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Physicochemical Characterization of Vegetable Oil Extract from Yellow Onion AMPOSTA of Meknes Region in Morocco

Saadia. Boulfane[1], Mehdi Taib[1], Abdellah Anouar[1], Said Hilali[2], Lahboub Bouyazza[1]
[1]Laboratory of Applied Chemistry and Environment, Hassan 1st University, Faculty of Sciences and Technologies, Settat, Morocco.
[2]Laboratory of Biochemistry, Neurosciences, Natural Resources and Environment; Hassan 1st University, Faculty of Science and Technology, Settat, Morocco.

***Abstract*:- This work aims to determine the physicochemical characteristics and the fatty acid, sterols and tocopherols from vegetable oil obtained by extraction with hexane from yellow onion seeds Allium cepa L. in the Boufkrane region of Meknes Morocco. The evaluation of the physicochemical parameters of the yielded the following results: acid number: 1 %; and peroxide value: 2 meq active O_2 / kg oil. The GC analysis of the acidic composition shows a predominance of linoleic acid (52.1 %) followed by oleic acid (32.3 %) and the sterolic composition reveals a predominance of β-sitosterol (63.7 %) as well as the presence of Campesterols and Δ–5 Avenasterol with percentages of 15.9 % and 10.6 % respectively. Finally, the analysis of tocopherols of this oil by HPLCUV gives a level of 1511 mg/Kg of total tocopherols with 84.9 % of α-tocopherols.**

Key words: Fatty acid, sterol, Allium cepa, α-tocopherols, β-sitosterol, linoleic acid.

I. INTRODUCTION

Onions are one of the most consumed vegetables around the world and in particular in Morocco. The bulb, the stem, the leaves are the most consumed parts, while the seeds are used very little. The seeds are currently considered a source of vegetable oil used in cosmetics and aromatherapy **[1]**. These oils are extracted from the seeds, kernels or pulp, either by nonpolar solvents such as hexane or petroleum ether **[2]**, or by cold mechanical pressure **[3]** or by supercritical extraction **[4]** and are obtained with a yield of 9 % to 45 %.

Each vegetable oil is characterized by its chemical composition of fatty acids, tocopherols and sterols which varies from species to species. The most abundant essential fatty acid in the vegetable oil of onion seeds is linoleic acid (ω-6) which represents an energy source and precursor of molecules playing an important role in inflammation such as prostaglandins or on platelet aggregation followed by oleic acid **[5, 6]**. The vegetable oils of onion seeds contain a high content of α, β and γ tocopherols **[6, 7]**, which have an important antioxidant property **[8]** and act as a masking agent and skin care agent.

This work falls within the framework of contributing to the valuation of a plant material the seeds of the yellow valence onion (Allium cepa L.) **[9]** from Morocco by extracting their vegetable oil in order to characterize it and determine its composition in fatty acids, tocopherols and sterols.

II. MATERIELS AND METHODS

II.1. Plant Material

The lot of the seeds of yellow onion Amposta was harvested in the month July 2020 at Boufkrane located in the region of Meknes in northern Morocco. The seeds were separated from their envelopes, freed of all impurities, dried in the sunlight, and then they are placed in an oven for 6 hours at 55ºC, then they were finely ground and conditioned at 25°C in a sealed vial before extracting the oil.

II.2. Extraction of oil

The oil was extracted from freshly crushed seeds, is placed in a cellulose cartridge. The oil was extracted with hexane in a Soxhlet apparatus. After 6 h of extraction, the oil was collected under vacuum and stored in pillboxes.

II.3. Physico-chemical characterization of the oil

II.3.1. Determination of free acidity

The free acidity, expressed as a percentage of oleic acid, was determined by 1 g of olive oil dissolved in 50 ml of ethanol. The mixture was titrated by a solution of potassium hydroxide 0.1 N in the presence of phenolphthalein **[10]**. The assay was carried out three times.

II.3.2 Determination of the peroxide value

1g of oil is dissolved in 12.2 ml of the acetic acid/chloroform mixture 3 : 2 (v/v). 15 ml of a saturated potassium iodide solution is added to the mixture. The latter is placed in the dark for 5 min. Then 60 ml of distilled water and 1 ml of a starch solution are added (a purple color appears). The resulting mixture was titrated with a 0.01N sodium thiosulfate solution **[11]**. The dosage was carried out thrice.

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II.4. Determination of fatty acid composition by Gas Chromatography

The methyl-esters of the fatty acids are obtained by the action of methanol in alkaline medium of glycerides and free fatty acids, according to the protocol of standard NF ISO 129666 - 2 NF ISO129666 - 4. To 0.1 g oil one adds 2 ml of heptane and 0.2 ml of of 2N methanolic potash. After agitation during 30 seconds, the heptanic higher phase is recovered. The methyl esters of fatty acids obtained were analyzed by gas chromatography HP (model 6890) equipped with a flame ionization detector (T = 260 ° C). The column used is a capillary column Carbowax type size 30 m x 0.32 mm x 0.25 µm. The carrier gas was nitrogen at a flow rate of 2.5 ml / min. The oven temperature program was 140 ° C to 200 ° C, from 210 to 245 ° C with a gradient of 10 °C / min every 10 min. The identification of the peaks was carried out in the presence of standard compounds and various percentages of fatty-acids were calculated by means of an automatic integrator. All the analyzes were carried out in triple and the results are expressed by averages and standard deviations.

II.5. Determination of total sterols

The sterol fraction was determined according to the method described by the COI **[12]**. After saponification of the oils with potassium hydroxide in ethanolic solution while using α - cholestanol as an internal standard. The unsaponifiable was then extracted with ethyl ether. The sterol fraction was separated from the unsaponifiable extract by thin layer silica gel chromatography. The sterols recovered from the silica gel were transformed into trimethylsilylethers and analyzed by gas chromatography using an Agilent type chromatograph equipped with an HP 5 capillary column (5% diphenyl and 95% dimethylpolysiloxane) of 30 m long, 0.25 mm inner diameter and 0.1 µm film diameter. The temperature of the oven, the injector and the detector is 300 ºC. The gas vector is hydrogen with a flow rate of 40 ml / min. The volume injected was 1 µl. The identification of the peaks was carried out in the presence of the witnesses and the calculation of the different percentages of sterols was done using an automatic integrator.

II.6. Determination of the composition of tocopherols content

The unsaponifiable fraction of the yellow onion Amposta oil was analyzed by normal phase HPLC **[13]** to determine the tocopherols content. A solution prepared from 20 mg of oil per milliliter of hexane and isooctane (99 %) / 2-propanol (1 %) filtered through a millipore filter 0.45µm diameter. The manual injector is provided with an injection loop of 20 µl and the column is K.romasil100 SIL parameters (C18, 5µm, 4.6 x 250 mm). The solvent mixture in the isocratic conditions consisted of hexane and isopropanol to HPLC (99 : 1% v : v). The flow of the eluent was 1ml/min and pressure of 33 bar with a fluorescence detector at a wavelength of 290-330 nm. Peaks were identified by injection of tocopherols standards (Sigma Aldrich product). The calibration curves were plotted by taking a dilution range from 0.3 to 8 mg /ml.

III. RESULTS AND DISCUSSIONS

III.1. Physicochemical characteristics of oil from Amposta yellow onion seeds.

Acidity index: it is expressed as a percentage of oleic acid, the assay gave the following result: 0.1± 0.01 %.The value found for free acidity is lower than that reported by Aiboudi et al. who obtained the value 1.55 % **[5]**. The very low value of free acidity can be explained by the good extraction and conservation of this oil.

Peroxide value: the result is expressed in milliequivalents of active oxygen per kilogram of oil (meq active O_2 / kg oil). The value of the peroxide value found is 2 ± 0.01 meq active O_2 / kg oil. This found result clearly explains the good conservation of this oil after its extraction. Indeed, the peroxide value of an oil reflects its oxidation state. The higher the value, the more peroxidised it is and thus the lower its keeping qualities **[14]**.

The values found for the acid number and the peroxide number meet the standards of Codex Alimentarius **[15]**.

III.2. Fatty acid composition of the oil extracted from the seeds of yellow Amposta

The results of the gas chromatography are shown in the tables below:

Table 1: Percentage of fatty acid of the oils studied

Acide gras	Percentage %
Myristic acid C14:0	0.1
Palmitic acid C16:0	7.8
Palmitoleic acid C16:1	0.3
Stearic acide C18:0	2.7
Oleic acid C18:1	32.3
Linoleic acid C18:2	52.1
Linolenic acidC18:3	0.1
Arachidic acid C20:0	0.2
Gadoleic acid C20:1	0.3
SFA	10.8
UFA	85.1

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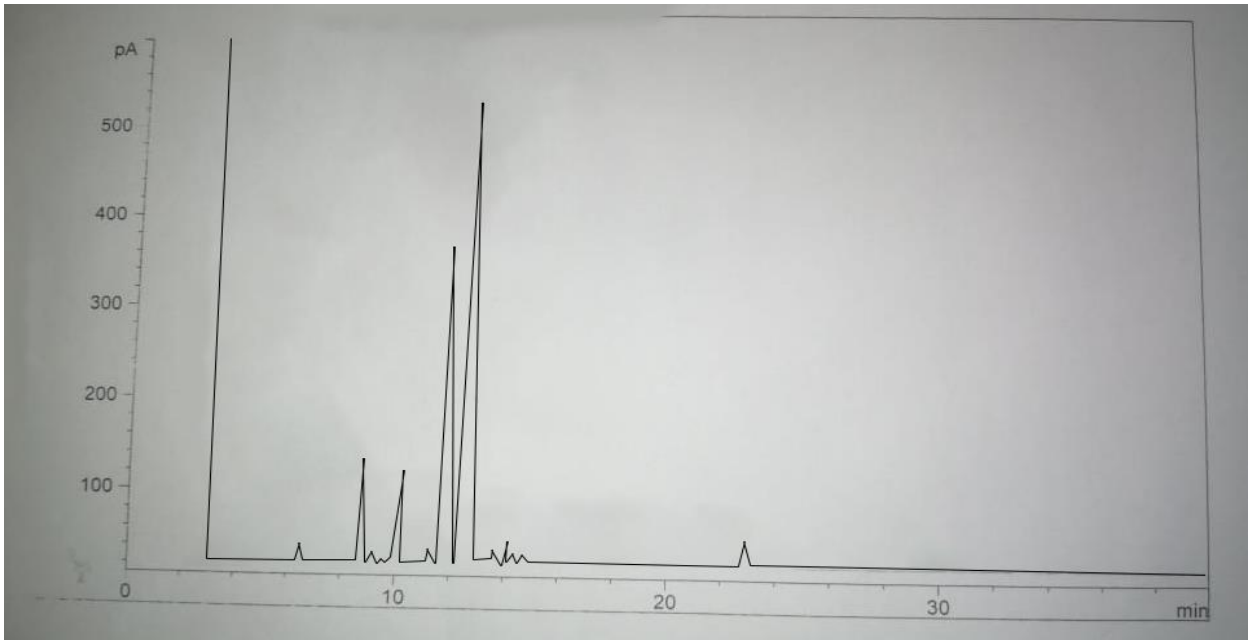

Fig 1 : Chromatogram of the fatty acid composition of the seed yellow onion Amposta oil

The fatty acid composition is another essential aspect of the qualitative evaluation of oils. Indeed, this parameter plays a very important role in the classification of oils and consequently facilitates its marketing. It is also one of the means used to ascertain the adulteration of oil and to detect, if necessary, fraud in the marketed oils **[16]**.

The results obtained for the oil studied show that the major fatty acid is linoleic acid (C18: 2) 52.1% followed by oleic acid (C18: 1) 32.3% then palmitic acid (C16: 0) 7.8% which makes it possible to classify this oil in the linoleic - oleic category. The minor fatty acids, the percentages of which hardly exceed 3 %, are formed by palmitoleic, stearic, linolenic, myristic, arachidic and gadoleic acid.

The ratio of unsaturated fatty acids to saturated fatty acids (SFA / UFA) is higher (7.88) which gives the oil greater stability to auto-oxidation and an important nutritional value **[17, 18]**.

III.3. Total sterols composition of the yellow onion seed Amposta oil

Sterols are an essential component of cell membranes, and we find them in both animals and plants. All sterols have the same nucleus which is the sterolic nucleus, but they differ in their side chain.

Table 2 : Total sterol composition of oil from Amposta yellow onion seeds

Stérols	**Percentage %**
Brassicasterol	0.2
Campesterol	15.9
Stigmasterol	0.3
β-Sitostérol	63.7
Δ-5-Avenasterol	10.6
Δ-7-Stigmasterol	0.6
Δ-7-Avenasterol	0.9
Cholesterol	3.8

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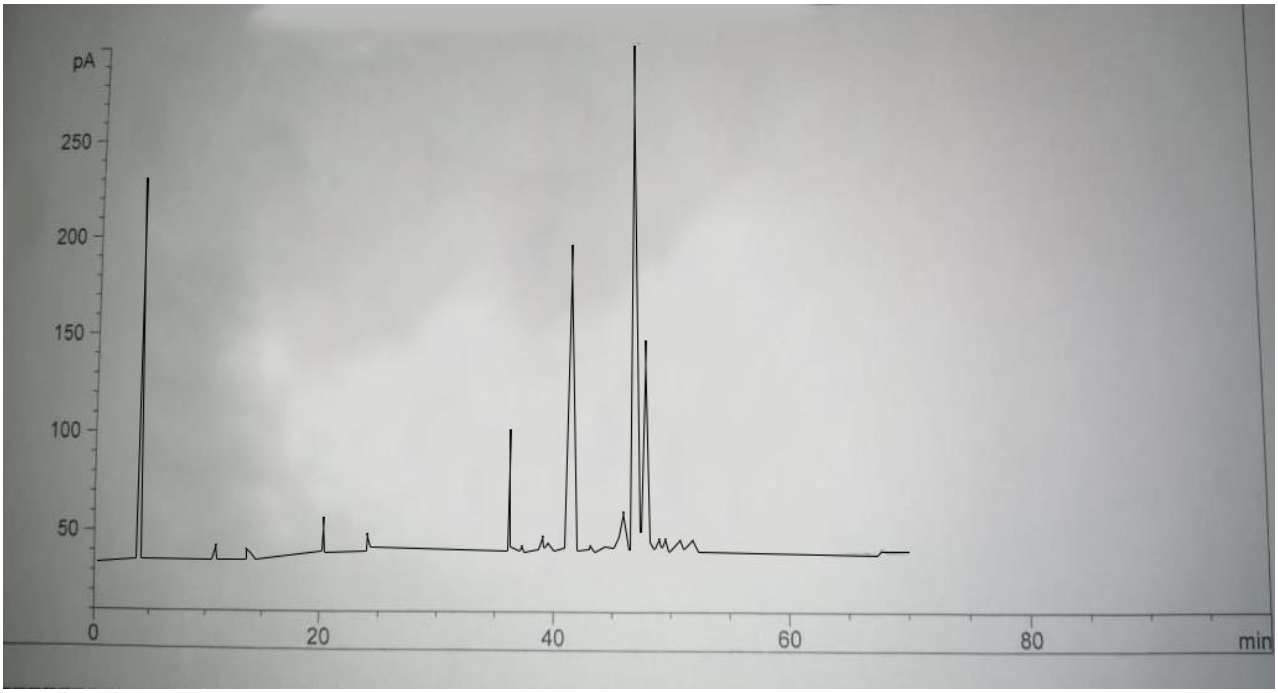

Fig 2 : Chromatogram of the Sterol fraction of the seed yellow onion Amposta oil

From Table 2 and Figure 2, we note the dominance of β-Sitosterol, the content of which is 63.7%. This oil contains a particularly high amount of β-Sitosterol, a substance that opposes the intestinal absorption of cholesterol **[19]**. Campesterol is always higher than Stigmasterol with percentages of 15.9% and 0.3% respectively. The percentage of Δ-5-Avenasterol is around 10.6 % while that of Cholesterol does not reach 4 %.

III.4. Composition of the tocopherols oil from Amposta yellow onion seeds

Tocopherols exert their antioxidant action by inhibiting radical degradation reactions of unsaturated fatty acids **[20, 21]**. Analysis of the chromatogram in figure 3 and table 3, shows the existence of three peaks corresponding to α, β, and γ –tocopherols : α-tocopherols is the majority product of tocopherols with a percentage of 84.9% followed by β -tocopherols 12.8% and finally γ -tocopherols 2.3%. We note the absence of δ -tocopherols. The total tocopherols are of the order of 1511mg / Kg which shows that this oil could exert a significant antioxidant activity.

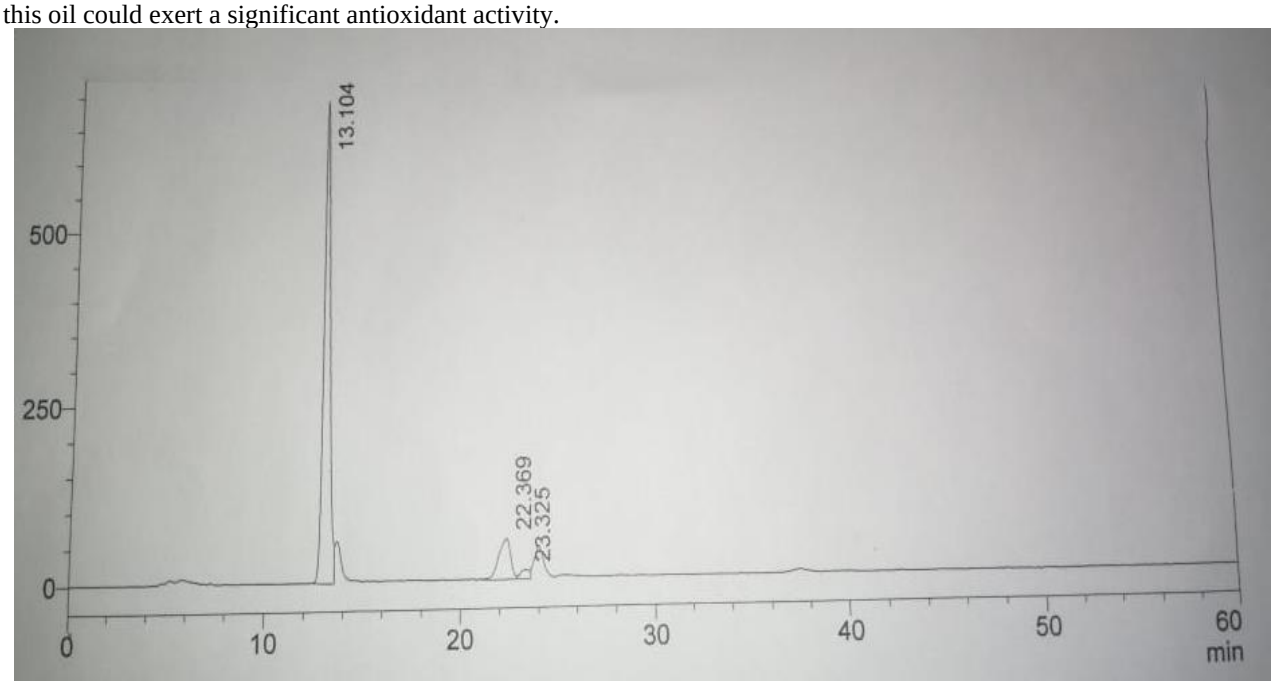

Fig 3 : Chromatogram of the composition of total tocopherols in the Amposta yellow onions seed oil.

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Table 3 : Composition of total tocopherols in the seed oil of yellow onion Amposta

Tocophérols	Percentage (%)
α-tocopherol	84.9
β-tocopherol	12.8
γ-tocopherol	2.3
δ-tocopherol	-
Total tocopherols (mg/kg)	1511

CONCLUSION

The fatty acid composition revealed that the oil studied is very rich in unsaturated fatty acids: C18: 1 and C18: 2. Indeed, the ratio of unsaturated fatty acids to saturated fatty acids is high and equal to 7.88, hence their interest in the preservation of oils and in human nutrition.

Analysis of the sterolic composition of the oil showed that the most important component is β-Sitosterol with a percentage of 63.7 %. It is a sterol characteristic of olive oil. The second most important component is Campesterol. The third component mainly presented is Δ-5-Avenasterol. The fourth component is Cholesterol. The percentages found for the oil studied are in international food standards.

The composition of total tocopherols shows that this oil studied contains an appreciable amount of α, β and γ –tocopherols. These are considered to be natural antioxidants that protect it against oxidation, they give it better stability during storage.

The high content of tocopherol and sitosterol allowed us to conclude that this oil could be used as an additive in food and as an ingredient in cosmetics as a preservative that can increase the shelf life of products.

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