Pol. J. Environ. Stud. Vol. 33, No. 4 (2024), 3733-3745
DOI: *10.15244/pjoes/177756* ONLINE PUBLICATION DATE: 2024-04-09

Original Research

Study of the Physiological Behavior of Some Plants in Response to Climate Change Conditions

Abdelouahid Laftouhi[1]*, Mary Anne W. Cordero[3], Mohamed Adil Mahraz[1], Hannou Zerkani[4], Anouar Hmamou[1], Amine Mounadi Idrissi[1], Tagnaout Imane[4], Noureddine Eloutassi[1], Zakia Rais[1], Abdslam Taleb[2], Mustapha Taleb[1]

[1]Laboratory of Electrochemistry, Modeling and Environment Engineering (LIEME), Sidi Mohamed Ben Abdellah University, Faculty of Sciences Fes, Morocco
[2]Environmental Process Engineering Laboratory- Faculty of Science and Technology Mohammedia, - Hassan II University of Casablanca, Morocco
[3]Basic Science Department, College of Medicine Princess Nourah bint Abdulrahman University Riyadh Kingdom of Saudi Arabia
[4]Chemistry of bioactive molecules and the environment, Faculty of Sciences, Moulay Ismail University, B.P. 11201, Zitoune Meknes, Morocco

Received: 18 October 2023
Accepted: 28 December 2023

Abstract

In the context of addressing climate change, it becomes essential to anticipate how it will affect plant biodiversity and the way plants adapt physiologically and morphologically to challenging environmental circumstances. To gain a comprehensive understanding of how plants adapt to adverse climatic conditions, we conducted a year-long study with three distinct water stress levels: 25% (sample 1), 50% (sample 2), and 75% (sample 3). The findings revealed a general decrease in primary metabolites (including proteins, carbohydrates, dietary fiber, lipids, and essential minerals like Mg, Fe, K, and Mn) as the water stress level increased. In contrast, secondary metabolites (such as alkaloids, flavonoids, saponins, tannins, and coumarins) exhibited an increase with rising water stress, although a decline became evident as conditions worsened. The same trend was observed in essential oil yield. Furthermore, gas chromatography analysis of essential oils from the plants indicated significant alterations in their chemical composition due to the influence of stressful environmental conditions.

Keywords: climate change, primary metabolites, secondary metabolites, essential oil yield

*e-mail: Laftouhiabdelouahid1993@gmail.com

Introduction

Since ancient times, herbal medicine has been in great demand and keeps increasing day by day today, especially in industrialized countries [1]. Noting that the World Health Organization (WHO) has stated that about 65% to 80% of the world's humanity in evolving countries use aromatic and medicinal plants for their primary health care [2]. On the other hand, developing countries [3], which view other difficult economic situations [4], do not have the means to use modern medicine [5]. Therefore, they have access to traditional medicine to heal themselves [6]. Currently, plant-based products offer a compelling reservoir of contemporary active ingredients for combating diverse diseases [7]. Aromatic and medicinal plants hold a crucial role in human existence [8], brimming with virtues that can be harnessed for a multitude of purposes [9]. Aromatic and medicinal plants represent an extraordinary source of antidotes and constitute a reservoir of plant-based medicine [10]. Man retains ancestral relationships with aromatic and medicinal plants for his needs [11].

Indeed, climate change has significant implications for plant life cycle events, ecosystem services, and cultural practices [12]. Climate change can alter the timing of key life cycle events in plants, including flowering, fruiting, and leaf senescence [13]. Warmer temperatures can cause these events to occur earlier than usual, disrupting the natural synchrony between plants and their pollinators, or seed dispersers [14]. Impacts on plant reproduction and ecosystem dynamics: The disruption of plant-pollinator or plant-seed disperser interactions can have cascading effects on plant reproduction and ecosystem dynamics. Reduced pollination or seed dispersal can lead to decreased plant fitness, altered plant community compositions, and changes in ecosystem processes such as nutrient cycling and trophic interactions [15]. Overall, climate change can have far-reaching consequences for plant-related ecological processes, ecosystem services, and cultural practices [16]. Understanding and addressing these impacts are crucial for effective conservation efforts, sustainable resource management, and the preservation of traditional knowledge and practices associated with plants [17].

However, climate change is currently modifying the morphological and physiological processes of aromatic and medicinal plants, which can negatively affect the chemical composition of secondary metabolites and essential oils.

This study aims to evaluate the influence of stressful conditions on the physiological behavior of some plants in response to climate change.

Materials and Methods

Study Area

Situated in the northern part of the Kingdom of Morocco, the province of Taounate covers an expansive area of 5616 km^2. As of the 2014 census, its population stood at 662,246 inhabitants. Notably, approximately 87% of this population calls the rural areas home, emphasizing the predominantly agrarian nature of the province. Taounate, with its abundant natural resources, strong rural identity, human capital, and rich cultural heritage, holds substantial promise for sustainable development. Harnessing these valuable assets can play

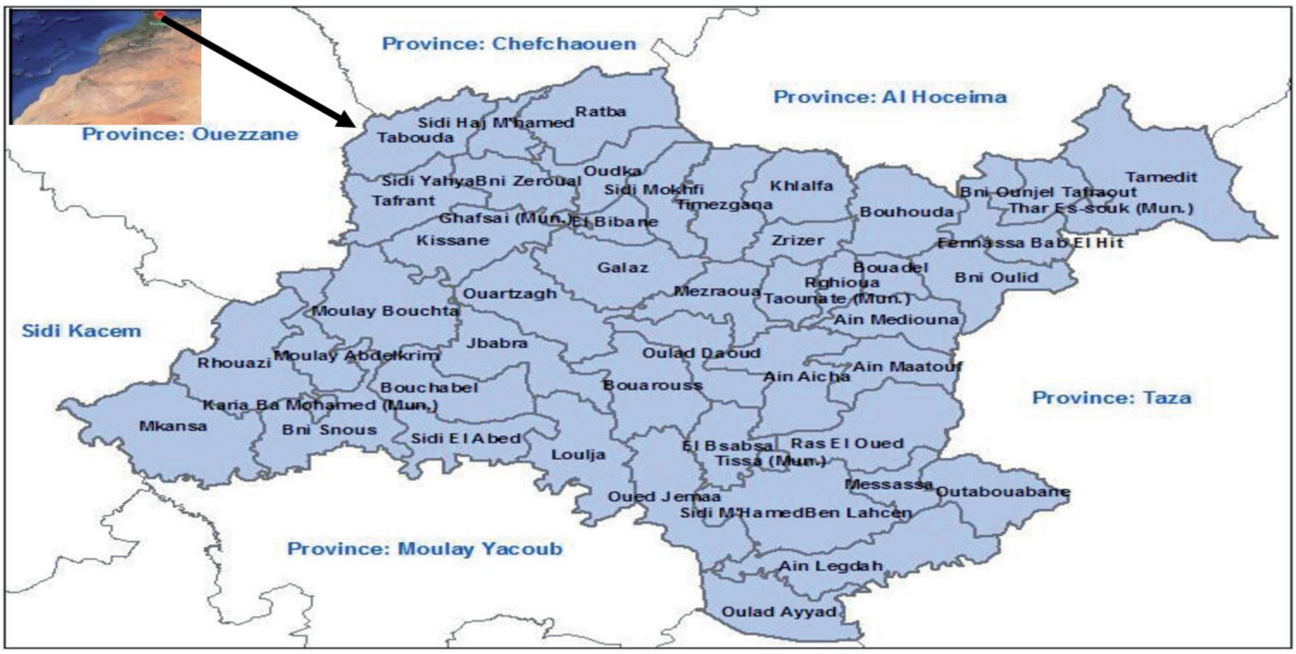

Fig. 1. Map of the study area (Taounate, North Morocco).

a pivotal role in fostering prosperity and enhancing the well-being of both the province and its residents.

Subculture of the Sample

Before transplanting, significant time and effort are dedicated to meticulously preparing the soil in our two fields. Several weeks in advance, we engage in thorough soil cultivation to thwart the proliferation of weeds, ensure a well-balanced blend of various soil factors, and mitigate the risk of root asphyxia when watering. This precise groundwork is geared towards creating optimal conditions for plant growth.

Furthermore, post-transplantation, our commitment extends to the regular removal of weeds on a weekly basis. This ongoing practice is crucial in preventing unwanted competition with our desired plants for vital nutrients. By eradicating these weeds, we guarantee that the intended plants have exclusive access to the resources they require for their optimal development.

By employing these diverse strategies, we ensure the cultivation of a thriving and robust crop in our fields.

The planting of the three sample sets was executed under varying levels of water stress for a year: 25% water stress for sample 1, 50% for sample 2, and 75% for sample 3. The study area encompasses 90 square meters, divided equally into three sections, each spanning 30 square meters [18].

Phytochemical Screening

The qualitative analysis process is geared towards establishing the presence or absence of specific compounds or groups of compounds in the samples. It encompasses a range of tests and techniques, including color reactions, precipitation reactions, various chromatographic methods (such as thin-layer chromatography and column chromatography), and spectroscopic approaches. Through the application of these methods, researchers can discern the categories of metabolites present in the plant samples, which may encompass alkaloids, flavonoids, tannins, terpenoids, saponins, phenolic compounds, and other chemical constituents.

Following the identification of the major families of primary and secondary metabolites via qualitative analysis, the next step involves quantitative assays to determine the concentrations or levels of these metabolites. The quantification of secondary metabolites was carried out according to the protocols of Gössling et al., Wang et al., and Sautner et al. [12-14]. These methodologies are designed to provide a reliable framework for accurately measuring and determining the levels or concentrations of the identified metabolites in plant samples.

Quantitative analysis employs a range of techniques, including spectrophotometry, high-performance liquid chromatography (HPLC), gas chromatography (GC), mass spectrometry (MS), and various other analytical methods. These techniques offer a more precise determination of the quantity or concentration of specific metabolites present in the samples. The quantitative data acquired through these analyses plays a critical role in evaluating the potential therapeutic or bioactive properties of the plants under study.

The insights obtained through phytochemical screening, encompassing both qualitative and quantitative dimensions, bear substantial significance for advancing research and development in domains like natural product chemistry, pharmacology, and plant-derived therapeutics. It offers valuable insights into the chemical composition of plant samples, aids in the identification of potential bioactive compounds, and enhances our understanding of their therapeutic properties and potential applications [15-17].

Essential Oils

In this study, the plant material utilized consisted of dried leaves, which were carefully preserved in the shade to maintain their chemical composition. To obtain the essential oils from these dried leaves, a Clevenger-type apparatus was employed. This type of apparatus is widely recognized for its application in the hydro-distillation method of essential oil extraction.

Approximately 100 grams of the dried leaves were loaded into the apparatus, and the extraction procedure was conducted using hydro-distillation. Hydro-distillation is a method that employs water as a solvent to extract the volatile aromatic compounds, including essential oils, from the plant material. The design of the apparatus facilitates the separation of essential oils from the water and the subsequent collection of these valuable oils [18].

Gas Chromatography (GC)

In the gas phase chromatography analysis, the extracted essential oils underwent further examination using a flame ionization detector (FID) in conjunction with two capillary columns of differing polarities. Specifically, the capillary columns employed were OV. 101 (25 meters long, with a diameter of 0.22 mm and a film thickness of 0.25 µm) and Carbowax 20 M (also 25 meters in length, with a diameter of 0.22 mm and a film thickness of 0.25 µm).

The analysis was conducted with a programmed oven temperature, where the temperature ranged from 50 to 200ºC. This temperature gradient increased at a rate of 5ºC per minute, facilitating a gradual temperature rise during the analysis. This programmed temperature sequence was designed to effectively separate and detect various volatile compounds within the essential oils based on their boiling points and volatility.

In this chromatographic system, helium gas was utilized as the carrier gas, flowing at a rate of 0.8 ml per minute. The carrier gas serves as the mobile phase,

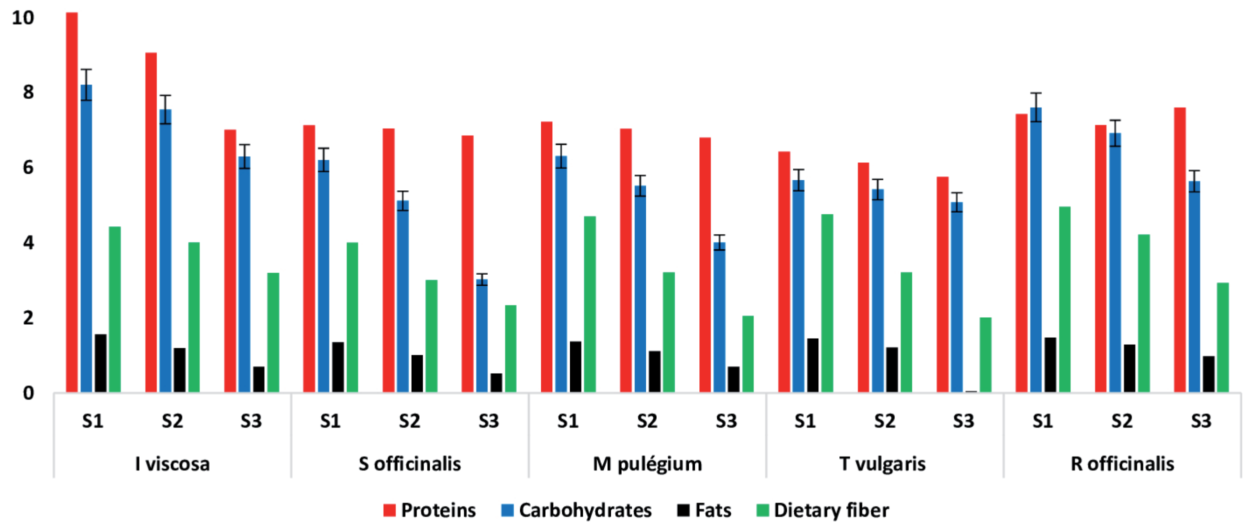

Fig. 2. Percentage of nutritional values (S1: Sample 1; S2: Sample 2; S3: Sample 3).

transporting the vaporized essential oil compounds through the capillary columns and towards the detector.

The flame ionization detector (FID) is a commonly employed detector in gas chromatography. It functions by measuring the ionization current generated through the combustion of organic compounds in a hydrogen-air flame. The FID furnishes a quantitative measurement of the separated compounds, as the ionization current is directly proportional to their concentration.

By implementing gas phase chromatography with the FID detector, combined with the use of various capillary columns, this analysis offers detailed insights into the individual chemical constituents found within the essential oils. This technique facilitates the identification and quantification of specific compounds, thereby contributing to a deeper understanding of the chemical composition and potential therapeutic properties of the essential oils [22, 25].

Results and Discussion

Quantitative Analyses

Primary Metabolites

The data in Fig. 2 reveals substantial variations in the composition of simple food components among the five plant species under study. Proteins consistently represent the highest percentage, with values ranging from 5.76% to 10.13%. Carbohydrates follow, with percentages spanning from 3.02% to 8.21%, while dietary fibers range from 2.01% to 4.7%, and lipids vary from 0.05% to 1.56%.

Furthermore, it's evident that the composition of these simple food components responds to changes in water stress levels. Generally, these percentages decrease as water stress severity intensifies. This phenomenon is observed across all plant species studied.

Examining specific plants:

Inula viscosa: Proteins range from 7.01% to 10.13%, carbohydrates from 6.3% to 8.21%, lipids from 0.7% to 1.56%, and dietary fiber from 3.2% to 4.43%.

Salvia officinalis: Proteins range from 6.86% to 7.13%, carbohydrates from 3.02% to 6.21%, lipids from 0.52% to 1.35%, and dietary fiber from 2.34% to 4%.

Mentha pulegium: Proteins range from 6.8% to 7.23%, carbohydrates from 4.01% to 6.31%, lipids from 0.7% to 1.37%, and dietary fiber from 2.01% to 4.7%.

Thymus vulgaris: Proteins range from 5.76% to 6.43%, carbohydrates from 5.08% to 5.76%, lipids from 0.05% to 1.45%, and dietary fiber from 2.01% to 4.76%.

Rosmarinus officinalis: Proteins range from 7.14% to 7.61%, carbohydrates from 5.64% to 7.61%, lipids from 0.98% to 1.48%, and dietary fiber from 2.93% to 4.96%.

These findings highlight both the distinctive nature of each plant's composition and the impact of varying water stress levels on the nutritional content of these plants. The findings have been validated by [26-28], revealing that water deficiency directly affects photosynthetic activity and the translocation of mineral salts from the soil to the roots. Additionally, [29-31] has affirmed that water scarcity influences both growth and nutrient supply. Similarly, [32-36] they discovered that environmental stresses have a detrimental impact on primary metabolites and their concentrations.

The results regarding the variation in mineral composition are depicted in Fig. 3, revealing that the most prevalent mineral compounds are Mg, Fe, Mg, K, and Mn. These minerals exhibit varying percentages, both among different plant species and within different samples of the same species. In terms of the highest percentage of mineral compounds, *Inula viscosa* takes the lead, followed by *Mentha pulegium*, *Thymus*

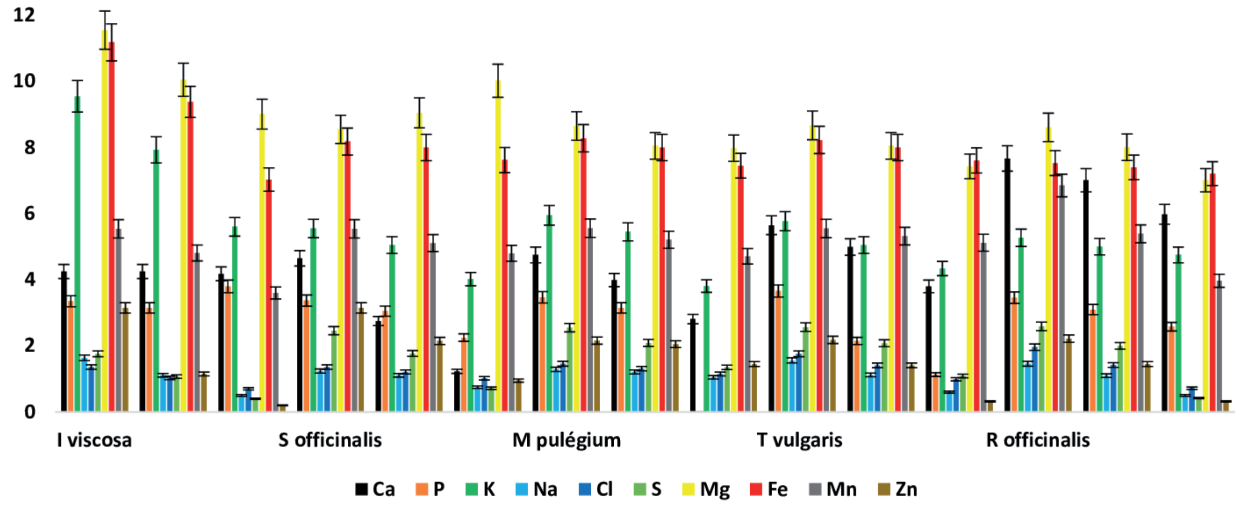

Fig. 3. Content of mineral compositions.

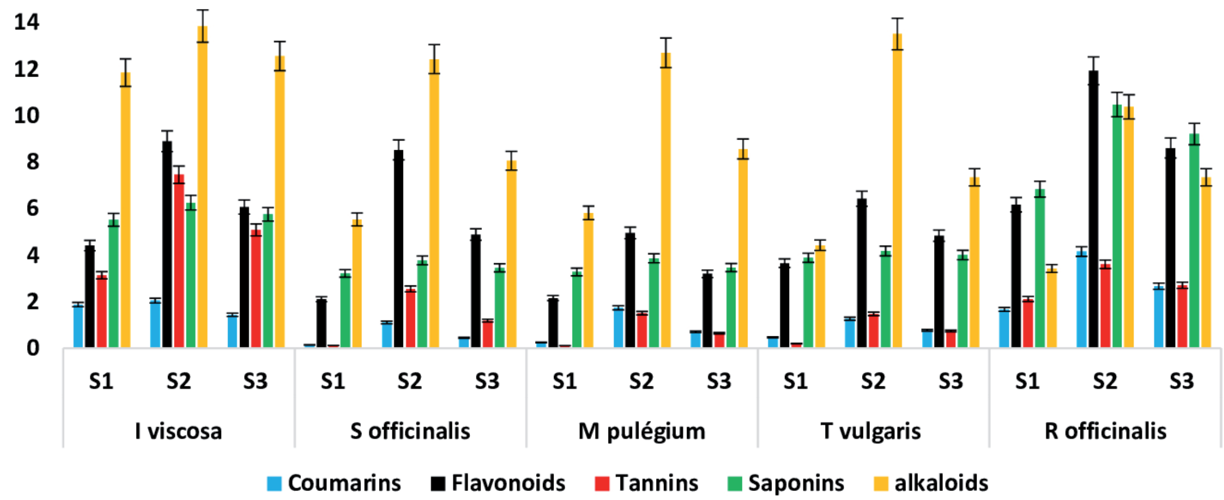

Fig. 4. Secondary metabolite content.

vulgaris, *Rosmarinus officinalis*, and finally *Salvia officinalis*. Thus, the percentage of mineral compounds decreased from sample one to sample two, to sample three by decreasing the irrigation, as found in [37], which found that water stress slows down the passage of water to the roots, which causes the passage of nutrients to decrease.

Secondary Metabolites

In Fig. 4, the percentages of secondary metabolites in the extracts from the five plants most widely used by the rural population are presented in relation to the decrease in water availability. Sample two consistently stands out with higher concentrations of these metabolites, while sample three generally exhibits relatively lower concentrations compared to sample one. Generally, the percentage of secondary metabolites in successive order is as follows: alkaloids, flavonoids, saponins, tannins, and coumarins.

For a more detailed breakdown of each plant's secondary metabolites with their respective percentages:

Inula viscosa: Alkaloids range from 11.85% to 13.85%, flavonoids from 4.42% to 8.9%, tannins from 3.14% to 7.46%, saponins from 5.52% to 6.26%, and coumarins from 1.44% to 2.05%.

Salvia officinalis: Alkaloids range from 5.54% to 12.43%, flavonoids from 2.11% to 8.53%, tannins from 0.12% to 2.55%, saponins from 3.22% to 3.78%, and coumarins from 0.15% to 1.11%.

Mentha pulegium: Alkaloids range from 5.82% to 12.7%, flavonoids from 2.16% to 4.96%, tannins from 0.11% to 1.51%, saponins from 3.28% to 3.87%, and coumarins from 0.24% to 1.74%.

Thymus vulgaris: Alkaloids range from 4.43% to 13.51%, flavonoids from 3.66% to 6.43%, tannins from 0.2% to 1.47%, saponins from 3.89% to 4.18%, and coumarins from 0.47% to 1.27%.

Rosmarinus officinalis: Alkaloids range from 3.43% to 10.38%, flavonoids from 6.17% to 11.93%, tannins

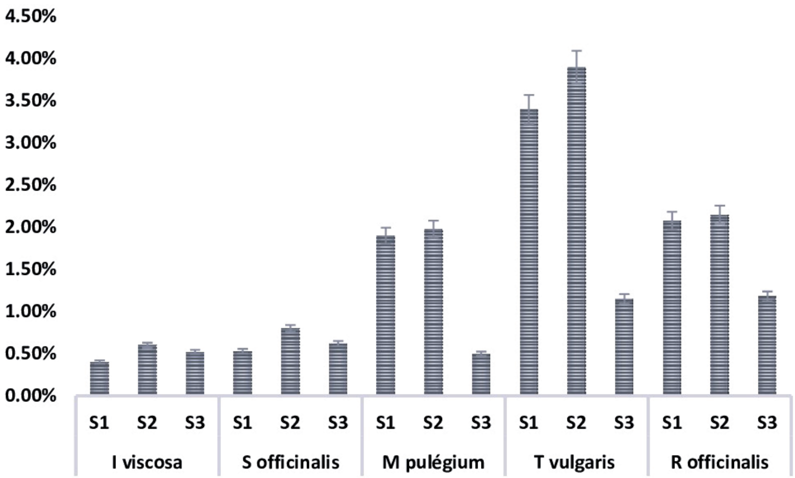

Fig. 5. Essential oil yield.

Table 1. Chemical compositions of essential oils of *Inula viscosa*.

Compounds	RI	S1	S2	S3
1 1,8-dehydro-cineole	991	0,09	0,17	0,21
2 n-nonanal	1100	0,09	0,21	0,34
3 para-mentha-1,5-diene-8-ol	1170	0,06	0,64	1,92
4 α-terpineol	1188	1,09	1,20	2,45
5 α-copaene	1376	3,36	2,98	4,38
6(E)-β-damascenone	1384	2,29	1,44	3,20
7(Z)-β-damascenone	1387	0,93	0,04	0,03
8 1-tetradecene	1389	1,05	1, 51	0, 90
9 α-cedrenes	1419	4,84	2, 77	1, 61
10 (E)-caryophyllene	1441	1,43	1,78	2,05
11 the aromatics	1455	0,09	1,11	1,78
12 geranyl acetone	1460	1,61	1,43	2,09
13 allo-aromatic dendrines	1467	1,19	1,94	2,05
14 cis-support adiene	1477	0,04	1,05	0,73
15 β-chamigrenes	1479	1,49	1,09	0,51
16 γ-muurolene	1490	0,05	1,07	0,90
17 β-cell size	1491	-	-	0,57
18 10,11-epoxy-calamenene	1492	3, 51	2, 60	-
19 δ-cell size	1493	2, 90	1, 81	1,89
20 cis-β-guaiene	1500	1,67	1,78	0,98
21 α-muurolene	1501	1,09	1,05	0,07
22 epizonarene	1505	0,71	1,03	0,02
23 α-cuprenene	1523	2, 09	3,15	1,82
24 δ-cadinene	1538	0, 80	1, 73	0,88
25 α-cadienene	1540	1, 39	2,03	2,78
26 α-copaen-11-ol	1545	1, 92	2,36	0,09
27 α-calacorene	1563	7,08	11,90	26,07

Table 1. Continued.

28(E)-nerolidol	1580	10,82	6,85	6,90
29 caryophyllene oxide	1589	2,35	2,94	1,95
30 1-hexadecene	1596	3,05	1,09	1,81
31 phokienol	1600	1,95	1,82	1,81
32 guaiol	1619	10,3	1,39	1,68
33 isongifolan-7-α-ol	1628	0,06	1,09	1,38
34 1-epi-cubenol	1631	2,63	2,93	0,05
35 muurola-4,10(14)-diene-1-β-	1632	1,92	3,92	3,06
36 gymnomitron	1640	0,05	1,01	1,54
37 epi-α-cadinol	1651	1,45	2,27	1,29
38 cedr-8(15)-en-9-α-ol	1653	9,17	7,98	4,75
39 α-eudesmol	1654	1,39	1,01	0,98
40 α-cadinol	1667	0,02	0,04	0,43
41 14-hydroxy-(Z)-	1669	0,91	1,61	1,47
42 14-hydroxy-9-epi-(E)-	1681	1,03	1,17	1,19
43 ishwarone	1699	0,03	0,03	0,48
44 epi-nootkatol	1747	0,04	0,98	0,69
45 8-α-11-elemodiol	1767	0,06	1,72	2,03
46 β-costol	1768	1,27	1,09	1,28
47 13-hydroxy-valencene	1790	2,79	2,89	2,69
	Total	94,91%	95,37%	96,84%

Table 2. Chemical compositions of essential oils of *Mentha pulegium.*

Compounds		Content of %		
	RI	S1	S2	S3
a-Pinene	937	1,4	0,1	1,7
Cyclohexanone-3-methyl	952	0,7	0,2	Tr
b-Pinene	974	0,1	0,3	0,2
Myrcene	992	0,5	0,1	0,5
Octanol-3	995	2,4	0,5	0,1
d-2-Carene	1003	Tr	0,1	0,7
Limonene	1030	1,3	0,1	0,5
p-Mentha-3,8-diene	1071	2,1	0,1	0,9
Menthone	1150	0,1	0,2	0,4
Pinocarvone	1166	1,3	0,1	1,9
Isomenthol	1182	0,8	-	0,1
Menthol	1171	2,6	0,1	3,8
Dihydrocarvone	1193	5,1	-	8,8
R(+)-pulegone	1236	71,6	73,4	61,1
Carvone	1240	5,4	-	10,1

Table 2. Continued.

a-Peperitone	1251	0,9	-	0,9
Piperitenone	1349	2,1	24,1	7,3
Caryophyllene	1418	0,2	0,1	0,3
Germacrene D	1475	0,1	0,1	0,1
g-Eudesmol	1630	0,4	0,1	0,2
a-Eudesmol	1649	0,6	0,1	0,1
	Total	99,3%	99,7%	99,7%

Table 3. Chemical compositions of essential oils of *Thymus vulgaris*.

Compounds		Content of %		
	RI	S1	S2	S3
a-Pinene	937	3,7	1,1	1,5
Sabinene	965	0,4	0,2	0,2
b-Pinene	975	2,8	0,9	0,1
Myrcene	984	0,1	0,7	0,4
α-Terpinene	1009	1,4	0,1	2,4
p-Cymene	1013	8,5	0,4	3,9
1,8-Cineole	1025	0,1	0,3	0,5
Limonene	1032	0,9	1,7	1,8
Y-Terpinene	1050	0,2	0,9	22,3
linalol	1086	0,8	0,5	0,4
Camphre	1127	0,9	2,8	2,1
transPinocarveol	1127	0,7	0,3	0,3
Borneol	1153	0,7	0,6	0,2
Terpinen-4-ol	1165	0,2	0,3	1,8
Carvacrylmethylether	1231	0,1	0,6	0,1
Thymol	1290	6,9	4,1	34,7
Carvacrol	1298	69,4	83,3	26,1
(E)-Caryophyllene	1420	0,1	0,3	0,1
Aromadendrene	1438	0,1	0,1	-
Alloaromadendrene	1458	0,1	0,1	-
Ledene	1493	-	0,1	0,6
Spathulenol	1564	-	0,1	-
Caryophyllene oxyde	1571	0,1	0,1	0,1
Total		98,2%	99,6%	99,6%

from 2.12% to 3.61%, saponins from 6.84% to 10.48%, and coumarins from 1.67% to 4.16%.

These results underscore how variations in water availability impact the composition of secondary metabolites in these plants. Sample two consistently displays higher concentrations, suggesting its response to fluctuating environmental conditions. These findings carry implications for comprehending the potential bioactive properties of these plants and their adaptability to fluctuating water conditions, which hold

Table 4. Chemical compositions of essential oils of *Rosmarinus officinalis.*

Compounds	RI	Content of %		
		S1	S2	S3
Alpha-pinene	939	9,85	8,23	9,04
Camphene	954	4,22	4,04	4,08
Beta-pinene	979	3,30	0,34	9,17
a-Terpinene	1017	0,66	0,14	0,56
p-Cymene	1025	2,59	0,36	2,08
Limonene	1028	0,05	Tr	Tr
Cineole	1030	45,44	55,71	43,07
Beta-myrcene	1048	3,35	1,44	1,56
Linalool	1097	0,99	0,12	0,85
Camphre	1146	17,89	21,93	21,99
Bornéole	1169	1,99	2,44	0,41
a-Terpineole	1199	3,92	4,05	1,38
Verbenone	1205	0,54	0,13	0,65
Acetate de Bornyle	1289	5,09	1,03	5,42
B-Caryophyllene	1419	0,06	0,02	0,06
a-Caryophyllene	1423	0,04	Tr	0,12
Total		99,98%	99,98%	99,98%

significant relevance for rural communities [38] Water stress has been identified as a factor that can impact and elevate the content of secondary metabolites [39]. This observation aligns with [40], who determined that high levels of secondary metabolites are associated with stress conditions. Furthermore, [41] noted that water-deficient plants exhibit heightened levels of secondary metabolites. In concurrence, [42] and [43] found that plants under adverse climatic conditions display the highest concentrations of secondary metabolites. Also [39-42] confirmed that stressful environmental conditions lead to the modification of the secondary metabolite profile of plants.

Essential Oils Yield and Gas Chromatography

The data regarding the yield of essential oil is depicted in the graph presented in Fig. 5. It's evident that *Thymus vulgaris* yields the highest amount of essential oil, followed by *Rosmarinus officinalis*, *Mentha pulegium*, *Salvia officinalis*, and lastly, *Inula viscosa*. Across all five plants, sample two consistently stands out with the highest essential oil yield compared to the other two samples. This observation implies that sample two demonstrates the highest essential oil production efficiency for each of the studied plant species, as supported by [48], which indicates that plants experiencing moderate stress exhibit the highest essential oil yield compared to those under severe stress.

Additionally, [49, 50] reported that non-irrigated plants display the highest content of essential oils [51], and those who found that oil yield increases under stress conditions, [52] indicated that increasing temperatures can increase essential oil content. Thus, [48-52] found that water stress and increased temperature can cause alterations in the physiological and morphological processes of plants and the yield and chemical compositions of essential oils.

The chemical composition analysis of essential oils from the five plants (Tables 1, 2, 3, 4, and 5) reveals that the primary compounds in these essential oils are influenced by the reduction in irrigation. In most of the plants studied, the major compounds tend to increase under moderate water stress conditions. However, as environmental conditions deteriorate further, the predominant compounds in the essential oils begin to decrease. This suggests that the composition of essential oils is dynamic and responsive to changes in water availability, with a shift towards increased production of major compounds under moderate stress, but a subsequent decline as water stress intensifies [58]. The content of thymol increases with water stress, as did [59], who found that the yield of camphor increases with increasing levels of water stress, and [60], who obtained that the yield of 1,8-cineole, camphene, and β-pinene increases under stressful conditions.

The identification of major compounds in *Mentha pulegium* essential oils, such as polygonum, piperitenone,

Table 5. Chemical compositions of essential oils of *Salvia*.

Compounds	Content of %		
	S1	S2	S3
α-thuyone	0,87	10,89	0,06
manool	11,78	13,01	13,12
β-caryophyllène	12,54	6,44	14,0
α-humulène	10,71	0,67	10,04
viridiflorol	6,08	7,01	6,08
1,8-cinéol	8,80	7,08	9,0
manool	7,70	0,55	6,95
β-caryophyllène	8,78	4,60	8,20
camphre	6,11	4,84	5,84
β-thuyone	3,75	3,18	2,70
atisirène	0,01	7,78	0,02
β-pinène	0,01	4,97	0,03
myrcène	3,01	0,01	4,10
bornéol	0,97	4,09	0,04
cis-hydrate de sabinène	0,95	2,76	0,06
camphène	1,08	1,89	1,85
α-pinène	1,84	1,01	1,93
α-humulène	0,01	2,79	0,06
limonène	2,76	0,65	2,86
α-terpinène	0,29	0,01	0,70
allo-aromadendrène	0,56	0,01	0,34
γ-terpinène	0,01	4,29	0,01
trans-pinocamphone	0,01	2,02	0,01
cis-hydrate de sabinène	0,10	0,06	0,17
acétate de bornyle	0,19	1,52	0,1
atisirène	0,30	0,08	0,77
α-terpinolène	0,33	2,90	0,1
Total	90,55%	95,12%	89,15%

carvone, and dihydrocarvone offer valuable insights into the chemical makeup of this medicinal plant. However, the influence of climate change on the abundance and composition of these compounds adds another layer of complexity to our understanding of *Mentha pulegium* essential oil production. Climate factors, specifically temperature and precipitation, play a pivotal role in shaping the chemical profile of these essential oils.

As illustrated in the accompanying figure, variations in climate conditions result in differences in the yield and distribution of these compounds among different samples. Pulegone, identified as the primary compound, exhibits varying percentages across the samples, with sample two demonstrating the highest concentration. These findings underscore the sensitivity of *Mentha pulegium* to climate change and its potential impact on essential oil production. Comprehending the underlying mechanisms behind these variations is crucial for predicting and managing the effects of climate change on medicinal plant resources. Extensive analysis and investigation are needed to unravel the intricate relationship between climate parameters and the adaptive responses of *Mentha pulegium*. This will provide invaluable insights into its medicinal properties and potential implications for traditional medicine and the development of natural products [47-49]. Additionally, other studies, such as those cited in [64, 65], support the finding that water stress significantly affects the abundance of the majority of compounds. Moreover, [66] asserts that ecological factors exert a substantial influence on the predominant compounds found in plants. These collective findings underscore the significant impact of stressors, encompassing water stress and ecological factors, on the composition and concentration of major compounds within medicinal plants. Such insights are of utmost importance for comprehending how these plants adapt to environmental conditions. They can also play a pivotal role in fine-tuning cultivation practices, thereby ensuring the quality and efficacy of plant-based products, especially in the context of shifting environmental factors. This knowledge is invaluable for sustainable resource management and the continued utilization of these plants for various applications, including traditional medicine and natural product development. The chemical compounds of plant essential oils are also modified by stressful conditions, as they found [67-71].

Conclusions

Aromatic and medicinal plants hold a pivotal role in our daily lives, emphasizing the pressing need to preserve them to ensure their sustainability for future generations. Currently, due to the combined impact of climate change and human activities, this natural wealth is deteriorating, both in terms of biodiversity and physiological and morphological aspects. It becomes imperative to anticipate the consequences of climate change on the physiological processes of these plants.

The results obtained from our study reveal that, across all three samples of the five plant species examined, the content of carbohydrates, lipids, proteins, and dietary fibers decreases as the level of irrigation decreases. In contrast, for secondary metabolites, sample two consistently displays the highest percentage compared to the other two samples. Similarly, in the case of essential oil yield, there is an initial increase in the content of essential oils during the first two years. However, as climatic conditions become more severe, the essential oil yield starts to decline. Moreover, chromatographic analysis illustrates that the major compounds within the essential oils of these plants undergo alterations under stress conditions.

Based on these findings, it becomes clear that the preservation of these crucial plant resources and an in- depth understanding of their responses to environmental stressors are essential for ensuring their sustained availability and the quality of essential oils and other plant-based products.

Conflict of Interest

The authors declare no conflict of interest.

Fundings

This research is funded by Princess Nourah bint Abdulrahman University Researchers Supporting Project number (PNURSP2023R147), Princess Nourah bint Abdulrahman University, Riyadh, Saudi Arabia.

Reference

1. SOW E.H., BATHIERY O. vulnérabilité de la commune de djilor (région de fatick) aux changements climatiques et stratégies d'adaptation des communautés. African Journal on Land Policy and Geospatial Sciences. **5**, 832, **2022**.
2. BOUCHMAA N., MRID R.B., KABACH I., ZOUAOUI Z., KARROUCHI K., CHTIBI H., ZYAD A., CACCIOLA F., NHIRI M. Beta vulgaris subsp maritima: a valuable food with high added health benefits. Applied Sciences. **12**, 1866, **2022**.
3. ASRAOUI F., KOUNNOUN A., CADI H.E., CACCIOLA F., MAJDOUB Y.O.E., ALIBRANDO F., MANDOLFINO F., DUGO P., MONDELLO L., LOUAJRI A. phytochemical investigation and antioxidant activity of globularia alypum l. Molecules. **26**, 759, **2021**.
4. BAALI F., BOUMERFEG S., NAPOLI E., BOUDJELAL A., RIGHI N., DEGHIMA A., BAGHIANI A., RUBERTO G. Chemical composition and biological activities of essential oils from two wild algerian medicinal plants: *mentha pulegium* l. and *lavandula stoechas* l. Journal of Essential Oil Bearing Plants. **22**, 821, **2019**.
5. BOUYAHYA A., ET-TOUYS A., BAKRI Y., TALBAUI A., FELLAH H., ABRINI J., DAKKA N. Chemical composition of *mentha pulegium* and *rosmarinus officinalis* essential oils and their antileishmanial, antibacterial and antioxidant activities. Microbial Pathogenesis. **111**, 41, **2017**.
6. CHO N.H., SHAW J.E., KARURANGA S., HUANG Y., DA ROCHA FERNANDES J.D., OHLROGGE A.W., MALANDA B. IDF diabetes atlas: global estimates of diabetes prevalence for 2017 and projections for 2045. Diabetes Research and Clinical Practice. **138**, 271, **2018**.
7. MARONE D., MASTRANGELO A.M., BORRELLI G.M., MORES A., LAIDÒ G., RUSSO M.A., FICCO D.B.M. Specialized metabolites: physiological and biochemical role in stress resistance, strategies to improve their accumulation, and new applications in crop breeding and management. Plant Physiology and Biochemistry. **172**, 48, **2022**.
8. TAÏBI S., ANZA F.Z.H., ZEROUAL S. Etude de l'impact des changements climatiques sur la disponibilité des ressources en eau basée sur les simulations du modèle climatique régional RCA4 : cas du bassin de Ain DALIA (Algérie). Algerian Journal of Environmental Science and Technology, **7** (2), **2021**.
9. ASKARY M., BEHDANI M.A., PARSA S., MAHMOODI S., JAMIALAHMADI M. Water stress and manure application affect the quantity and quality of essential oil of Thymus daenensis and *Thymus vulgaris*. Industrial Crops and Products. **111**, 336, **2018**.
10. SHAWON R.A., KANG B.S., LEE S.G., KIM S.K., JU LEE H., KATRICH E., GORINSTEIN S., KU Y.G. Influence of drought stress on bioactive compounds, antioxidant enzymes and glucosinolate contents of Chinese cabbage (Brassica rapa). Food Chemistry. **308**, 125657, **2020**.
11. DEMBELE F., SAMAKE S., TOGOLA I., KAREMBE M. Etudes ethnobotanique et phytochimique despeces ornementales utilisees au Mali: cas de *Terminalia catappa* (L.) dans le district de Bamako. International Journal of Applied Research. **7**, 01, **2021**.
12. GÖSSLING S., DOLNICAR S. A review of air travel behavior and climate change. WIREs Climate Change. **14**, **2023**.
13. WANG Y., PINEDA-MUNOZ S., MCGUIRE J.L. Plants maintain climate fidelity in the face of dynamic climate change. Proceedings of the National Academy of Sciences U.S.A. **120**, e2201946119, **2023**.
14. SAUTNER Z., VAN LENT L., VILKOV G., ZHANG R. firm-level climate change exposure. The Journal of Finance. **78**, 1449, **2023**.
15. ROBBINS SCHUG G., BUIKSTRA J.E., DEWITTE S.N., BAKER B.J., BERGER E., BUZON M.R., DAVIES-BARRETT A.M., GOLDSTEIN L., GRAUER, A.L., GREGORICKA L.A., HALCROW S.E., KNUDSON, K.J., LARSEN C.S., MARTIN D.L., NYSTROM K.C., PERRY M.A., ROBERTS C.A., SANTOS A.L., STOJANOWSKI C.M., SUBY J.A., TEMPLE D.H., TUNG T.A., VLOK M., WATSON-GLEN T., ZAKRZEWSKI S.R. Climate change, human health, and resilience in the Holocene. Proceedings of the National Academy of Sciences U.S.A. **120**, e2209472120, **2023**.
16. SIMPSON N.P., MACH K.J., CONSTABLE A., HESS J., HOGARTH R., HOWDEN M., LAWRENCE J., LEMPERT R.J., MUCCIONE V., MACKEY B., NEW M.G., O'NEILL B., OTTO F., PÖRTNER H.-O., REISINGER A., ROBERTS D., SCHMIDT D.N., SENEVIRATNE S., STRONGIN S., VAN AALST M., TOTIN E., TRISOS C.H. A framework for complex climate change risk assessment. One Earth. **4**, 489, **2021**.
17. NOGUEIRA L.M., CRANE T.E., ORTIZ A.P., D'ANGELO H., NETA G. climate change and cancer. Biomarkers & Preventio. **32** (7), 869, **2023**.
18. LAFTOUHI A., ELOUTASSI N., ECH-CHIHBI E., RAIS Z., ABDELLAOUI A., TALEB A., BENIKEN M., NAFIDI H.-A., SALAMATULLAH A.M., BOURHIA M., TALEB M. The impact of environmental stress on the secondary metabolites and the chemical compositions of the essential oils from some medicinal plants used as food supplements. Sustainability. **15**, 7842, **2023**.
19. TOGOLA I., KONARÉ M.A., DIAKITÉ M., DIARRA N., TOUNKARA F., SANOGO R., DEMBÉLÉ D. stages of development of datura innoxia mill., a plant used in traditional medicine in mali. American Journal of Innovative Research and Applied Sciences. **9** (2), 200, **2019**.
20. SOULAMA S., NACOULMA O., NAGTIERO MEDA R., BOUSSIM J., MILLOGO-RASOLODIMBY J. Teneurs

en coumarines de 15 ligneux fourragers du Burkina Faso. International Journal of Biological and Chemical Sciences. **7**, 2283, **2014**.

21. BAGUIA-BROUNE F.D.M., N'GAMAN K.C.C., MAMYRBEKOVA-BEKRO J.A., VIRIEUX D. Saponines des racines de Securidaca longipedunculata (Polygalaceae): quantification et évaluation anti-oxydante. Revue Nature et Technologie. **10** (2), 25, **2018**.
22. LAFTOUHI A., ELOUTASSI N., DRIOUA S., ECH-CHIHBI E., RAIS Z., ABDELLAOUI A., TALEB A., BENIKEN M., TALEB M. impact of water stress and temperature on metabolites and essential oil of *Rosmarinus officinalis* (phytochemical screening, extraction, and gas chromatography). Journal of Ecological Engineering. **24**, 237, **2023**.
23. HAMADOU H.H., KALLO M.S., MANZO L.M., MOUSSA I., ADAMOU R. Criblage phytochimique et dosage des polyphénols du Detarium microcarpum Guill. et Perr. utilisé dans le traitement des maladies parasitaires au Niger. Afrique SCIENCE. **14** (5), 390, **2018**.
24. BOUTERFAS K., MEHDADI Z., LATRECHE A., HAZEM Z. Quantification de quelques polyphénols de Marrubium vulgare L. du mont de Tessala (Algérie occidentale) pendant les deux périodes de végétation et de floraison. Les technologies de laboratoire, **8** (31), **2013**.
25. BENIAICH G., HAFSA O., MALIKI I., BIN JARDAN Y.A., EL MOUSSAOUI A., CHEBAIBI M., AGOUR A., ZOUIRECH O., NAFIDI H.A., KHALLOUKI F., BOURHIA M., TALEB M. GC-MS characterization, in vitro antioxidant, antimicrobial, and in silico nadph oxidase inhibition studies of anvillea radiata essential oils. Horticulturae. **8**, 886, **2022**.
26. SOULAMA S., NACOULMA O., NAGTIERO MEDA R., BOUSSIM J., MILLOGO-RASOLODIMBY J. Teneurs en coumarines de 15 ligneux fourragers du Burkina Faso. International Journal of Biological and Chemical Sciences. **7**, 2283, **2014**.
27. WEATHERS M., HATHAWAY J.M., TIRPAK R.A., KHOJANDI A. evaluating the impact of climate change on future bioretention performance across the contiguous united states. Journal of Hydrology. **616**, 128771, **2023**.
28. ZHU Y., XIONG C., WEI Z., CHEN Q., MA B., ZHOU S., TAN J., ZHANG L., CUI H., DUAN G. Impacts of global change on the phyllosphere microbiome. New Phytologist. **234**, 1977, **2022**.
29. APPLEQUIST W.L., BRINCKMANN J.A., CUNNINGHAM A.B., HART R.E., HEINRICH M., KATERERE D.R., VAN ANDEL T. Scientists' warning on climate change and medicinal plants. Planta Medica. **86**, 10, **2020**.
30. YANG H., XIAO X., LI J., WANG F., MI J., SHI Y., HE F., CHEN L., ZHANG F., WAN X. Chemical compositions of walnut (*Juglans* spp.) oil: combined effects of genetic and climatic factors. Forests. **13**, 962, **2022**.
31. HESSINI K., WASLI H., AL-YASI H.M., ALI E.F., ISSA A.A., HASSAN F.A.S., SIDDIQUE K.H.M. graded moisture deficit effect on secondary metabolites, antioxidant, and inhibitory enzyme activities in leaf extracts of rosa damascena mill. var. trigentipetala. Horticulturae. **8**, 177, **2022**.
32. AHAMMED G.J., WU Y., WANG Y., GUO T., SHAMSY R., LI X. Epigallocatechin-3-Gallate (EGCG): A unique secondary metabolite with diverse roles in plant-environment interaction. Environmental and Experimental Botany. **209**, 105299, **2023**.
33. HUMBAL A., PATHAK B. Influence of exogenous elicitors on the production of secondary metabolite in plants: A review ("VSI: secondary metabolites"). Plant Stress. **8**, 100166, **2023**.
34. KOPECKÁ R., KAMENIAROVÁ M., ČERNÝ M., BRZOBOHATÝ B., NOVÁK J. International Journal of Molecular Sciences. **24**, 6603, **2023**.
35. MACHADO J., VASCONCELOS M.W., SOARES C., FIDALGO F., HEUVELINK E., CARVALHO S.M.P. young tomato plants respond differently under single or combined mild nitrogen and water deficit: an insight into morphophysiological responses and primary metabolism. Plants. **12**, 1181, **2023**.
36. PÉREZ-LLORCA M., POLLMANN S., MÜLLER M. ethylene and jasmonates signaling network mediating secondary metabolites under abiotic stress. International Journal of Molecular Sciences. **24**, 5990, **2023**.
37. ALHAITHLOUL H.A., SOLIMAN M.H., AMETA K.L., EL-ESAWI M.A., ELKELISH A. Changes in ecophysiology, osmolytes, and secondary metabolites of the medicinal plants of mentha piperita and catharanthus roseus subjected to drought and heat stress. Biomolecules. **10**, 43, **2019**.
38. GAO S., WANG Y., YU S., HUANG Y., LIU H., CHEN W., HE X. Effects of drought stress on growth, physiology and secondary metabolites of two adonis species in northeast china. Scientia Horticulturae. **259**, 108795, **2020**.
39. JAN R., ASAF S., NUMAN M., LUBNA K.M. Plant secondary metabolite biosynthesis and transcriptional regulation in response to biotic and abiotic stress conditions. Agronomy. **11**, 968, **2021**.
40. AHMED S., GRIFFIN T.S., KRANER D., SCHAFFNER M.K., SHARMA D., HAZEL M., LEITCH A.R., ORIANS C.M., HAN W., STEPP J.R., ROBBAT A., MATYAS C., LONG C., XUE D., HOUSER R.F., CASH S.B. Environmental factors variably impact tea secondary metabolites in the context of climate change. Frontiers in Plant Science. **10**, 939, **2019**.
41. ASHRAF M.A., IQBAL M., RASHEED R., HUSSAIN I., RIAZ M., ARIF M.S. Environmental stress and secondary metabolites in plants. in: plant metabolites and regulation under environmental stress. Elsevier, pp. 153, **2018**.
42. SANCHO-KNAPIK D., SANZ M.Á., PEGUERO-PINA J.J., NIINEMETS Ü., GIL-PELEGRÍN E. Changes of secondary metabolites in Pinus sylvestris L. needles under increasing soil water deficit. Annals of Forest Science. **74**, 24, **2017**.
43. CHÁVEZ-ARIAS C.C., RAMÍREZ-GODOY A., RESTREPO-DÍAZ H. Influence of drought, high temperatures, and/or defense against arthropod herbivory on the production of secondary metabolites in maize plants. A review. Current Plant Biology. **32**, 100268, **2022**.
44. BORIM A.J., OCAMPOS F.M.M., CATOIA R., DOKKEDAL A.L., COLNAGO L.A., CECHIN I., SALDANHA L.L. NMR-based metabolomics reveals effects of water stress in the primary and specialized metabolisms of *Bauhinia ungulata* l. (fabaceae). Metabolites. **13**, 381, **2023**.
45. LÓPEZ-HIDALGO C., LAMELAS L., CAÑAL M.J., VALLEDOR L., MEIJÓN M. Untargeted metabolomics revealed essential biochemical rearrangements towards combined heat and drought stress acclimatization in Pinus pinaster. Environmental and Experimental Botany. **208**, 105261, **2023**.
46. NICOLAS-ESPINOSA J., GARCIA-IBAÑEZ P., LOPEZ-ZAPLANA A., YEPES-MOLINA L., ALBALADEJO-

MARICO L., CARVAJAL M. Confronting secondary metabolites with water uptake and transport in plants under abiotic stress. International Journal of Molecular Sciences. **24**, 2826, **2023**.

47. SALAM U., ULLAH S., TANG Z.H., ELATEEQ A.A., KHAN Y., KHAN J., KHAN A., ALI S. Plant metabolomics: an overview of the role of primary and secondary metabolites against different environmental stress factors. Life. **13**, 706, **2023**.
48. SARMOUM R., HAID S., BICHE M., DJAZOULI Z., ZEBIB B., MERAH O. Effect of salinity and water stress on the essential oil components of rosemary (*Rosmarinus officinalis* l.). Agronomy. **9**, 214, **2019**.
49. KESHAVARZ, H. Study of water deficit conditions and beneficial microbes on the oil quality and agronomic traits of canola (*Brassica napus* L.). Grasas y Aceites. **71**, 373, **2020**.
50. RAHIMI A., MOHAMMADI M.M., SIAVASH MOGHADDAM S., HEYDARZADEH S., GITARI H. Effects of stress modifier biostimulants on vegetative growth, nutrients, and antioxidants contents of garden thyme (*thymus vulgaris* l.) under water deficit conditions. Journal of Plant Growth Regulation. **41**, 2059, **2022**.
51. XU B., GAO M., HU W., ZHAO W., WANG S., ZHOU Z. Individual and interactive influences of elevated air temperature and soil drought at the flowering and boll-forming stage on cottonseed yield and nutritional quality. The Crop Journal. **10**,128, **2022**.
52. SABER M., HARHAR H., EL HATTABI L., ZENGIN G., BOUYAHYA A., TABYAOUI M. Chemical composition and antioxidant activities of essential oils and extracts from cones of Tetraclinis articulata (Vahl) Masters. International Journal of Secondary Metabolite. **8**, 352, **2021**.
53. ABAZA A.S.D., ELSHAMLY A.M.S., ALWAHIBI M.S., ELSHIKH M.S., DITTA A. Impact of different sowing dates and irrigation levels on NPK absorption, yield and water use efficiency of maize. Scientific Reports. **13**, 12956, **2023**.
54. ASGHARI J., MAHDAVIKIA H., REZAEI-CHIYANEH E., BANAEI-ASL F., AMANI MACHIANI M., HARRISON M.T. Selenium nanoparticles improve physiological and phytochemical properties of basil (*Ocimum basilicum* l.) under drought stress conditions. Land. **12**, 164, **2023**.
55. ASLANI Z., HASSANI A., ABDOLLAHI MANDOULAKANI B., BARIN M., MALEKI R. Effect of drought stress and inoculation treatments on nutrient uptake, essential oil and expression of genes related to monoterpenes in sage (*Salvia officinalis*). Scientia Horticulturae. **309**, 111610, **2023**.
56. MAHMOUD N., ABDOU M.A.H., SALAHELDIN S., SOLIMAN W.S., ABBAS A.M. The impact of irrigation intervals and npk/yeast on the vegetative growth characteristics and essential oil content of lemongrass. Horticulturae. **9**, 365, **2023**.
57. PUNETHA A., KUMAR D., CHAUHAN A., SURYAVANSHI P., PADALIA R.C., UPADHYAY R.K., VENKATESHA K.T. Soil moisture stress induced changes in essential oil content and bioactive compounds in German chamomile (*Chamomilla recutita* L.). Journal of Essential Oil Research. **35**, 289, **2023**.
58. KHALID M.F., ZAKIR I., KHAN R.I., IRUM S., SABIR S., ZAFAR N., AHMAD S., ABBAS M., AHMED T., HUSSAIN S. Effect of water stress (drought and waterlogging) on medicinal plants. Springer Nature, pp. 169, **2023**.
59. PODDA A., POLLASTRI S., BARTOLINI P., PISUTTU C., PELLEGRINI E., NALI C., CENCETTI G., MICHELOZZI M., FRASSINETTI S., GIORGETTI L., FINESCHI S., DEL CARRATORE R., MASERTI B. Drought stress modulates secondary metabolites in *Brassica oleracea* L. convar. *acephala* (DC) Alef, var. *sabellica* L. Journal of the Science of Food and Agriculture **99**, 5533, **2019**.
60. FARHOUDI R. Effet du stress de la sécheresse sur les constituants chimiques, la photosynthèse et les propriétés antioxydantes de l'huile essentielle de *Rosmarinus officinalis*. Journal of Medicinal Plants and By-products. **1**, 17, **2013**.
61. AHMED Z., EL HASSAN S., FATIMA M., MAHDI C., ABDELLAH C., MUSTAPHA T. Effects of extraction technique and solvent on phytochemicals, antioxidant, and antimicrobial activities of cultivated and wild rosemary (*Rosmarinus officinalis* l.) from taounate region (northern morocco). Biointerface Research in Applied Chemistry. **12**, 8441, **2021**.
62. HASNAE C., KAMAL H., ADIL E., NOUREDDINE E., ABDELAZIZ S., FOUAD M., DIL T., SHIV K. Heat and Drought Stress Impact on Phenology, Grain Yield, and Nutritional Quality of Lentil (*Lens culinaris Medikus*). Frontiers in Nutrition. **7**, **2020**.
63. ALBERGARIA E.T., OLIVEIRA A.F.M., ALBUQUERQUE U.P. The effect of water deficit stress on the composition of phenolic compounds in medicinal plants. South African Journal of Botany. **131**, 12, **2020**.
64. CHANSON L.,JIMING J., JIANQIANG I. Effects of severe water stress on maize growth processes in the field. Sustainability. **11**, 5086, **2019**.
65. HAN Z., ZUO Q., WANG C., GAN R. Impacts of climate change on natural runoff in the yellow river basin of china during 1961-2020. Water. ***15***, 929, **2023**.
66. EL HADDAD N., CHOUKRI H., GHANEM M.E., SMOUNI A., MENTAG R., RAJENDRAN K., HEJJAOUI K., MAALOUF F., KUMAR S. High-temperature and drought stress effects on growth, yield and nutritional quality with transpiration response to vapor pressure deficit in lentil. Plants**.** **11**, 95, **2021**.
67. BOUREMANI N., CHERIF-SILINI H., SILINI A., BOUKET A.C., LUPTAKOVA L., ALENEZI F.N., BARANOV O., BELBAHRI L. Plant growth-promoting rhizobacteria (pgpr): a rampart against the adverse effects of drought stress. Water. **15**, 418, **2023**.
68. DEL BUONO D., REGNI L., PROIETTI, P. Abiotic stresses, biostimulants and plant activity. Agriculture. **13**, 191, **2023**.
69. GHADIRNEZHAD SHIADE S.R., FATHI A., TAGHAVI GHASEMKHEILI F., AMIRI E., PESSARAKLI M. Plants' responses under drought stress conditions: Effects of strategic management approaches – a review. Journal of Plant Nutrition. **46**, 2198, **2023**.
70. GUL M.U., PAUL A., CHEHRI A. Hydrotropism: understanding the impact of water on plant movement and adaptation. Water. **15**, 567, **2023**.
71. STRADA S., FERNÁNDEZ-MARTÍNEZ M., PEÑUELAS J., BAUWENS M., STAVRAKOU T., VERGER A., GIORGI F. Disentangling temperature and water stress contributions to trends in isoprene emissions using satellite observations of formaldehyde. 2005-2016. Atmospheric Environment. **295**, 119530, **2023**.