Moroccan Journal of Chemistry
ISSN: 2351-812X
http://revues.imist.ma/?journal=morjchem&page=login
Yatim & al. / Mor. J. Chem. 10 N°4 (2022) 787-799
DOI: https://doi.org/10.48317/IMIST.PRSM/morjchem-v10i4.34416

Morphological and chemical characterization of carob pulps collected from four moroccan regions

M. YATIM[a*], T. EL-ASKRI[a], Y. SEHLI, A. AMECHROUQ[b], A. EL YAACOUBI[c], T. AINANE[c], A. RAHOU[a], M. HAFIDI[a], R. ZOUHAIR[a].

[(a)] Laboratory of Plant Biotechnology and Molecular Biology, Department of Biology, Faculty of Sciences, Moulay Ismail University, B.P. 11201, Zitoune, Meknes, Morocco

[(b)] Laboratory of Molecular Chemistry and Natural Substance, Moulay Ismail University, Faculty of Science, P.B 11201 Zitoune, Meknes, Morocco

[(c)] University of Sultan Moulay Slimane, Higher School of Technology, PB 170, Khenifra, Morocco

* *Corresponding author: m.yatim@edu.umi.ac.ma*

Received 15 April 2022,
Revised 07 Sept 2022,
Accepted 10 sept 2022

Abstract

The present research is included in a strategy of conservation and management of genetic resources for Carob tree (*Ceratonia Siliqua* L.) in Morocco. It has as one of its objectives, the evaluation of the morphological and chemical characteristics of four Moroccan ecotypes of carob pulps. The study related to morphological characterization reveals that there was a significant difference between the four populations from Meknes, Fez, Khemisset and Marrakech collected in 2018 and 2019. The lipid profile of carob pulp extracts analyzed by GC-MS is constituted of fatty acids, hydrocarbons, non-oxygenated compounds, sterols and tocopherols. The concentration of these compounds varies according to the population and year of collection. According to the results, the fatty acid contents are relatively high in 2019 with the dominance of palmitic acid. Additionally, our study approved the dominance of 1-Hexacosene, Tetracosane, Heptadecane. 1-iodo Hexadecane, 1-chloro Octadecane and 1, 54-dibromo Tetrapentacontane. Furthermore, it is noted that Stigmasterol, gamma-Sitosterol, beta-Sitosterol, alpha-Tocospiro A, alpha-Tocospiro B and Vitamin E are present in the samples. Moreover, the hierarchical analysis based on the results of the morphological and chromatographic characterization of the four populations' pulps identify three groups. The first group included populations P3 and P4 respectively from Marrakech and Khemisset. The second group presented by the population P1 from Meknes. The population P2 from Fez constituted the third group.

Keywords: *fatty acid, GC/MS, hydrocarbons, Ceratonia siliqua L pulp, sterols, tocopherols.*

1. Introduction

In order to valorize and conserve plant bio resources, Morocco has adopted programs to promote the agriculture of several species with ecological and socio-economic benefits. The Carob tree (Ceratonia siliqua L.) is a Mediterranean plant [1][2]. It is actually one of the most performing forest, fruit, and forage trees that exist in Morocco [3]. The principal spontaneous populations of carob trees in association with the olive tree, the lentisk or the argan tree are situated in Fez, Marrakech, Agadir, Essaouira, Taza, El Hoceima, Beni Mellal, and Khenifra, [4]. According to the Food and Agriculture Organization (FAO) in October 2021, the carob production in Morocco is estimated at 21501 tons with 10161 ha of area cultivated in 2019 [5]. However, different studies proved that the plant oil contains various bioactive components with biological activities [6] and different therapeutic properties [7]. Previous researches reported the antimicrobial, cytotoxic and pharmaceutical activities of carob [8][9]. However, limited studies were realized on carob oils [10][11]. Therefore, the research aim is to determine the lipid fraction composition by GC-MS of pulp extracts obtained from four different provenances in Morocco.

2. Materials and methods

1.1. ***Plant material***

The quantity of 1500 g of pods from four carob populations were collected in August 2018 and 2019 from different Moroccan regions (Fez, Meknes, Khemisset and Marrakech) (Table 1). Seeds were separated manually and crushed using an electric grinder.

Table 1. Geographic data of Ceratonia siliqua L. fruit collection stations.

Population	Sex	Origin	Latitude N	Longitude W	Altitude (m)	Geographic area
P1	Female	Meknes	33° 53′ 42"	5° 33′ 17″	560	Tray saïs
P2	Female	Fez	34° 03′ 00″	4° 58′ 59″	579	Tray saïs
P3	Female	Marrakech	31° 37′ 48″	8° 00′ 00″	450	Haut Atlas
P4	Female	Khemisset	33° 49′ 0″	6° 4′ 0″	409	Central Board

1.2. ***Morphological characterization of carob fruits***

The morphological characters of the fruits (pods and seeds) were determined on fifteen pods per population of carob (*Ceratonia siliqua* L.) in a randomized manner. Six characters were measured at the pod, namely length, width, thickness, number of seeds, fresh pod weight (g) and yield (seed weight/pod weight *100).

1.3. ***Determination of dry matter***

The dry matter (DM) was determined by drying at 100 °C to 105°C, under atmospheric pressure until a practically constant mass was obtained [12]. The water content (%) of the plant material is given by the following equation:

$$\text{Moisture } (\%) = \frac{P-P1}{M} * 100 \qquad (1)$$

With **P**: mass in g of the test sample before drying, **P1**: mass in g of the test sample after drying and M: mass of the biological material. From the water content, we determine the dry matter content which is given by the following equation:

$$Taux\ de\ matière\ sèche\ (\%) = 100 - \ teneur\ en\ eau\ (\%) \qquad (2)$$

1.4. ***Oil extraction***

The extraction of the fatty content was realized with hexane in a Soxhlet extraction system during three hours on 20g of pulp previously crushed. At the end of the extraction, the solvent was evaporated in a rotating evaporator under vacuum, with moderate temperature (+40°C). The extracts are placed in well-sealed opaque glass bottles and stored (4°C), protected from light and air [13].

1.5. Extraction yield

The yield of oil is the ratio between the weight of oil extracted and the weight of plant material used [14]. The yield (%) is obtained by the following equation:

$$R = \frac{Ph}{Pv} * 100 \qquad (3)$$

R: oil yield in percent, **Ph**: oil weight in grams, **Pv**: weight of plant material in grams.

1.6. Analysis by gas chromatography coupled with mass spectrometry GC-MS

Trans-esterification of the oil was effected. The samples were analyzed by gas chromatography (GC) (Agilent 7890A Series GC) coupled with mass spectrometry (MS) equipped with a multimode injector and a column 123-BD11 (15 m, 0.32 mm, 0.10 µm). Each sample solubilized in chloroform was injected into the column at split mode (1/2 split) using helium as vector gas at 4 mL/min. The temperatures of the ion source and the quadrupole were 230 °C and 150 °C, respectively. The temperature program was started at 40 °C and maintained for 1 min. It increased at 15 °C / min to 160 °C then held 2 min, further raised to 230 °C at 7 °C / min then increased to 360 ° C at 30 °C / min and finally constant for 5 min. The composition was determined from the airs of the peaks. Identification was performed using the 2014 NIST MS library.

1.7. Data Statistical Analysis

Data were analyzed by ANOVA using GraphPad Prism 6 software. All data were reported as mean ± standard deviation (SD). The difference in means was compared by Tukey's test at the $p \leq 0.05$ and measurements were performed in triplicate. Hierarchical clusters were performed using SPSS 26.0 software using the Intergroup Distance Aggregation method, which is based on the proximity distance matrix. The resulting dendrogram was normalized to Pearson correlation. Different letters on the tables indicate different significance at $p \leq 0.05$

3. RESULTS AND DISCUSSIONS

3.1. Morphological characterization of carob pods *Ceratonia siliqua*

The investigation of genetic variability is crucial in plant selection. To select genetic resources, the identification of variability by morphological characterization must be done. Indeed, the results obtained on pods from four populations collected from the two consecutive years 2018 and 2019 (Table 2). They showed that morphological parameters related to the size of carob pods are variable from the population to another. The pod length varies between 9.473±1.055cm and 12.067±1.017cm in 2018 and 9.669±1.022cm and 13.150±1.128cm in 2019, the width of pod varies between 1.432±0.066cm and 1.623±0.126cm in 2018 and 1.544±0.17cm and 1.756±0.211cm in 2019. Similarly, the thickness of pods varies between 0.388±0.053cm and 0.544±0.078cm in 2018 and 0.425±0.066cm and 0.565±0.032cm in 2019, the fresh weight of pod (g) ranges from 3.506±1.107g to 6.784±1.078 in 2018 and 3.772±0.325 to 7.261±0.346g in 2019. The seed yield shows the highest variation, since it varies from 23.161±1.123% to 29.072±1.325% in 2018 and from 25.447±1.234% to 31.359±1.328% and the number of seeds per pod varies from 8.463±0.142 to 11.723±1.115 in 2018 and 10.054±1.103 and 12.755±1.105. This analysis of the agro-morphological

diversity of carob pods (Ceratonia siliqua L.) from the four Moroccan regions showed significant phenotypic variations for mean length, fresh weight, number of seeds/pod and pod yield. In addition, the pod width and thickness shows practically no significant difference between the populations. On the same populations investigated, the relative values for length, width, thickness, number of seeds per pod and fresh pod weight (g) varied from 9, 947±1.047 cm to 21.400±1.160 cm, 1.600±0.130cm to 1.713±0.119 cm, 0.373±0.046 to 0.653±0.062 cm, 7.510±0.892 to 17.133±1.187 and 3.765±0.456g to 14.650±1.137g, respectively [15]. Moreover, the research carried out by El Kahkahi et al, in 2014 [16] on forty-seven populations of carob trees from different sites in Morocco also revealed important variations in the main descriptive morphological characters. On the other hand, in Beni Mellal (Middle Atlas), the population of carob trees is marked by a length between 12 and 15 cm, a width of 1.5 to 2.5 cm, a thickness of pod of 0.4 to 1.37 cm and a number of seeds of 10.67 to 12.5 [17].

Table 2. Morphological characterization of pods from four Moroccan carob populations.

All data are shown as mean ± SD (n = 15).

$^{a-d}$: values with a different subscript in a column are significantly different (P ≤ 0.05).

	Length (cm)		Width (cm)		Thickness (cm)		Fresh pod weight (g)		Number of seeds/pods		Yield (%)	
Population	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**
P1	10,112±0,340^{a}	11,430±0,352^{a}	1,623±0,126^{a}	1,756±0,211^{a}	0,544±0,078^{a}	0,565±0,032^{a}	5.742±1.086^{c}	6.273±0.441^{a}	8.463±0.142^{c}	10.054±1.103^{a}	29,072±1,325^{a}	31,359±1,328^{c}
P2	10,664±1,026^{a}	11,106±0,549^{a}	1,432±0,066^{a}	1,632±0,075^{a}	0,388±0,053^{a}	0,425±0,066^{a}	3.506±1.107^{b}	3.772±0.325^{c}	8.521±1.066^{a}	10.424±1.085^{b}	23,161±1,123^{c}	28,155±1,324^{a}
P3	9,473±1,055^{a}	9,669±1,022^{a}	1,517±0,143^{a}	1,544±0,173^{a}	0,520±0,055^{a}	0,534±0,104^{a}	6.784±1.078^{b}	7.455±0.512^{a}	9.533±1.117^{b}	11.043±1.101^{a}	25,687±1,530^{b}	25,447±1,234^{b}
P4	12,067±1,017^{b}	13,150±1,128^{b}	1,523±0,064^{a}	1,548±0,067^{a}	0,511±0,045^{a}	0,519±0,077^{a}	6,166±0,310^{c}	7.261±0.346^{c}	11,723±1,115^{b}	12.755±1.105^{c}	25,423±1,112^{b}	25,984±1,087^{b}

3.2. Water content and dry matter content (%)

The dry matter content of carob pulps in all four populations was estimated between 94.303±1.028 and 95.103±1.035 in 2018, and 95.258±1.422 to 95.683±1.115 in 2019 (Table 3). It was elevated in the pulps from Meknes (P1) and Fez (P2) compared to those from Khemisset (P4) and Marrakech (P3) without significant difference. As it was concerned the water content of the pulps carob populations chosen, it varies from 4.897±0.175 (P1) to 5.697±0.146% (P4) in 2018 and from 5.132±0.152% (P3) to 5.525±0.164 (P4) in 2019. Our results were more elevated with that obtained by El Kahkahi et al (2015) [18].They revealed that the dry matter content of pulps in Moroccan different populations was estimated between 82.58% and 87.34% with a water content that varies between 12.66% and 17.45%.

Table 3. Water and dry matter content of carob pulp (%)

	$\%H_2O$		%MS	
	2018	**2019**	**2018**	**2019**
P1	4.897±0.175^{a}	4.317±0.157^{a}	95.103±1.035^{a}	95.683±1.115^{a}
P2	5.080±0.164^{a}	4.742±0.178^{b}	94.920±1.172^{a}	95.258±1.422^{a}
P3	5.553±0.150^{b}	5.132±0.152^{c}	94.447±1.062^{a}	94.868±1.152^{a}
P4	5.697±0.146^{b}	5.525±0.164^{a}	94.303±1.028^{a}	94.475±1.328^{a}

$\%H_2O$ water content; %MS dry matter content; Values in the same column with letters are significantly different at the p ≤0.05 (Values represent the mean of three replicates ± Standard Error).

3.3. Oil content of carob pulp

The results reveal the oil content of pulps which varied from 0.102±0.003 (P1) to 0.431±0.010 (P4) in 2018 and from 0.145±0.032% (P1) to 0.456±0.047 (P4) in 2019 (Table 4). The carob pulps contained very little oil content compared to the seeds. It varied according to the population analyzed. These variations in content can be explained by the effect of variety or geographical origin that affects the oil yield [19]. Our results are similar to the data reported in the literature. According to Papaefstathiou et al (2018) [20], the oil yield of the three Cyprus carob pulps was estimated with 0.21±0.02 and 0.23± 0.01% as it is also reported by Oziyci et al in 2014 [21].

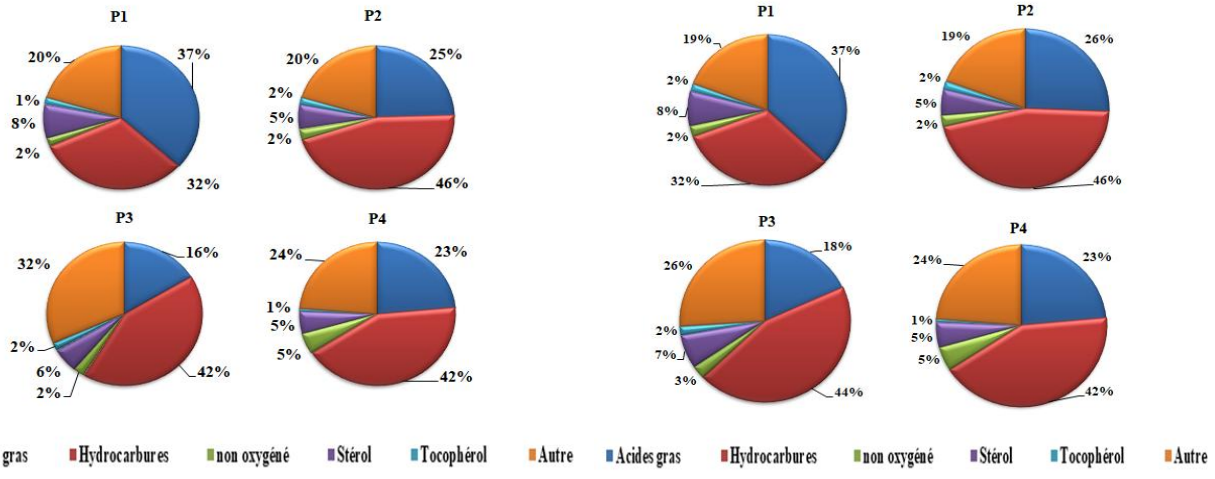
Table 4. Oil content (%)of carob pulp.

	%MG	
	2018	**2019**
P1	0.102±0.003[a]	0.145±0.032[a]
P2	0.346±0.007[b]	0.416±0.054[b]
P3	0.302±0.004[b]	0.344±0.043[b]
P4	0.431±0.010[c]	0.456±0.047[c]

%MG: oil content; Values in the same column with letters are significantly different at the threshold p ≤0.05; n= 3.

3.4. Determination of the pulp extracts composition (%)

The lipid fraction composition by gas chromatography of carob pulps extracts collected in 2018 and 2019 (Figure 1) showed a profile constituted by fatty acid, hydrocarbons, non-oxygenated compounds, sterols and tocopherols. The concentration of these compounds depends on the population analyzed and the collection year. For fatty acids, it was recorded that the values of 16.32 (P3) to 57.12 (P1) in 2018 and 18.21(P3) to 56.72 (P1) in 2019. The Hydrocarbons percentage showed values from 11.71% (P1) to 42.32% (P3) in 2018 and 12.32% to 44.63% in 2019. For non-oxygenated compounds, a percentage of 1.99% (P1) to 4.98% (P4) in 2018 and 2.32% to 3.74% in 2019 was obtained. In addition, the sterols obtained a percentage from 4.98% (P4) to 7.59% (P1) in 2018 and 5.28 (P2) to 7.28% (P1) in 2019. For the tocopherols, the percentage of 0.62% (P4) to 1.74% (P3) in 2018 and 0.71% (P4) to 1.76% (P3) in 2019 was obtained.



(A) **(B)**

Figure 1. Determination of the carob pulp extract composition of the four populations collected in 2018 **(A)** and 2019 **(B)**.

a. Fatty acid composition (%) of carob pulp *Ceratonia siliqua*

Table 5. Fatty acid composition of carob pulp extracts (*Ceratonia siliqua*) (%)

(All data are given as the mean ± SD (n = 3) [a-b]: Values with a different letter in a row differed significantly ($P \leq 0.05$))

		P1		P2		P3		P4	
Fatty acid		**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**
Hexanoïc acid / Caproïc acid	**C_6 : 0**	0.980±0.102[a]	1.118±0.028[a]	0.550±0.043[a]	0.562±0.063[a]	0.370±0.031[a]	0.382±0.043[a]	1.460±0.078[b]	1.440±0.063[b]
Heptanoic acid	**C_7 : 0**	0.023±0.012[a]	0.041±0.011[a]	0.030±0.008[a]	0.047±0.034[a]	0.430±0.075[a]	0.444±0.062[a]	0.060±0.021[a]	0.060±0.022[a]
Octenoic acid	**C_8 : 1**	0.050±0.010[a]	0.122±0.023[a]	0.030±0.010[a]	0.052±0.011[a]	0.102±0.021[a]	0.112±0.022[a]	0.180±0.031[a]	0.180±0.023[a]
Octanoic acid	**C_8 : 0**	0.210±0.050[a]	0.267±0.062[a]	0.290±0.051[a]	0.302±0.071[a]	0.120±0.043[a]	0.125±0.033[a]	0.630±0.043[a]	0.524±0.103[a]
Nonaoic acid	**C_9 : 0**	0.090±0.022[a]	0.135±0.032[a]	0.060±0.011[a]	0.060±0.012[a]	0.122±0.054[a]	0.122±0.032[a]	0.180±0.062[a]	0.165±0.042[a]
Decanoïc acid / Capric acid	**C_{10} : 0**	0.110±0.043[a]	0.121±0.020[a]	0.090±0.031[a]	0.084±0.016[a]	0.124±0.051[a]	0.123±0.022[a]	0.210±0.041[a]	0.211±0.061[a]
lauric acid / Dodecanoïc acid	**C_{12} : 0**	0.230±0.010[a]	0.230±0.041[a]	0.150±0.042[a]	0.143±0.031[a]	0.670±0.071[a]	0.665±0.073[a]	0.300±0.032[a]	0.288±0.034[a]
Tetradecanoic acid / Myristic acid	**C_{14} : 0**	0.540±0.054[a]	0.563±0.052[a]	0.550±0.042[a]	0.542±0.052[a]	0.920±0.072[a]	1.028±0.094[a]	0.600±0.053[a]	0.255±0.053[a]
Palmitoleic acid / Hexadecenoïc acid	**C_{16} : 1**	3.290±0.156[a]	3.321±0.113[a]	1.328±0.065[b]	1.345±0.062[b]	2.720±0.103[a]	2.718±0.104[a]	2.120±0.067[c]	2.132±0.054[c]
Pentadecylic acid / Pentadecanoïc acid	**C_{15} : 0**	1.043±0.044[a]	1.113±0.102[a]	3.548±0.032[b]	3.554±0.073[b]	2.430±0.121[b]	2.455±0.112[b]	1.390±0.082[c]	1.440±0.084[c]
Palmitic acid	C_{16} **: 0**	15.957±0.210[a]	16.057±0.114[a]	15.646±0.126[b]	16.021±0.113[b]	16.360±1.013[b]	16.575±1.013[b]	13.980±1.022[c]	13.965±0.103[c]
Margaric acid ,heptadecanoic acid	**C_{17} : 0**	2.157±0.043[a]	2.167±0.110[a]	1.280±0.053[a]	1.308±0.053[a]	2.761±0.106[a]	2.883±0.113[a]	2.890±0.084[b]	2.897±0.063[b]
Linoleic acid	**C_{18} : 2**	15.410±0.132[a]	15.522±1.107[a]	11.506±0.311[b]	11.545±0.212[b]	10.420±0.104[a]	10.655±1.011[a]	10.640±1.004[c]	10.723±1.023[c]
Oleic acid	**C_{18} : 1**	39.060±1.013[a]	39.145±1.124[a]	36.510±0.321[b]	36.634±1.011[b]	36.810±1.021[c]	36.965±1.052[c]	35.830±1.032[d]	36.065±1.009[d]
Stearic acid / Octadecanoïc acid	**C_{18} : 0**	5.680±0.450[a]	5.832±0.112[a]	9.112±0.074[b]	9.142±0.145[b]	6.070±0.117[b]	6.354±0.108[b]	5.370±0.201[c]	5.450±0.106[c]
Arachidic acid / Eicosanoïc acid	**C_{20} : 0**	2.350±0.113[a]	2.355±0.124[a]	3.970±0.054[b]	3.976±0.107[b]	2.338±0.107[a]	2.341±0.078[a]	3.580±0.107[b]	3.587±0.121[b]
Béhénic acid / Docosanoïc acid	**C_{22} : 0**	1.560±0.102[a]	1.634±0.107[a]	1.820±0.055[b]	1.034±0.057[a]	1.521±0.067[a]	1.252±0.055[a]	1.660±0.081[b]	1.675±0.073[b]
Lignoceric acid / Tetracosanoïc acid	**C_{24}: 0**	3.480±0.056[a]	3.522±0.151[a]	3.530±0.061[b]	3.043±0.011[c]	1.242±0.117[c]	1.264±0.061[c]	5.780±0.112[d]	5.778±0.065[d]
Cerotic Acid / Hexacosanoïc acid	**C_{26} : 0**	5.640±0.064[a]	5.623±0.127[b]	6.850±0.119[b]	7.571±0.105[b]	7.670±1.006[b]	7.812±0.111[b]	6.430±0.121[c]	6.443±0.072[c]
Montanic acid / Octacosanoïc acid	**C_{28} : 0**	2.140±0.101[a]	1.112±0.075[b]	3.150±0.077[b]	3.035±0.125[b]	6.800±0.112[c]	5.725±0.115[c]	6.710±0.075[d]	6.722±0.115[d]

The determination of fatty acid composition in carob pulp extracts collected in 2018 and 2019 by gas chromatography (Table 5) showed that there is no significant difference between the two extracts of the populations examined. The unsaturated fatty acid content was about 51.502% (2018) and 51.809% (2019). However, for the populations studied,

the saturated fatty acid content was about 48.499% (2018) and 48.191% (2019). On the one hand, there are somewhat elevated fatty acid contents in 2019 compared to those obtained in 2018. On the other hand, the dominance of palmitic acid is reported with values of 15.957±0.210, 15.646±0.126, 16.360±1.013, 13.980±1.022 in 2018 and 16.057±0.114, 16.021±0.113, 16.575± 1.013 and 13.965±0.103 in 2019, the linoleic acid with values of 15.410±0.132%, 11.506±0.311, 10.420±0.104 and 10.640±1.004 in 2018 and 15.522±1.107, 11.545±0.212, 10.655±1.011 and 10.723±1.023, the oleic acid with values of 39.060±1.013, 36.510±0.321, 36.810±1.021 and 35.830±1.032% in 2018 and 39.145±1.124, 36.634±1.011, 36.965±1.052 and 36.065±1.009 in 2019, the Stearic acid with values of 5.680±0.450, 9.112±0.074, 6.070±0.117, and 5.370±0.201% in 2018 and 5.832±0.112, 9.142±0.145, 6.354±0.108, and 5.450±0.106 in 2019 and Cerotic Acid with 5. 640±0.064, 6.850±0.119, 7.670±1.006, and 6.430±0.121 in 2018 and 5.623±0.127, 7.571±0.105, 7.812±0.111, and 6.443±0.072 in 2019 for the populations P1, P2, P3, and P4, respectively. An important diversification in fatty acid composition was revealed for carob pulp oil by Youssef et al (2013) [22]. According to these authors, pulp oil is composed mainly of four fatty acids including oleic, linoleic, palmitic and stearic acids with 40.45%, 23.19%, 11.01% and 3.08%. The fatty acid composition of two Greek carob pod varieties at different growth stages includes Palmitic Acid 15.90 - 21.90, Stearic Acid 1.90 - 3.70, Oleic Acid 16.20 - 57.01 and Linoleic Acid 6.00 - 29.10 [23]. According to these authors, the values increased the next year to 17.85±0.49 - 22.05±0.21 for palmitic acid, 2.54±0.08 - 3.61±0.10 for stearic acid, 13.63±0.61 - 38.45±0.40 for oleic acid and 9.42±0.68 - 42.23±0.47 for linoleic acid [24]. The composition of carob leaf extracts collected from 3 different sites revealed 12 fatty acids (C:14 to C: 24) where palmitic, stearic, oleic and linoleic and linolenic acid were dominant respectively with 5.24±0.63 to 13.87±0.21%, 13.10±0.22 to 14.98±0.14%, 3.30±0.11 to 6.56±0.16%, 14.02±0.25 to 15.88±0.14%, and 29.40±0.26 to 35.78±0.75% [19]. In addition, It was founded that long chain fatty acids such as cerotic acid (C26:0) which was detected by [25]. These authors showed that the oil of *Tetracarpidium conophorum* contains cerotic acid with 3.42% and 3.52%. Ayaji et al in 2006 [26] reported the presence of cerotic acid in the seed extracts of *Brachystegia eurycoma* and *Mucuna flagellipes* (Fabaceae) with 4.14% and 2.71% respectively. In the fruits of *Rosa majalis* L. and the leaves of *Vaccinium myrtillus* L. the cerotic acid represents 0.08±0.01 and 0.11±0.01 mg/g respectively [27].

b. The hydrocarbon composition (mg/100g) of the carob pulp extract

The determination of hydrocarbons by gas chromatography was performed (Table 6). A significant difference between the four carob populations was observed between 2018 and 2019. However, we registered the dominance of 1-Hexacosene with values that varied from 21.024±0.15 (P1) to 47.644±0.231 (P3) in 2018 and from 21.133±0.102 (P1) to 48.212±0.173 (P3) in 2019. Therefore, we found the detection of Tetracosan with 8.500±0.035 (P3) to 19.343±0.053 (P4) in 2018 and 8.782±0.022 (P3) to 19.654±0.025 (P4) in 2019. The values were between 5.299±0.429 (P1) and 25.624±0.323 (P2) in 2018 and 6.132±0.214 (P1) and 25.689±0.154 (P2) in 2019 of the Heptadecane. The analysis of the volatile composition of unroasted and roasted Egyptian carob pods showed 1.35 and 0.10% of hydrocarbons with octadecane representing a value of 0.37 ± 0.64 and 0.04 ± 0.04% [28]. El-Rafe et al. in 2020 [29] showed the presence of n-Heptadecane (0.05%), Octadecane (0.07%), 1-Nonadecene (0.07%), 1-Eicosene (0.55%), Eicosane (0.08%) and Heneicosane (0.28%). In addition, the chemical composition of different leaves of *Broussonetia luzonica* (Moraceae), a plant endemic to the Philippines, demonstrated the presence of Heneicosan (3.291%), Tricosan (6.315%), Tetracosan (8.626%) Triacontane (7.341%), Hexacosan (5.534%) and Octacosan (1.66%) [30].

Table 6. Hydrocarbon content (mg/100g MG) of carob pulp extracts (*Ceratonia siliqua*)

(All data are given as the mean ± SD (n = 3) [a-b]: Values with a different letter in a row differed significantly ($P \leq 0.05$))

		P1		P2		P3		P4	
Hydrocarbon		**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**
$C_{13}H_{28}$	Tridecane	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.022±0.006[a]	0.032±0.003[a]	0.603±0.062[b]	0.623±0.012[b]
$C_{12}H_{26}$	Dodecane	0.122±0.012[a]	0.213±0.021[a]	4.046±0.125[b]	4.241±0.102[b]	6.897±0.165[c]	7.116±0.015[c]	0.819±0.023[d]	0.849±0.039[d]
$C_{15}H_{28}$	7-Pentadecyne	0.000±0.000[a]	0.000±0.000[a]	4.841±0.107[b]	4.921±0.122[b]	0.000±0.000[a]	0.000±0.000[a]	2.886±0.024[c]	2.921±0.032[c]
$C_{15}H_{32}$	Pentadecane	0.000±0.000[a]	0.000±0.000[a]	9.095±0.187[b]	9.155±0.045[b]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]
$C_{16}H_{30}$	7-Hexadecyne	0.163±0.013[a]	0.181±0.008[a]	0.000±0.000[b]	0.000±0.000[b]	1.361±0.071[a]	1.422±0.042[a]	0.000±0.000[a]	0.000±0.000[a]
$C_{17}H_{36}$	Heptadecane	5.299±0.429[a]	6.132±0.214[b]	25.624±0.323[b]	25.689±0.154[b]	8.591±0.152[c]	8.765±0.075[c]	14.604±0.043[c]	14.822±0.048[c]
$C_{18}H_{38}$	Octadecane	0.000±0.000[a]	0.000±0.000[a]	6.259±0.120[a]	6.622±0.101[b]	21.205±0.43[d]	22.025±0.081[c]	36.920±0.023[c]	37.432±0.015[c]
$C_{19}H_{38}$	1-Nonadecene	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.258±0.032[a]	0.267±0.026[b]
$C_{20}H_{42}$	Eicosane	0.570±0.067[a]	0.612±0.043[a]	0.000±0.000[b]	0.000±0.000[b]	0.000±0.000[b]	0.000±0.000[b]	0.000±0.000[b]	0.000±0.000[b]
$C_{21}H_{44}$	Heneicosane	1.922±0.141[a]	1.982±0.074[a]	0.000±0.000[b]	0.000±0.000[b]	23.020±0.212[c]	23.322±0.062[d]	7.065±0.045[e]	7.155±0.023[e]
$C_{24}H_{50}$	Tetracosane	8.875±0.054[a]	9.918±0.065[b]	10.996±0.231[b]	11.042±0.401[c]	8.500±0.035[b]	8.782±0.022[a]	19.343±0.053[b]	19.654±0.025[c]
$C_{25}H_{50}$	Z-12-Pentacosene	0.000±0.000[a]	0.000±0.000[a]	0.450±0.031[b]	0.462±0.112[c]	0.000±0.000[a]	0.000±0.000[b]	0.948±0.032[d]	0.978±0.045[d]
$C_{26}H_{52}$	1-Hexacosene	21.024±0.154[a]	21.133±0.102[a]	27.768±0.341[a]	28.675±0.523[b]	47.644±0.231[c]	48.212±0.173[b]	37.006±0.121[d]	37.215±0.053[d]
$C_{28}H_{38}$	Octacosane	0.000±0.000[a]	0.000±0.000[a]	0.415±0.014[b]	0.462±0.032[b]	6.988±0.141[c]	6.991±0.074[c]	2.197±0.032[d]	2.537±0.062[e]
$C_{29}H_{58}$	Z-14-Nonacosane	0.000±0.000[a]	0.000±0.000[a]	4.737±0.212[b]	4.924±0.142[b]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]
$C_{29}H_{52}$	Nonacos-1-ene	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	1.180±0.065[b]	1.207±0.052[b]	1.939±0.032[c]	1.973±0.054[c]
$C_{31}H_{64}$	2-Methyltriacontane	0.763±0.073[a]	0.842±0.082[a]	0.000±0.000[b]	0.000±0.000[b]	0.000±0.000[c]	0.000±0.000[c]	0.000±0.000[d]	0.000±0.000[d]
$C_{35}H_{70}$	17-Pentatriacontene	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	2.632±0.042[b]	2.909±0.051[c]	1.809±0.054[d]	2.129±0.026[e]
$C_{36}H_{74}$	Hexatriacontane	0.173±0.054[a]	0.217±0.038[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]
$C_{44}H_{90}$	Tetratetracontane	0.000±0.000[a]	0.000±0.000[a]	0.726±0.052[b]	0.825±0.032[b]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]	0.000±0.000[a]

c. Composition of un-oxygenated compounds (mg/100g) of the carob tree pulp *Ceratonia siliqua*

For the chemical composition of unoxygenated compounds, the analysis of carob pulp extracts from four Moroccan populations collected during 2018 and 2019 of carob (Table 7) permitted the characterization of 1-iodo Hexadecane, 1- chloro Octadecane and 1,54-dibromo-Tetrapentacontane in all populations during the two collecting years (2018, 2019). The research on *Symplocos crataegoides* stem revealed the availability of 1-iodo Hexadecane (0.57%) and 1-iodo-octadecane (0.46%) [31]. Similarly, Khammas et al in 2020 [32] reported 1-iodo Hexadecane with 8.21% in the hexane extract of *Azadirachta indica*. However, the composition of *Physalis angulate* L. leaves and seeds showed the presence of 1-chloro Octadecane, with values of 2.84 and 1.74% respectively [33]. The majority of these compounds have pharmaceutical and nutritional benefits [33]. The research conducted on those organic compounds has confirmed their activity against various pathogens [34].

d. Sterol composition (mg/100g MG) of carob pulp extract Ceratonia siliqua

The characterization of sterols showed the detection of Stigmasterol, gamma-Sitosterol, and beta-Sitosterol (Table 8) with little variation between 2018 and 2019. The contents of gamma-Sitosterol are relatively important for all the four populations. They vary between 5.899±0.132 to 17.878±0.124 mg/100g MG in 2018 and between 6.853±0.132 to

17.878±0.077 mg/100g MG in 2019. In this context, the research conducted on carob fruits in different Egyptian regions revealed the availability of beta-Sitosterol with 0.02 to 0.07%. The sterols content varies in response to the fruit ripening stage and its roasting [35]. Also, the analysis of the leaves methanolic extract of *Broussonetia luzonica* allowed the detection of the gamma-Sitosterol (14.754%) and the beta-Sitosterol (3.065%) [30].

Table 7. Content of non-oxygenated compounds in carob pulp extracts *Ceratonia siliqua* (mg/100g MG)

(All data are given as the mean ± SD (n = 3) $^{a-b}$: Values with a different letter in a row differed significantly ($P \leq 0.05$))

		P1		P2		P3		P4	
non-oxygenated		**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**
$C_{16}H_{33}I$	Hexadecane, 1-iodo-	0.000±0.000^{a}	0.000±0.000^{a}	20.160±0.130^{b}	20.171±1.160^{b}	6.292±0.122^{c}	6.452±0.075^{c}	10.124±0.054^{a}	10.354±0.054^{a}
$C_{18}H_{37}Cl$	Octadecane, 1-chloro-	0.295±0.035^{a}	0.302±0.032^{a}	8.541±0.412^{b}	8.541±0.321^{b}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}
$C_{18}H_{37}I$	Octadecane, 1-iodo-	0.885±0.075^{a}	0.965±0.045^{a}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}
$C_{22}H_{44}Br_2$	Docosane	0.000±0.000^{a}	0.000±0.000^{a}	0.622±0.033^{b}	0.625±0.102^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{a}	0.000±0.000^{a}
$C_{22}H_{46}S$	1-Docosanethiol	0.264±0.042^{a}	0.321±0.041^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}
$C_{32}H_{64}Br_2$	Dotriacontane, 1,32-dibromo-	0.244±0.034^{a}	0.353±0.023^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000^{a}	0.000±0.000
$C_{54}H_{108}Br_2$	Tetrapentacontane, 1,54-dibromo-	0.336±0.032^{a}	0.423±0.023^{a}	6.881±0.109^{a}	6.881±0.121^{a}	1.119±0.033^{a}	1.119±0.046^{a}	10.468±0.075^{a}	10.475±0.055^{a}

Table 8. Sterol content of carob pulp extracts (in mg/100g MG)

(All data are given as the mean ± SD (n = 3) $^{a-b}$: Values with a different letter in a row differed significantly ($P \leq 0.05$))

		P1		P2		P3		P4	
Sterols		**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**
$C_{29}H_{48}O$	Stigmasterol	0.895±0.125^{a}	0.954±0.023^{a}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}
$C_{29}H_{50}O$	gamma,-Sitosterol	5.899±0.132^{a}	6.853±0.132^{a}	9.302±0.052^{b}	10.042±0.093^{b}	12.554±0.142^{c}	12.723±0.054^{c}	17.878±0.124^{d}	17.878±0.077^{d}
$C_{29}H_{50}O$	beta,-Sitosterol	0.925±0.101^{a}	1.623±0.031^{a}	0.000±0.000^{b}	0.000±0.000^{b}	3.993±0.101^{b}	4.163±0.066^{b}	3.576±0.112^{c}	3.576±0.014^{c}

e. Tocopherol composition (mg/100g) of carob pulp Ceratonia siliqua

The gas chromatography analysis of carob pulp extracts showed the presence of alpha-Tocospiro A in population 1 with 0.447±0.037 and 0.452±0.095 mg/g MG, alpha-Tocospiro B in P2 with 0.267±0.043 and 0.272±0.052 and in P3 with 5.264±0.074 5.532±0.073 mg/g MG in 2018 and 2019 respectively (Table 9). The E vitamin was detected with contents that varied from 0.325±0.053 to 2.671±0.054 mg/g MG in 2018 and from 0.341±0.056 to 2.677±0.036in 2019 (Table 9). In the hexanic extract of *Spondias mombin* leaves, the gamma tocopherol and the E vitamin were identified with contents of 0.89 and 7.82% respectively [36]. Moreover, alpha Tocospiro A and alpha Tocospiro B waer tocopherols that have antioxidant properties according to previous studies [37][38]. In addition, both high temperatures and humidity reduce the tocopherol content [39]. The hierarchical analysis based on the results of morphological and chromatographic characterization of pulps from four Moroccan carob populations collected during 2018 and 2019 identify three groups (Figure 2). The first group includes the P3 and P4 from Marrakech and Khemisset. It is characterized by a content of fatty acids approximately 20.98%, 43.24% of hydrocarbons, 3.41% of non-oxygenated organic compounds, 5.74% of sterol and 0.21% of tocopherol. The second group includes P1 from Meknes. It is characterized by an estimated average fatty acid content of 36.94%, 7.54% of sterol and 31.92 of hydrocarbons. The population 2 from Fez constitutes the third group which is marked by a fatty acid content of 25.08%, a high content of hydrocarbons 45.60%, 5.36% of sterol and 1.66% of tocopherol.

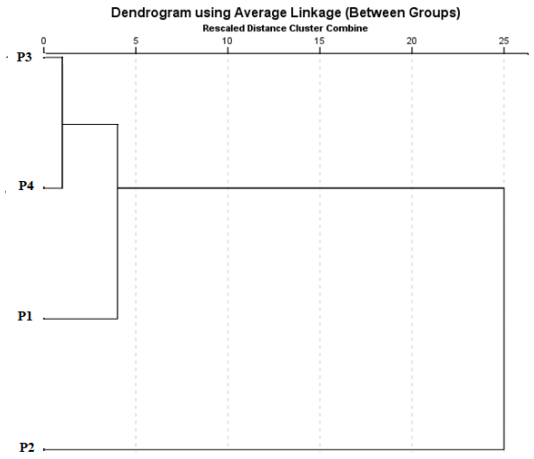


Figure 2. Hierarchical classification of four populations of Moroccan carob pulps.

Table 9. Tocopherol content of carob pulp extracts (mg/100g MG)

(All data are given as the mean ± SD (n = 3) $^{a-b}$: Values with a different letter in a row differed significantly ($P \leq 0.05$))

		P1		P2		P3		P4	
Tocopherol		**2018**	**2019**	**2018**	**2019**	**2018**	**2019**	**2018**	**2019**
$C_{29}H_{50}O_4$	alpha,-TocospiroA	0.447±0.037^{a}	0.452±0.095^{a}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}
$C_{29}H_{50}O_4$	alpha,-TocospiroB	0.803±0.102^{a}	0.845±0.081^{a}	0.267±0.043^{b}	0.272±0.052^{b}	5.264±0.074^{c}	5.532±0.073^{c}	0.000±0.000^{d}	0.000±0.000^{d}
$C_{29}H_{50}O_2$	Vitamin E	0.325±0.053^{a}	0.341±0.056^{a}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	0.000±0.000^{b}	2.671±0.054^{c}	2.677±0.036^{c}

Conclusion

The analysis of the agro-morphological diversity of carob pods (Ceratonia siliqua L.) from four Moroccan regions showed significant phenotypic variations. The gas chromatographic analysis of the pulp extracts made was done to determine a profile constituted by fatty acids, hydrocarbons, non-oxygenated compounds, sterols and tocopherols. The concentration of these compounds varies according to the population analyzed and the year of their collection. Moreover, the statistical analysis showed the presence of a significant difference between the extracts in 2018 and 2019. The results obtained revealed the dominance of Palmitic, Linoleic, Oleic, Stearic and Cerotic acid for fatty acids, 1-Hexacosene, Tetracosane, heptadecane for hydrocarbons. Concerning the non-oxygenated compounds, it was reported that the dominance of 1-iodo Hexadecane, 1- chloro Octadecane and 1,54-dibromo-Tetrapentacontane with the presence of Stigmasterol, gamma-Sitosterol and beta-Sitosterol with the dominance of gamma-Sitosterol for the sterols Based on the results of morphological and chromatographic characterization of pulps from four Moroccan carob tree populations collected in 2018 and 2019, the hierarchical analysis to identify three groups. The first group includes the P3 and P4 from Marrakech and Khemisset. The second group consists of the P1 from Meknes. Population 2 from Fez constitutes the third group.

References

[1] V. Cavallaro, C. Maucieri, C. Patanè, G. Fascella, and A. Pellegrino, "Polyphenols leaching and seed dormancy in carob (Ceratonia siliqua L .) in relation to hot water treatment," *Acta Physiol. Plant.*, vol. 43, no. 141, pp. 1–10, 2021, doi: 10.1007/s11738-021-03308-z.

[2] K. El Oumlouki, G. Salih, A. Jilal, H. Dakak, M. El Amrani, and A. Zouahrt, "Comparative study of the mineral

composition of carob pulp (Ceratonia siliqua L.) from various regions in Morocco," *Moroccan J. Chem.*, vol. 9, no. 4, pp. 741–753, 2021, doi: 10.48317/IMIST.PRSM/morjchem-v9i3.21872.

[3] H. Sbay, "Le caroubier au Maroc Un arbre d'avenir," *le Cent. Rech. For. Rabat*, no. May 2008, 2008.

[4] Ait Chitt M., Belmir M., and Lazrak A., "Production de plants sélectionnés et greffés de caroubier," *Transf. Technol. en Agric.*, vol. 153, no. 037, pp. 77–80, 2007.

[5] FAO, "(2021, October 14). The Food and Agriculture Organization of the United Nations https://www.fao.org/faostat/fr/#data/QCL," 2021.

[6] I. Maliki, A. El Moussaoui, H. Elmsellem, M. Ramdani, and K. Elbadaoui, "Phytochemical screening and the antioxidant, antibacterial and antifungal activities of aqueous extracts from the leaves of Salvia officinalis planted in Morocco," *Moroccan J. Chem.*, vol. 9, no. 2, pp. 354–368, 2021, doi: 10.48317/IMIST.PRSM/morjchem-v9i2.24840.

[7] S. A. Ismaili, I. Marmouzi, K. Sayah, H. Harhar, E. A. Faouzi, S. Gharby, B. Himmi, S. Kitane, M. A. El Belghiti "Chemical analysis and anti-oxidation activities of the Moroccan Milk Thistle," *Moroccan J. Chem.*, vol. 4, no. 3, pp. 4-3 (2016) 695-702, 2016, [Online]. Available: https://revues.imist.ma/index.php/morjchem/article/view/4845.

[8] A. Ben Hsouna, M. Trigui, R. Ben Mansour, R. M. Jarraya, M. Damak, and S. Jaoua, "Chemical composition, cytotoxicity effect and antimicrobial activity of Ceratonia siliqua essential oil with preservative effects against Listeria inoculated in minced beef meat," *Int. J. Food Microbiol.*, vol. 148, no. 1, pp. 66–72, 2011, doi: 10.1016/j.ijfoodmicro.2011.04.028.

[9] I. Fadhil Abdul-Husin, "Study of the Effect of Carob (Ceratoniasiliqua L.) Extract Activity as Antibiotic from UTI," *Al-Qadisiyah J. Agric. Sci. (P-ISSN 2077-5822 , E-ISSN 2617-1479)*, vol. 8, no. 1, pp. 17–25, 2018, doi: 10.33794/qjas.vol8.iss1.48.

[10] N. Tlili, F. Sakouhi, W. Elfalleh, S. Triki, and a. Khaldi, "Fatty acids , total phenols and proteins of Caper, Acacia, Cactus and Carob seeds," *Asiat. J. Biotechnol. Resour.*, vol. 2, no. 04, pp. 384–390, 2011, doi: 03.2011/AJOBR-2011/02(04)/384-390.

[11] B. Matthaus and M. M. Özcan, "Lipid evaluation of cultivated and wild carob (Ceratonia siliqua L .) seed oil growing in Turkey," *Sci. Hortic. (Amsterdam).*, vol. 130, no. 1, pp. 181–184, 2011, doi: 10.1016/j.scienta.2011.06.034.

[12] S.Nielsen, "Food analysis, food science texts series.," *Springer Berlin.*, 2010, [Online]. Available: https://doi.org/10.1007/978-1-4419-1478-16.

[13] M. S. A. Kechebar, S. Karoune, K. Laroussi, and A. Djellouli, "Phenolic Composition and Antioxidant Activities of Opuntia ficus Indica L . Cladodes Related to Extraction Method," *Int. J. Sci. Res.*, vol. 9, no. 6, pp. 744–749, 2017.

[14] A. Akrout, R. Chemli, I. Chref, and M. Hammami, "Analysis of the essential oil of Artemisia campestris L.," *Flavour Fragr. J.*, vol. 16, no. 5, pp. 337–339, 2001, doi: 10.1002/ffj.1006.

[15] M. Yatim, R. El Kahkahi, I. Es-sbata, T. El-askri, S. El oirdi, T. Lakhlifi, A. Belhaj, M. Hafidi, & R. Zouhair "Effects of Pre-sowing Treatments and Abiotic Stress on the Germination of Ceratonia siliqua Seeds of Four Moroccan Biomes," *Annu. Res. Rev. Biol.*, vol. 35, no. 12, pp. 11–31, 2020, doi: 10.9734/ARRB/2020/v35i1230307.

[16] R. El Kahkahi, R. Zouhair, M. Ait Chitt, and R. Errakhi, "Morocco carob (Ceratonia siliqua L .) populations : Morphological variability of Pods and Kernel," *Int. J. pure Appl. Biosci.*, vol. 2, no. 4, pp. 38–47, 2014.

[17] K. Elfazazi, M. Jbilou, A. Assaidi, M. Benbati, and H. Harrak, "Morphological and Biochemical Variability of Moroccan Carob (Ceratonia siliqua L .) Produced in Beni Mellal Region," vol. 5, no. 4, pp. 14–21, 2017.

[18] R. El Kahkahi, R. Zouhair, M. Diouri, M. Ait Chitt, and R. Errakhi, "Morphological and biochemical characterization of Morocco carob tree (Ceratonia siliqua L .)," *Int. J. Biol. Med. Res.*, vol. 6, no. 2, pp. 4946–4952, 2015.

[19] S. Dallali, F. Aloui, H. Selmi, and H. Sebei, “Comparison of the chemical composition and the antioxidant activity of the leaves of Carob tree (Ceratonia siliqua L .) collected in three sites of Djebel Zaghouan (Tunisia) Comparaison de la composition chimique et de l ’ Activité antioxydante des feu,” *J. new Sci.*, vol. CI, no. 21, pp. 3429–3438, 2018, doi: E-ISSN 2286-5314.

[20] E. Papaefstathiou, A. Agapiou, S. Giannopoulos, and R. Kokkinofta, “Nutritional characterization of carobs and traditional carob products,” *Food Sci. Nutr.*, vol. 6, no. 8, pp. 2151–2161, 2018, doi: 10.1002/fsn3.776.

[21] H. R. Oziyci *et al.*, “Mineral composition of pods and seeds of wild and grafted carob (Ceratonia siliqua L.) fruits,” *Sci. Hortic. (Amsterdam).*, vol. 167, pp. 149–152, 2014, doi: 10.1016/j.scienta.2014.01.005.

[22] M. K. E. Youssef, M. M. El-manfaloty, and H. M. Ali, “Assessment of Proximate Chemical Composition , Nutritional Status , Fatty Acid Composition and Phenolic Compounds of Carob (Ceratonia Siliqua L .),” vol. 3, no. 6, pp. 304–308, 2013, doi: 10.5923/j.fph.20130306.06.

[23] S. A. Vekiari, G. Ouzounidou, M. Ozturk, and G. Görk, “Variation of quality characteristics in Greek and Turkish carob pods during fruit development,” *Procedia - Soc. Behav. Sci.*, vol. 19, pp. 750–755, 2011, doi: 10.1016/j.sbspro.2011.05.194.

[24] A. S. Vekiari, G. Ouzounidou, G. Gork, M. Ozturk, and M. Asfi, “Compositional changes of major chemical compounds in greek carob pods during development,” *Bull. Chem. Soc. Ethiop*, vol. 26, no. 3, pp. 343–351, 2012.

[25] W. A. Adebayo, K. A. Taiwo, and O. J. Olubiyo, “Effects of Different Solvent Polarity on the Quality Characteristics of Conophor Nut Oil,” *Int. J. Food Sci. Agric.*, vol. 5, no. 2, pp. 339–345, 2021, doi: 10.26855/ijfsa.2021.06.020.

[26] I. A. Ajayi, R. A. Oderinde, D. O. Kajogbola, and J. I. Uponi, “Oil content and fatty acid composition of some underutilized legumes from Nigeria,” *Food Chem.*, vol. 99, no. 1, pp. 115–120, 2006, doi: 10.1016/j.foodchem.2005.06.045.

[27] A. Savych, R. Basaraba, N. Muzyka, and P. Ilashchuk, “Analysis of fatty acid composition content in the plant components of antidiabetic herbal mixture by GC-MS,” *Pharmacia*, vol. 68, no. 2, pp. 433–439, 2021, doi: 10.3897/PHARMACIA.68.E66693.

[28] M. A. Farag and D. M. El-kersh, “Volatiles profiling in Ceratonia siliqua (Carob bean) from Egypt and in response to roasting as analyzed via solid-phase microextraction coupled to chemometrics,” *J. Adv. Res.*, vol. 8, no. 4, pp. 379–385, 2017, doi: 10.1016/j.jare.2017.05.002.

[29] H. M. El-Rafe, R. S. Mohammed, A. H. Abou Zeid, and A. A. Sleem, “Bioactivities and phytochemical studies of acrocarpus fraxinifolius bark wight arn,” *Egypt. J. Chem.*, vol. 63, no. 1, pp. 209–214, 2020, doi: 10.21608/EJCHEM.2019.15114.1917.

[30] F. P. Casuga, A. L. Castillo, and M. J. A. T. Corpuz, “GC–MS analysis of bioactive compounds present in different extracts of an endemic plant Broussonetia luzonica (Blanco) (Moraceae) leaves,” *Asian Pac. J. Trop. Biomed.*, vol. 6, no. 11, pp. 957–961, 2016, doi: 10.1016/j.apjtb.2016.08.015.

[31] N. Govindarajan, U. M. R. Cheekala, S. Arcot, S. Sundaramoorthy, R. Duraisamy, and I. Raju, “GC-MS analysis of n-hexane extract of stem bark of symplocos crataegoides buch.-ham. Ex d. Don,” *Pharmacogn. J.*, vol. 8, no. 6, pp. 520–524, 2016, doi: 10.5530/pj.2016.6.2.

[32] A. D. Khammas, N. A. Ali, M. Khalaf, and M. Amin, “Diagnosis of active compounds of ethanol and hexane extract of azadirachta indica by gas chromatograph mass spectrometer technique (gc-ms) and evaluation of its antagonism efficiency on the fusarium oxysporum f . Sp . Lycopersici growth in vitro,” *Plant Arch.*, vol. 20, no. 2, pp. 6741–6750, 2020.

[33] S. A. Salim, K. H. Abood, and M. A. Razzooqee, “Comparative study of antioxidant activity and secondary

metabolites between in vitro and ex vitro cultures of physalis angulate L.: An edible and medicinal plant," *Indian J. Ecol.*, vol. 47, pp. 145–152, 2020.
[34] M. Al Bratty *et al.*, "Phytochemical, Cytotoxic, and Antimicrobial Evaluation of the Fruits of Miswak Plant, Salvadora persica L.," *J. Chem.*, vol. 2020, no. Mic, p. 11, 2020, doi: 10.1155/2020/4521951.
[35] M. A. Farag *et al.*, "Variation in Ceratonia siliqua pod metabolome in context of its different geographical origin, ripening stage and roasting process," *Food Chem.*, vol. 283, no. September 2018, pp. 675–687, 2019, doi: 10.1016/j.foodchem.2018.12.118.
[36] O. C. Akanji, "Determination of bioactive constituents of Spondias mombin leaves by GC-MS analysis," *World J. Adv. Res. Rev.*, vol. 06, no. 03, pp. 149–165, 2020, doi: 10.30574/wjarr.
[37] S. Munné-Bosch and L. Alegre, "The Function of Tocopherols and Tocotrienols in Plants," *CRC. Crit. Rev. Plant Sci.*, vol. 21, no. 1, pp. 31–57, 2002, doi: 10.1080/0735-260291044179.
[38] I. Bano and G. S. Deora, "Preliminary phytochemical screening and GC-MS analysis of methanolic leaf extract of Abutilon pannosum (Forst. F.) Schlect. from Indian Thar desert," *J. Pharmacogn. Phytochem.*, vol. 8, no. 1, pp. 894–899, 2019.
[39] R. Rodríguez-Solana, A. Romano, and J. M. Moreno-Rojas, "Carob Pulp: A Nutritional and Functional By-Product Worldwide Spread in the Formulation of Different Food Products and Beverages. A Review," *Processes*, vol. 9, no. 1146, pp. 1–45, 2021, doi: https://doi.org/10.3390/ pr9071146.

(2022) ; https://revues.imist.ma/index.php/morjchem