives; GO: green olives; RO: red olives.



**Figure 1: Geographical map of studied area**

**Olives processing**

Following a screening preservatives having antioxidant, anti-inflammatory and maximum antimicrobial effects. We have preserved our table olives with (2% Thyme, 2% Parsley, 1% Garlic and 1% Nutmeg) and lemon juice of 3% at temperature not exceeding 28°C. The four preparations are prepared as follows (table2).

**Table 2: Four preparations of different artisanal table olives**

Samples	Preparations for three types of table olives
1	Parsley-garlic-nutmeg.
2	Parsley-thyme- garlic.
3	Garlic-thyme-nutmeg
4	Parsley-thyme-nutmeg

**Sensory analysis**

The sensory profile of samples was performed by a trained panel, composed of 20 assessors (6 males and 14 females). They were recruited because of their major role in the implementation of table olives sensory analysis method, samples were served in dishes in laboratory of ‘Human Pathology, Biomedicine and Environment’, Team Nutrition faculty of Medicine and Pharmacy. In order to avoid any order effect the olives were presented in a random order to the assessors. Water cups was used for mouth rinsing between each sample evaluation. Assessors evaluate the Attributes (appearance, aroma, flavour and texture) of the list by scoring their intensity in an unstructured line scale (0–10), 0 was the lowest and 10 were the highest score.

**Biochemical Analysis**

The nutritive components of artisanal table olives of the 12samples determined according to validated analytical methods: Vitamin C content was determined following the method described by [12]. Carotenoid estimation was done according to analytical procedure by [13]. Antioxidant capacity determinations paste olive extract in water was determined according to the method of [14]. Phenolic compounds extracting in water was analyzed using the modified isolation method described by [15], °Brix and %salt analysis were assessed using a digital refractometer (Krussoptronic, DR 301-95, Germany).

**Statistical analysis**

Experimental data was statistically analysed using PCA (Past Software); correlation analysis were done according to Pearson method and analysis of variance by Anova.

**RESULTS AND DISCUSSION**

**Biochemical and physicochemical analyses**

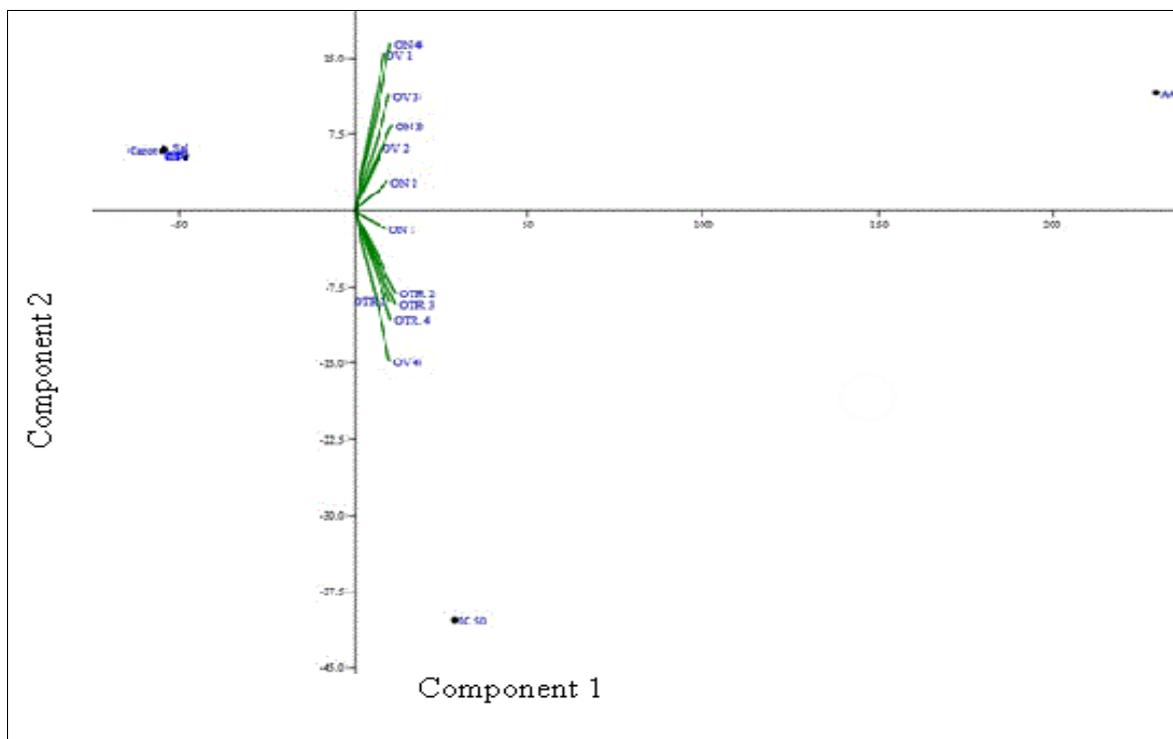
The values of phenolic compounds, vitamin C, carotenoid, antioxidants activities (IC 50), total soluble solids (°Brix) and % salt of all these samples analysed are summarized in (table 3). The results show that the black table olives from the city of Marrakech have the highest average concentration of phenolic compounds (0.37g / 100g), vitamin C (0.02g / 100g) and IC 50 (15.5g / 100g). Followed by green table olives from the Fez city which are rich in carotenoids (0.0029g / 100g) and vitamin C (0.02g / 100g). Concerning red table olives from the Chefchaoun location presented a high rate in % salt (2.625) and Brix (2.575g/100). We also deduced for the three types of olives compared to their biochemical test that the black table olives presented by the (samples 2 and 4) which showed the highest rate, followed by (samples 2 and 3) for green table olives and (samples 3 and 4) for red table olives. These results are relatively similar to [16;9], who they shown that the phenolic compounds of olive increases during ripening. Whereas [17] show that the green color of the tissue is caused by chlorophylls and carotenoids pigments and their concentrations fall progressively during ripening.

**Table 3: Quantitative distributions of phenolic compounds, vitamin C, carotenoids, antioxidants activities, ° brix and % salt in artisanal table olives**

artisanal olives	Samples	P.C*** (g/100g)	Vitamin C* (g/100g)	Carotenoids*** (g/100g)	% inhibition	AA** (mg/ml)	% salt	° Brix***
Green olives	1	0.2475	0.0088	0.00206	70.6564	1	3.1	3.3
	2	0.2301	0.0387	0.00317	60.6178	10	2.4	2.1
	3	0.2307	0.0247	0.00394	83.7838	10	2.3	2.2
	4	0.2182	0.0247	0.00256	81.0811	43,5	2.5	2.3
	Average	0.23	0.02	0.0029	74.03	16.125	2.575	2.475
Redolives	1	0.2398	0.007	0.000681	81.8533	36	2.4	2.3
	2	0.2503	0.007	0.000473	95.7529	39	2,6	2.5
	3	0.2707	0.0176	0.000573	94.2085	40	2,8	2.7
	4	0.2428	0.0018	0.000704	84.1699	39	2,7	2.8
	Average	0.25	0.01	0.00061	89	38.5	2.625	2.575
black olives	1	0.3704	0.0141	0.000145	72.973	24	1.8	1.7
	2	0.3812	0.0088	0.000581	77.6062	19	1.4	1
	3	0.3795	0.0194	0.000276	89.1892	15	1.1	0.7
	4	0.3411	0.0335	0.000158	87.2587	4	1.2	0.7
	Average	0.37	0.02	0.00029	81.76	15.5	1.375	1.025

\*\*\*Significance at P< 0,001; \*\*Significance at P<0, 01; \*Significance at P<0, 05. Values are given as mean of three repetitions standard deviation; (1,2,3,4) Green olives of Fes ; (1,2,3,4) Red olives of Chawn ; (1,2,3,4) Black olives of Marrakech ; PC :Phenolic compounds; IC 50 Inhibition concentration 50% of the extracts.

Variance analysis of these assays in the three types of olives shows that overall the constituents present variability in the artisanal samples studied. To have a dimensional view of the analysis obtained on the three types of olives, we used the principal components analysis PCA of all merged data done on several types of olives belonging to the three types of table olives (green olives, red olives and black olives). The results obtained are summarized in (figure 2).



**Figure 2. Principal component analysis (PC1 and PC2) of artisanal table olives**

Analysis shows that black table olives and green table olives are grouped and contribute positively to component 2 (PC2). The red olives are also grouped in individual pool and contribute negatively to component 2 PC2. These distributions show that the biochemical analysis carried out are able to group the table olives and to characterize them according to their biochemical contents. Antioxidants activity(AA) contribute to the positive part of component 1 and come into association with black table olives and green table olives. IC 50 contribute negatively to PC2 and strongly correlate with red olives. In summary the PCA study confirms the strong correlation, which was showed by a good separation between the antioxidant activity and the two types of olives (black table olives and green table olives).These results are in accordance with [18] who worked on another matrix (animal), to characterize dry-cured hams from four different processing methods and she showed a good separation among groups by principal component analysis evaluated by physico-chemical characteristics.

Panel sensory example is a group of individuals who have been specifically trained to recognize describe, and quantify sensory characteristics of the three olives tested (green, red and black olives) within the normal range of sensitivity, individuals can very significantly in their sensory perceptions. The values are the average taster’s notes for the twelve artisanal samples prepared (table 4).

**Table 4: The average taster’s notes for the twelve artisanal samples prepared.**

Samples preparation / Descriptors	1	2	3	4	5	6	7	8	9	10	11	12
Odorintensity	5,529	6,294	5,824	5,824	5,941	5,824	5,765	6,118	6,765	7,294	7,059	6,941
Pleasantodor	5,235	6,412	5,941	5,529	5,294	6,118	5,471	6,412	5,824	6,824	6,647	6,235
Color	4,588	4,706	5,176	4,941	7,412	7,412	6,235	6,765	8,000	8,412	8,235	8,000
Clearness	5,412	5,235	5,118	5,235	7,412	7,471	6,588	7,176	8,000	8,412	8,412	8,118
Brightness	5,000	5,471	5,824	4,706	6,647	6,765	6,176	6,471	7,647	7,118	7,765	6,588
Aromaappreciation	5,412	6,059	5,765	5,588	5,706	6,000	6,412	6,824	6,059	6,765	6,588	5,647
Bittering	5,706	5,706	5,882	5,353	6,059	6,000	5,941	6,235	6,588	6,588	6,588	6,000
harshness	5,941	5,941	6,294	6,412	4,824	5,000	4,882	5,353	6,294	6,059	6,118	5,235
Crisp chair	5,824	6,176	5,647	5,706	3,882	4,059	4,000	3,941	4,706	4,765	4,882	4,706
Formsmooth	6,588	6,235	6,000	5,765	6,882	6,176	6,353	6,412	3,882	4,412	4,647	4,941
Acidity/salt	6,471	6,294	6,059	5,765	5,529	5,706	6,000	6,588	5,353	6,118	5,353	4,588
Pleasantflavor	6,118	6,647	6,235	5,765	5,765	5,882	5,882	7,059	6,059	6,824	5,941	5,882
Acidity	4,941	5,059	6,294	5,647	6,471	6,176	6,000	6,353	5,176	5,529	4,765	4,529
Bitterness	3,059	2,941	3,471	2,765	3,588	4,176	5,176	4,294	4,176	4,882	4,706	4,824
Astringency	2,471	2,235	2,294	2,412	2,765	2,647	2,471	2,353	2,941	3,059	3,235	3,294
Pleasantafter taste	5,882	5,353	4,824	4,529	5,765	5,765	5,471	6,294	5,882	6,706	6,000	5,824

**Principal component analysis of sensory attributes of artisanal table olives.**

Statistical analysis of sensory panel data can be carried out to produce a consensus opinion of the group’s perceptions. This is one of the main benefits of using a panel, as an individual, due to low personal sensitivity, may on occasion overlook a particular attribute that is of importance to the consumer [19].

The principal component analysis (figure 3) of sensory attributes of artisanal table olives are grouped in three pools. The first is formed by Green table olives (sample 1, sample 2, sample 3, and sample 4) from Fes city, are show that (harshness and crisp chairs) are strongly correlated by contributory negatively to component 2 PC2. The second pool is formed by red table olives (sample 5, sample 6 and sample 8) from the Chefchaoun region. Are show that (Clearness, Form smooth, appreciation of aroma, bitterness, acidity and pleasant after taste) are strongly correlated and contributed negatively to component 2 PC2. The third pool is formed by black table olives (sample 9, sample 10, sample 11, and sample 12) from the Marrakech city show that (odor intensity, pleasant odour, brightness, bittering and astringency) are strongly correlated and contribute positively to component 1 (PC1).This result is in agreement with [18] who she showed a good separation between aroma and sensory analysis by principal component analysis. Besides in disagreement with [20] who applied (PCA) to sensory data and volatile compounds show a poor separation of samples according to cultivars.

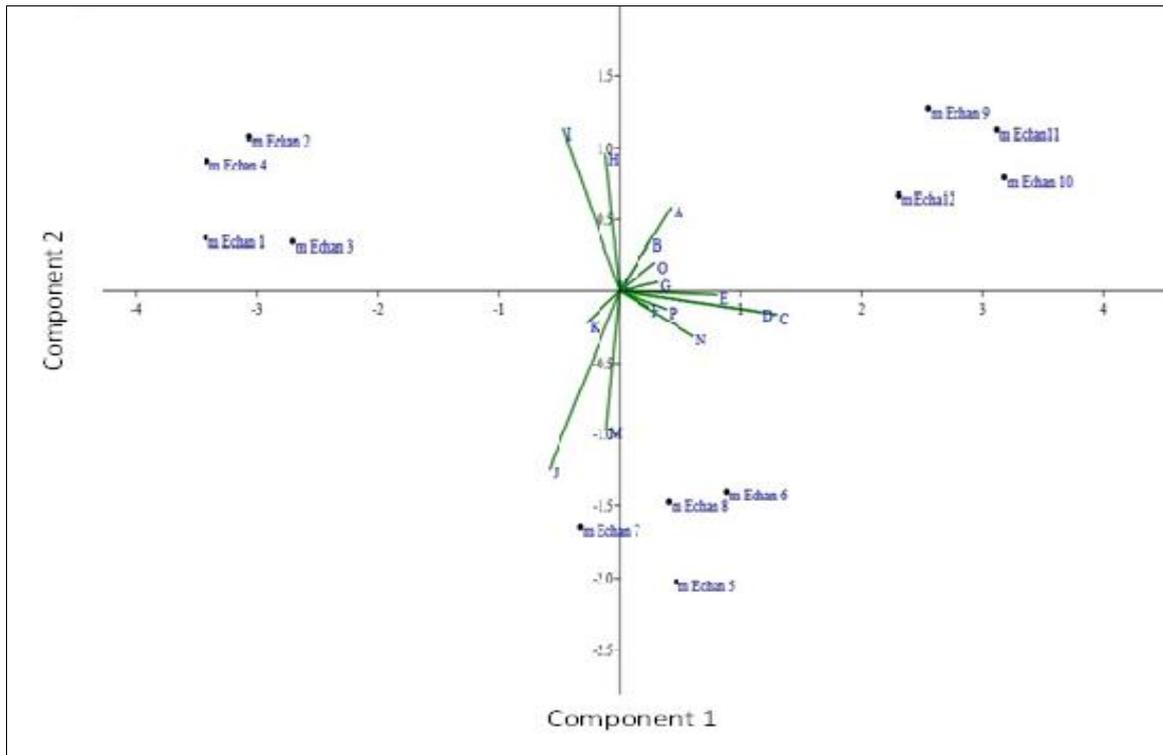


Figure 3. Principal component analysis (PC1 and PC2) of sensory attributes of artisanal table olives.

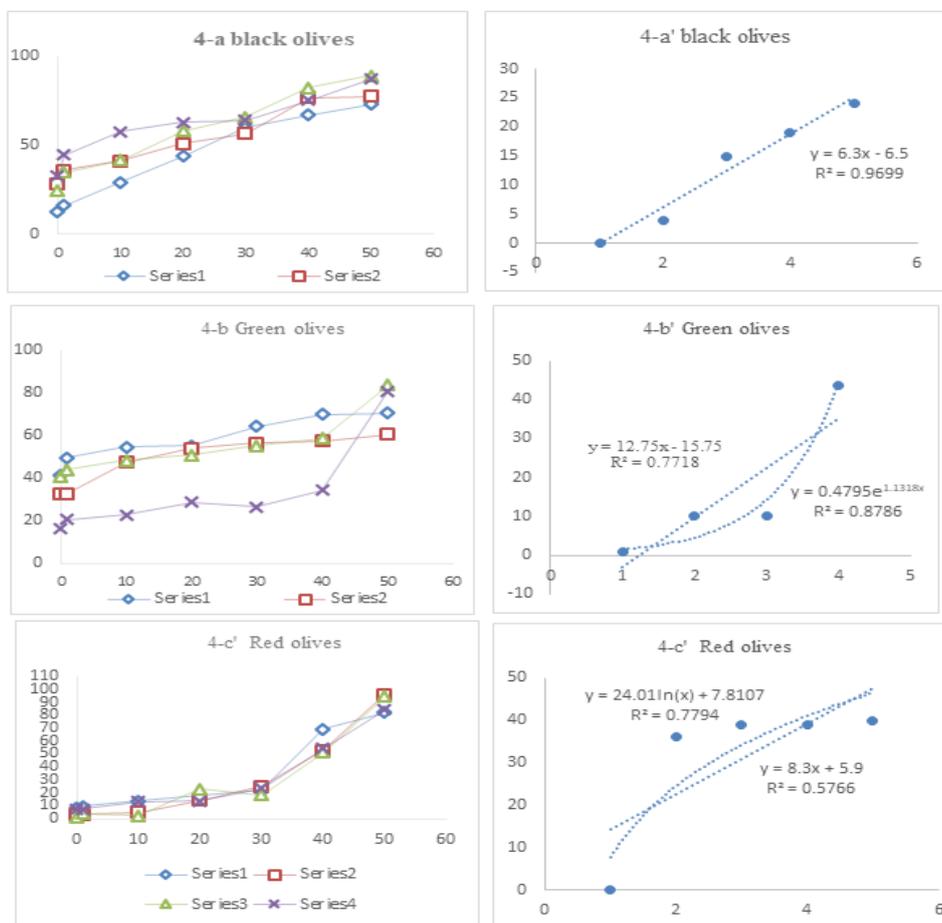
Table 5: Mathematical models between AA and phenolic compounds of twelve series to green, red and black olives

Olives	Series	Mathematical models	Coif.	Nature
Red olives	Series 1	$Y = 8.273e^{0,045x}$	$R^2 = 0.945$	exponential
	Series 2	$Y = 3.075e^{0,069x}$	$R^2 = 0.993$	
	Series 3	$Y = 1.787e^{0,083x}$	$R^2 = 0.879$	
	Series 4	$Y = 6.665e^{0,049x}$	$R^2 = 0.967$	
Black olives	Series 1	$y = 1.246x + 16,30$	$R^2 = 0.974$	Linear
	Series 2	$y = 0.975x + 31,51$	$R^2 = 0.962$	
	Series 3	$y = 1.246x + 29,85$	$R^2 = 0.979$	
	Series 4	$y = 0.894x + 41,45$	$R^2 = 0.913$	
Green olives	Series 1	$y = 0.533x + 46,54$	$R^2 = 0.915$	Exponential and Linear
	Series 2	$y = 0.554x + 36,90$	$R^2 = 0.842$	
	Series 3	$y = 0.665x + 40,26$	$R^2 = 0.820$	
	Series 4	$y = 17.19e^{0,024x}$	$R^2 = 0.806$	

**Correlation between antioxidant activity and inhibition concentration 50% of artisanal table olives**

The highest antioxidant potential was observed respectively in black table olives, green table olives and finally in red table olives. The figure 4-a-a' shows a linear model for antioxidant activity and a linear correlation between phenolic content and antioxidant activity. The figure 4-b-b' shows a linear and

exponential model antioxidant activity and a linear and exponential correlation between phenolic content and antioxidant activity. Figure 4-c-c' show the exponential model and logarithmic correlation between phenolic content and antioxidant activity.



**Figure 4: a-b-c Correlation between PC and inhibition concentration 50%; a'-b'-c' Correlation between antioxidant activity and phenolic compounds**

Results obtained show the existence of linear, logarithmic and exponential correlation between antioxidant activities and phenolic compounds. Moreover Pearson's correlation between DPPH assay and total phenol content ( $R^2=0.969$ - $R^2=0.878$ ;  $P<0,001$ ) showed the highest antioxidant capacity (Figure 4). These result are in agreement with [21] who found a good antioxidant activity of green olives by DPPH and ABTS assay and [22] was observed very high correlation between total phenols and antioxidant capacity measured by four methods (TRAP-DPPH- $\beta$  carotene and TAA) of some Spanish olive oils.

The mathematical analysis models of the antioxidant activities (expressed in% inhibition) and their correlation with the concentration in phenolic compounds shows linear, exponential models relations and existence a difference in the coefficient of direction equations. ( $a=1.246$ ) for black olives and ( $a=0.665$ ) for green olives and ( $a=8.273e^{0,045x}$ ) for red olives (Figure5).

### CONCLUSION

Biochemical and sensory analysis show that the olive type is an important factor for this fruit. Black olives show having phenolic compounds and antioxidant activity a high content, the best correlation between antioxidant activity and phenolic compounds concentration, appreciated by assessors and having a linear model. Whereas green olives with high content in vitamin C, carotenoids and a linear and exponential models. Finally red olives show a high content of sugars and salt with exponential models and logarithmic correlation

between phenolic content and antioxidant activity. The results confirms the strong correlation between biochemical and sensory analysis in artisanal olives.

#### REFERENCES

- [1] Morocco World News, (2017). Olive Production for 2017-2018 Season Breaks Morocco's Record.
- [2] Mahhou A., Jermmouni A., Hadiddou A., Oukabli A., Mamouni A. , 2014. Période de Récolte et caractéristiques de l'huile d'olive de quatre variétés en irrigué dans la région de Meknès. Rev. Maroc. Sci. Agron. Vét. 2:5–15.
- [3] Essadki M., Ouazzani N., Lumaret R., Moumni M., 2006. ISSR variation in olive-tree cultivars from Morocco and other Western countries of the Mediterranean basin. Genet. Resour. CropEvol. 53:475–482.
- [4] USAID Maroc, 2017. Rapport américaine pour le developpement international, élaboré par chemonics international (guide olive table AAI).
- [5] IOOC Home Page, 2011. (International Olive Oil Council Activities: World Olive Oil Figures: World Olive Oil Consumption).
- [6] Angerosa, F., 2002. Influence of volatile compounds on virgin olive oil quality evaluated by analytical approaches and sensor panels. *Eur. J. Lipid Sci. Technol.* 104, 639–660.
- [7] Baccouri, O., Bendini, A., Cerretani, L., Guerfel, M., Baccouri, B., Lercker, G., Zarrouk, M., Miled, D.D.B., 2008. Comparative study on volatile compounds from Tunisian and Sicilian monovarietal virgin olive oils. *Food Chem.* 111, 322–328.
- [8] Ribarova, F., Zanev, R., Shishkov, S., Rizov, N., 2003.  $\alpha$ -Tocopherol, fatty acids and their correlations in Bulgarian foodstuffs. *J. Food Compos. Anal.* 16, 659–667.
- [9] Hui Y.H., Barta J., Pilar Cano M., Gusek T.W., Sidhu J.S., Sinha N.K., 2006. Handbook of Fruits and Fruit Processing. 490-688.
- [10] Covas, M.I., Nyyssonen, K., Poulsen, H.E., 2006. The effect of polyphenols in olive oil on heart disease risk factors. *Ann. Int. Med.* 145, 333–431.
- [11] Covas, M.I., 2008. Bioactive effects of olive oil phenolic compounds in humans: Reduction of heart disease factors and oxidative damage. *Inflammopharmacology* 16, 216–218.
- [12] Barros L, Carvalho A.M, Ferreira I.C.F.R., 2010. Leaves, Flowers, Immature fruits and leafy flowered stems of *Malvasylvestris*: a comparative study of thenutraceutical potential and composition. *Food Chem. Toxicol* 48, 1466–1472.
- [13] Nayek, S., Choudhury, I.H., Jaishee, N., Roy, S., Resea, J., 2004. *Chemi Sci.* (2014) 63-69.
- [14] Jeong S.M., Kim S.Y., Kim D.R., Jo S.C., Nam K.C., Ahn D.U., Lee, S.C., 2004. *J. Agric. Food Chem.* 52, 3389-3393
- [15] Barros L., Calhelha, R.C., Vaz J.A., Ferreira I.C.F.R., Baptista, P., Estevinho, L.M., 2007. *Eur. Food Res. Techn*, 225, 151-156.
- [16] Ollivier, D., Boubault, E., Pinate, Ch., 2004. Article original Annales. Des falsifications, de l'expertise chimique et toxicologique, 2ème Semestre N° 169-196.
- [17] Roca M., M'inguez-Mosquera M.I., 2001. Changes in chloroplast pigments of olive varieties during fruit ripening. *J. Agric. Food Chem.* 49:832–839.
- [18] Petričević, S., Radovčić, N.M., Lukić, K., Listeš, E., Medić, H., 2018. Differentiation of dry-cured hams from different processing methods by means of volatile compounds, physico-chemical and sensory analysis. *Meat Science* 137, 217–227.
- [19] Jack, F.R., 2003. Development of guidelines for the preparation and handling of sensory samples in the Scotch whisky industry. *Journal of the Institute of Brewing* 109 (2), 114–119.
- [20] López-López, A., Higinio Sánchez A., Cortés-Delgado, A., de Castro, A., Montaño, A., 2018. Relating sensory analysis with SPME-GC-MS data for Spanish-style green table olive aroma profiling. *Food Science and Technology*, doi: 10.1016/j.lwt.2017.11.058.
- [21] Piscopo, A., De Bruno, A., Zappia, A., 2014. Marco Poiana. Antioxidant activity of dried green olives. *LWT - Food Science and Technology* 58, 49-54.
- [22] Gorinstein, S., Martin-Belloso, O., Katrich, E., Lojek, A., Gligelmo-Miguel, M., Haruenkit, R., Park, Y.S., Jung, S.T., Trakhtenberg, S., 2003. Comparison of the contents of the main biochemical compounds and the antioxidant activity of some Spanish olive oils as determined by four different radical scavenging tests. *Journal of Nutritional Biochemistry* 14, 154–159.