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An Investigation on the Morphology, Anatomy and Ecology of *Origanum onites* L.¹

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Abstract: Possible variations in the morphology, anatomy, ecology and phenology of *Origanum onites* L. (*Lamiaceae/Labiatae*) in relation to altitudinal variations was studied together with the differences in etheric oil content.

A decrease in the stem and lower leaf lengths was observed compared with an increase in the cortical area and vascular tissues in parallel with altitudinal increases. The number and size of stomata also varied with this factor. Etheric oil content was found to be different in relation to altitude.

Key Words: *Origanum onites*, morphology, anatomy, ecology.

Origanum onites'in morfolojisi, anatomisi ve ekolojisi üzerinde bir araştırma

Özet: Bu çalışmada, *Origanum onites* L.'in (*Lamiaceae/Labiatae*) yükseklik değişikliklerine bağlı olarak morfolojik, anatomik, ekolojik ve fenolojik özelliklerindeki değişiklikler ile eterik yağ miktarlarındaki farklılıklar araştırılmıştır.

Yükseklik artışına paralel olarak iletim dokusu ve korteks alanlarında bir artışa benzer şekilde bitkinin gövde ve alt yaprak boylarında bir azalma gözlenmiştir. Keza bu faktör ile stoma sayısı ve büyüklüğünde değişiklikler meydana gelmiştir. Eterik yağ miktarının yüksekliğe paralel olarak değiştiği görülmüştür.

Anahtar Sözcükler: *Origanum onites*, morfoloji, anatomi, ekoloji.

Introduction

Origanum onites L. is a member of the *Labiatae* family with many stems up to 65 cm tall and branches up to 13 cm long (1). It is commonly known as thyme, İzmir thyme, Bilyalı thyme or White thyme. 2-3 % of volatile oil is obtained from its aboveground parts through steam distillation. This oil includes phenolic substances such as carvacrol and thymol, (2). Thymol is one of the oldest antibiotics. In medicinal treatments it is used as an antiseptic, antibacterial, antispasmodic, antiasthmatic, expectorant and as a fungicide due to its volatile oils and terpenic materials. Thymol is also used as a starter in the synthesis of some pharmacological active ingredients, for tooth filling, in the preparation of antiseptic baths and soaps and in the preservation of some foods. It is commonly used as a spice also, and is thus exported in large quantities (3, 4).

Studies on the productivity and ontogenetic variability of *O. onites* under the ecological conditions of Bornova have been carried out and it has been noted that an ontogenetic variability exists in the amount of thymol + carvacrol and volatile oil (5). The same report reveals that the effective materials in the plant exist in variable amounts depending on different times of day and vegetative period. It has also been emphasized that the distillates of *Origanum* oil extracted from *Origanum* L. species, collected from different sites and plant species also varies (6).

Several authors (7, 8) have emphasized the effect of altitudinal variations in the plant morphogenesis.

The aim of this study was to investigate changes occurring in the morphology and anatomy as well as in the amount of volatile oil in *O. onites* in relation to factors which change in parallel with an increase in altitude.

Materials and Methods

Origanum onites L. (*Labiatae*) samples were collected from the lowest to the highest possible sites of İzmir Çatalkaya (1040 m), Spil (1514 m), Nif (1506 m) and Ödemiş-Bozdağ (2150 m). The specimens of *O. onites*

were identified by Yusuf Gemici and Erkuter Leblebici from Ege University and preserved in the herbarium center Afyon Kocatepe University.

Specimens investigated:

Plant sample	Herbarium number	Collection data
<i>Origanum onites</i> L.	A.K.Ü.M. 03	B2 Manisa : Spil mountain near Mevlevihane 180 m A. Gönüz. 28.05.1993
"	A.K.Ü.M. 04	B2 Manisa : Spil mountain near Sultanyaylası 340 m A. Gönüz 28.05.1993
"	A.K.Ü.M. 05	B2 Manisa : Spil mountain near Güvercinkaya 700 m A.Gönüz 28.05.1993
"	A.K.Ü.M. 06	B1 İzmir : Kemalpaşa Nif mountain near Kale üstü 300 m A. Gönüz 06.06.1993
"	A.K.Ü.M. 07	B1 İzmir : Kemalpaşa Nif mountain near Kozludere 600 m A. Gönüz 06.06.1993
"	A.K.Ü.M. 08	B1 İzmir : Kemalpaşa Nif mountain near Burçak tarlası 900 m A. Gönüz 06.06.1993
"	A.K.Ü.M. 09	B2 Manisa : Salihli Bozdağ- Ödemiş near road 400 m A. Gönüz 02.07.1993
"	A.K.Ü.M. 10	B2 Manisa : Salihli Bozdağ-Ödemiş near road 900 m A. Gönüz 02.07.1993
"	A.K.Ü.M. 11	B1 İzmir : Çatalkaya near road 270 m A. Gönüz 20.06.1993
"	A.K.Ü.M. 12	B1 İzmir : Çatalkaya near Fire Tower 1020 m A. Gönüz 20.06.1993

A.K.Ü.M. stands for the herbarium sheets at the Faculty of Science and Art of Afyon Kocatepe University.

Morphological measurements were made on the aboveground parts of the samples. Samples were fixed in 70 % alcohol for anatomical studies. Sartur (9) and Sudan III (10) reactivities were applied to the sections for a better understanding of some anatomical structures. The length and width of the stomata was measured with an acular micrometer using surface sections from the upper and lower parts of the leaf epidermis. The stomatal index was calculated according to the method described by Meidner and Mansfield (1968) (11) by determining the number of stomata and epidermal cells in a unit area.

$$SI = \frac{\text{Number of stomata in a unit area} \times 100}{\text{Number of stomata in a unit area} + \text{Number of epidermis cells in a unit area}}$$

Statistical analysis of the results with regard to morphology, anatomy and stomatal studies was carried out at the Computer Research Centre of Ege University.

The above and underground parts of the plant samples; collected during the flowering stage; were brought to the laboratory, cleaned, dried in an incubator at 80 °C for 24 hours, ground and prepared for analysis.

The amounts of total phosphorus and total potassium were determined with a Spectrophotometer at a wavelength of 436 nm.wavelength and an Eppendorf Flame Photometer, after wet digestion, total nitrogen was determined by the Kjeldahl method (12).

Soil samples of about 1kg taken from depths of 0-20 cm were brought to the laboratory, air-dried and passed through a 2 mm sieve. Soils textures were determined with Bouyoucos the Hydrometer Method", pH with a "Beckman pHmeter", CaCO₃ % with a Scheibler Calcimeter and total % salt content by Conductivity Bridge apparatus according to the methods outlined in detail by Öztürk et al (1987) (13). Organic matter content was determined according to the method outlined by Steubing (1965) (12).

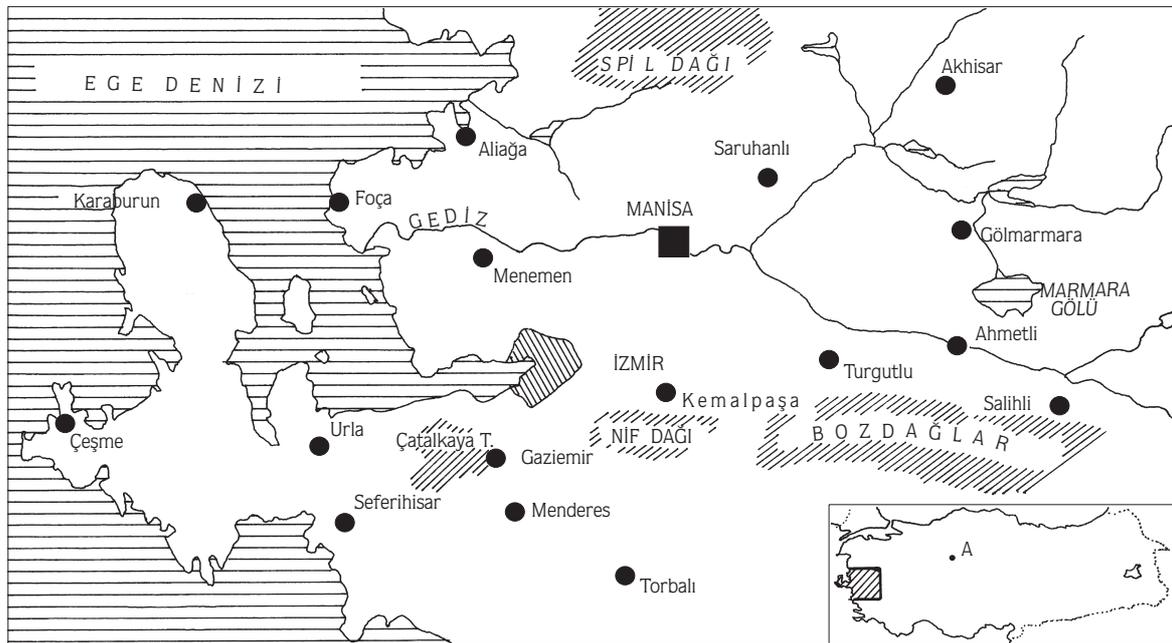
N, P, K in the samples were determined after wetdigestion, according to the Kjeldahl method and methods described by Loue (14), Bingham (15) and Pizer (16) respectively. Results were evaluated according to Öztürk et. al. (13).

The aboveground parts of the plant samples (Herba) were dried and the volatile oil content determined by steam distillation (2).

Results

Phenology

O. onites generally grows in rocky places in the cracks of rocks, in forests, mainly among pine trees. It completes its vegetative growth during the months of April and May and starts to blossom in mid May. Blooming continues until mid July. In July it starts to

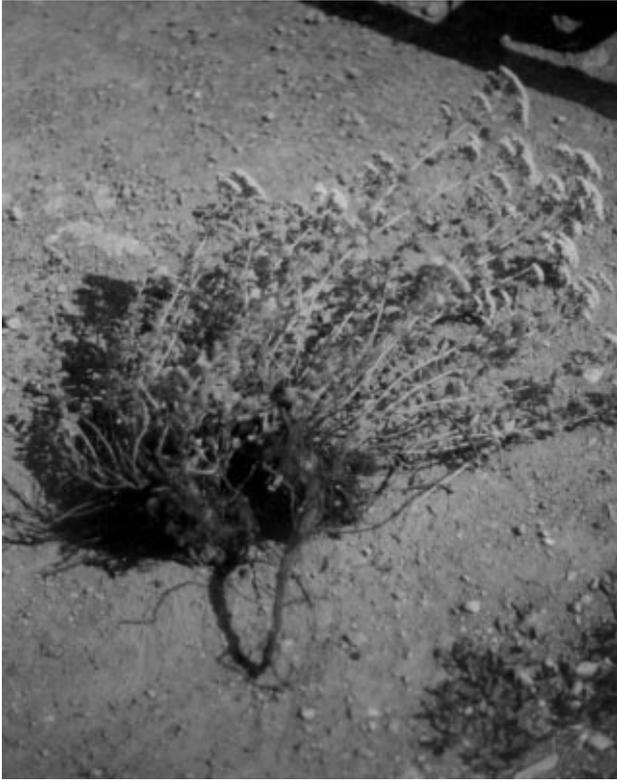
Figure 1. Map showing the collection sites of *O. onites* samples.

	Lower Altitudes	High Altitudes
1- Growth of Plant	–	–
2- First Budding	April	End of April - May
3- First Flowering	May	May - June
4- First Fruiting	July	July - August
5- Maturation of Fruits	July - August	August - September
6- Dispersal of Fruits	August - September	September
7- Leaf Fall	September	September - October
8- Plant Dies	October - November	November

Table 1. Phenological observations on *O. onites* at various altitudesTable 2. Morphological measurements on *O. onites*

Locality	Altitude (m)	Number of measured plants	Mean Stem length (cm)	Mean length and width of lower stem leaves (cm)	Mean Corimbos number	Mean length and width of bract (mm)	Mean sepal length and width (mm)	Mean petal length (mm)
Manisa	180	10	*38.8 ± 1.25	2.14 ± 0.06	21.0 ± 1.63	*4.05 ± 2.50	*2.64 ± 0.09	4.59 ± 0.08
Spil mountain	"	"	"	*2.11 ± 0.04	"	2.50 ± 0.00	1.62 ± 0.07	"
"	340	"	37.3 ± 0.94	2.08 ± 0.07	*15.6 ± 6.48	4.16 ± 0.05	2.65 ± 0.05	4.76 ± 0.09
"	"	"	"	1.83 ± 0.04	"	2.29 ± 0.23	*1.60 ± 0.00	"
"	700	"	37.5 ± 1.43	*1.99 ± 0.12	*14.0 ± 2.16	4.36 ± 0.09	2.86 ± 0.10	4.80 ± 0.08
"	"	"	"	*1.75 ± 0.07	"	2.00 ± 0.00	1.80 ± 0.00	"
Kemalpa_a Nif mountain	300	"	38.5 ± 1.08	2.22 ± 0.06	34.0 ± 1.70	4.15 ± 0.12	2.95 ± 0.05	4.77 ± 0.08
"	"	"	"	1.82 ± 0.06	"	2.50 ± 0.00	2.00 ± 0.00	"
"	610	"	38.0 ± 1.05	2.18 ± 0.04	33.8 ± 2.86	4.05 ± 0.05	*2.83 ± 0.10	4.72 ± 0.09
"	"	"	"	1.78 ± 0.04	"	2.10 ± 0.00	1.92 ± 0.07	"
"	900	"	40.1 ± 1.10	2.37 ± 0.04	28.6 ± 3.23	4.80 ± 0.05	3.05 ± 0.05	*4.82 ± 0.07
"	"	"	"	2.26 ± 0.05	"	2.80 ± 0.00	2.00 ± 0.00	"
Salihli Bozdağ	400	"	49.8 ± 1.23	*2.43 ± 0.08	24.1 ± 4.20	4.05 ± 0.08	2.98 ± 0.07	4.31 ± 0.21
"	"	"	"	2.28 ± 0.04	"	2.10 ± 0.00	2.00 ± 0.00	"
"	900	"	*50.0 ± 1.56	*2.47 ± 0.04	14.3 ± 2.58	4.85 ± 0.05	*3.32 ± 0.10	5.01 ± 0.09
"	"	"	"	2.30 ± 0.04	"	*3.50 ± 0.05	2.70 ± 0.00	"
Yzmir Çatalkaya	270	"	47.9 ± 1.10	2.15 ± 0.17	*37.2 ± 4.76	4.05 ± 0.05	2.73 ± 0.09	*2.72 ± 0.07
"	"	"	"	1.75 ± 0.19	"	2.50 ± 0.00	1.76 ± 0.05	"
"	1020	"	*45.5 ± 0.97	*1.93 ± 0.10	34.6 ± 1.26	*4.59 ± 0.05	2.81 ± 0.07	4.95 ± 0.05
"	"	"	"	*1.65 ± 0.07	"	2.50 ± 0.00	1.76 ± 0.08	"

*The difference between measured values was found to be significant at p<0.05 level.

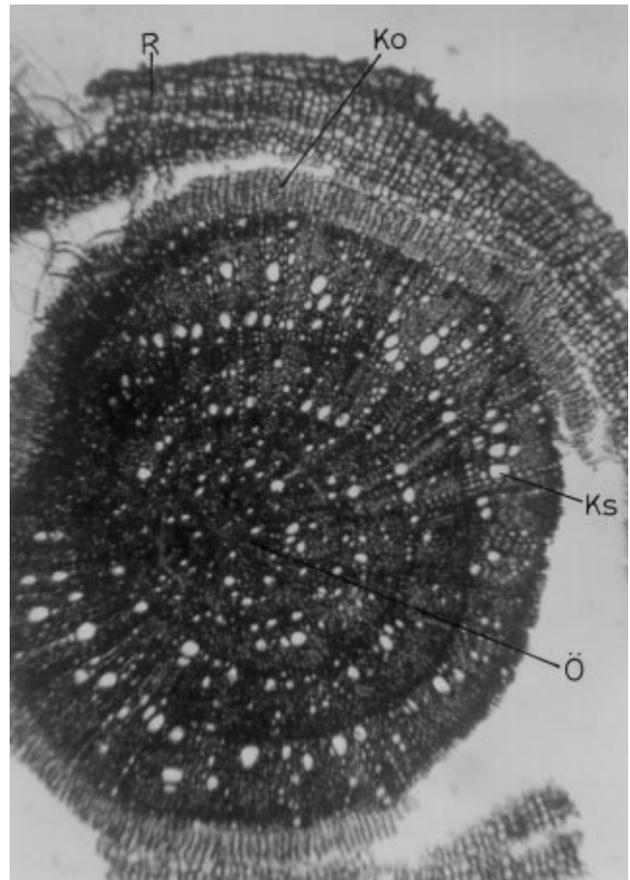


Figures 2a-b. General appearance of *O. onites* in its natural habitat.

produce fruits and about the end of August seeds mature. Fruit production continues until the end of September (Table 1).

Morphology

In the samples of *O. onites* collected from various altitudes (Figures 2 a, b), stem length, stem-leaf length and width, number of flowers, bract length and width, sepal length and width and petal length were measured. The results are given in table 2. It was observed that the length of samples growing at higher sites on Spil and Çatalkaya were shorter. However, no distinctive differences were noticed amongst the Bozdağ samples. In general, stems were longer in the high altitude samples from Nif. Similarly, it was observed that leaves at the lower parts of the stem were shorter at higher sites but no difference existed in the width of stem.

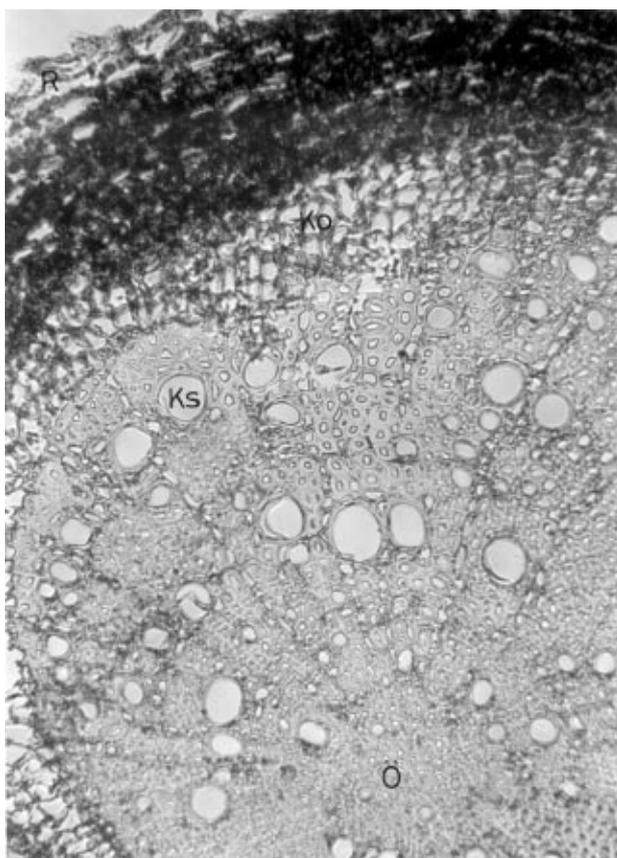


Figures 3. Cross-sections of the *O. onites* roots from different altitudes (Bozdağ 900 m - Çatalkaya 1020 m). R= Rhizoderm, Ks= Xylem, Ko= Cortex, Ö= Pith (3.2 x 6.3).

Table 3. Thicknesses of cortex and xylem in the roots of *O. onites*.

Locality	Altitude (m)	Mean Thickness of the Cortex (μ)	Mean Thickness of Xylem (μ)
Manisa Spil mountain	180	*173.96 \pm 7.33	1035.0 \pm 12.2
"	340	178.68 \pm 3.87	1039.5 \pm 10.1
"	700	*194.40 \pm 7.18	1098.0 \pm 9.50
Kemalpaşa Nif mountain	300	*176.58 \pm 3.54	*1095.0 \pm 17.3
"	610	194.40 \pm 8.72	1114.5 \pm 17.4
"	900	*203.83 \pm 2.97	*1194.0 \pm 7.70
Salihli Bozdağ	400	177.63 \pm 4.59	1096.5 \pm 11.1
"	900	*209.07 \pm 3.87	*1191.0 \pm 10.5
İzmir Çatalkaya	270	*116.85 \pm 4.97	*1080.0 \pm 15.8
"	1020	133.09 \pm 5.06	1156.5 \pm 11.1

*The difference between measured values was found to be significant at $p < 0.05$ level.



Figures 4. Cross sections of the *O. onites* roots from different altitudes (Bozdağ 900 m - Çatalkaya 1020 m). R= Rhizoderm, Ks= Xylem, Ko= Cortex, Ö= Pith (3.2 x 6.3).

It was noticed that samples from higher altitudes had larger but few flowers, with longer bracts, petals and carpels. In the samples from Nif and Bozdağ the latter were wider. However, no difference was seen amongst the samples from Spil or Çatalkaya.

Anatomical Observations

Anatomical sections taken from the root, stem and leaves of the plants were investigated in detail and the results obtained are outlined below.

Anatomy of Root

A torn and lignified rhizoderm layer connected with cortex forms the outermost layer of the roots. Cortex is multi-layered and suberized. Under the cortex layer phloem forms a thin layer followed by xylem which covers a large area and the parenchymatic pith occupies a narrow space (Figures 3, 4). The results of the measurements of the root cortex and xylem layers are given in table 3.

It was observed that there was a statistically significant increase in the cortex and xylem tissues of high altitude samples.

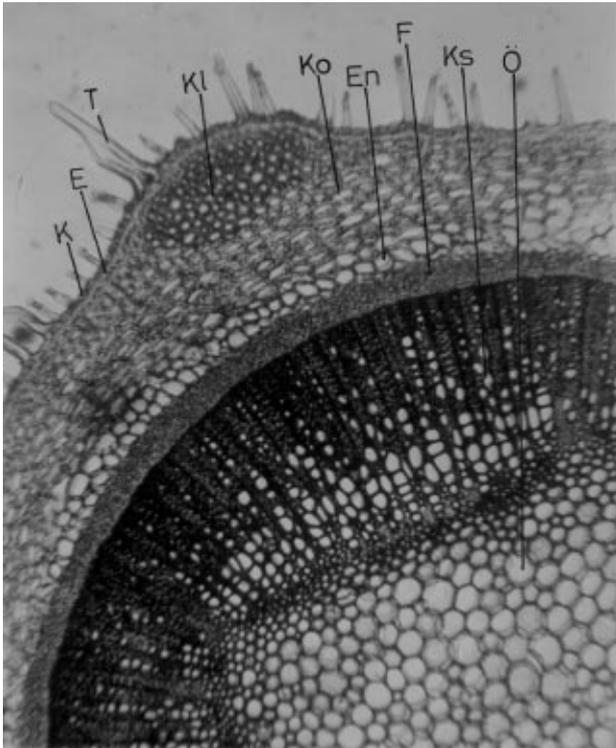
Anatomy of Stem

The stem is quadrangular. In the upper part there is a cuticular layer followed by a single thin layer of epidermis which embodies many papillose and villose hairs. Below the epidermis is collenchyma tissue; which is multi-layered at the four edges of the stem, but is reduced to 2-layers in the parts between the corners. Cortex and a

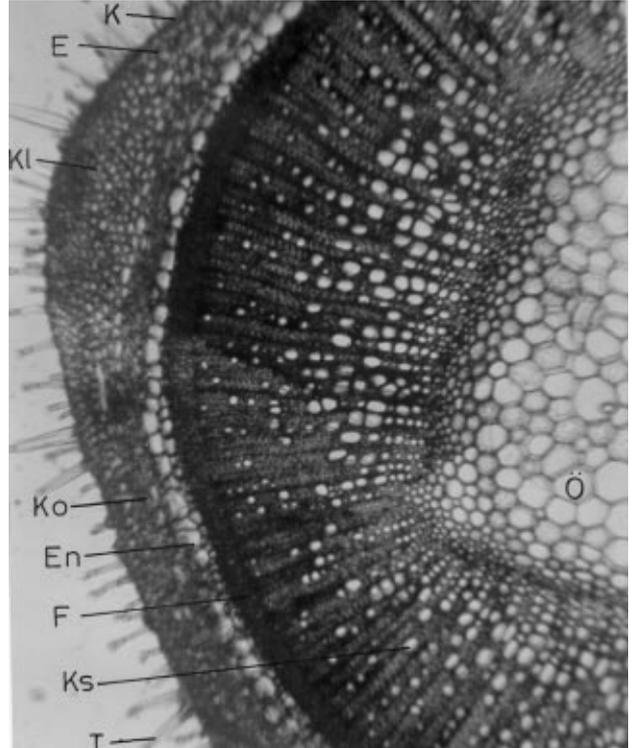
Table 4. Measurements taken from the stem cross sections of *O. onites*

Locality	Altitude (m)	Mean culticle thickness (μ)	Mean collenchyma thickness at four angles (μ)	Mean phloem thickness (μ)	Mean xylem thickness (μ)	Mean parenchymatic pith thickness (μ)
Manisa	180	*5.18 \pm 0.29	*255 \pm 10.0	*49.20 \pm 2.70	559.5 \pm 7.25	979.50 \pm 10.1
Spil mountain	340	5.34 \pm 0.33	261 \pm 14.4	57.12 \pm 2.97	579.0 \pm 7.77	*961.50 \pm 11.0
"	700	*7.75 \pm 0.33	*384 \pm 16.1	59.21 \pm 3.53	583.0 \pm 9.56	980.00 \pm 10.0
Kemalpaşa	300	*5.34 \pm 0.48	301 \pm 19.3	*73.38 \pm 3.31	*505.5 \pm 7.25	*1368.0 \pm 12.6
Nif mountain	610	5.55 \pm 0.56	*279 \pm 25.7	61.31 \pm 4.97	532.5 \pm 24.7	1231.5 \pm 22.9
"	900	*10.0 \pm 0.38	*489 \pm 31.0	61.31 \pm 4.97	561.0 \pm 21.4	*1933.5 \pm 8.50
Salihli Bozdağ	400	*5.39 \pm 0.35	316 \pm 8.50	45.59 \pm 3.54	*495.6 \pm 10.0	*942.00 \pm 9.49
"	900	6.96 \pm 0.43	342 \pm 24.2	*66.54 \pm 2.53	*607.5 \pm 7.91	1149.0 \pm 31.0
Izmir Çatalkaya	270	5.03 \pm 0.36	*324 \pm 22.6	52.40 \pm 5.52	*484.5 \pm 14.2	*928.50 \pm 18.0
"	1020	*10.0 \pm 0.38	*496 \pm 14.5	58.16 \pm 5.21	592.5 \pm 12.7	1132.5 \pm 7.91

*The difference between measured values was found to be significant at $p < 0.05$ level.



Figures 5. Cross sections of the *O. onites* roots from different altitudes (Bozdağ 900 m - Çatalkaya 1020 m). R= Rhizoderm, Ks= Xylem, Ko= Cortex, Ö= Pith (3.2 x 6.3).



Figures 6. Cross sections of the *O. onites* roots from different altitudes (Bozdağ 900 m - Çatalkaya 1020 m). R= Rhizoderm, Ks= Xylem, Ko= Cortex, Ö= Pith (3.2 x 6.3).

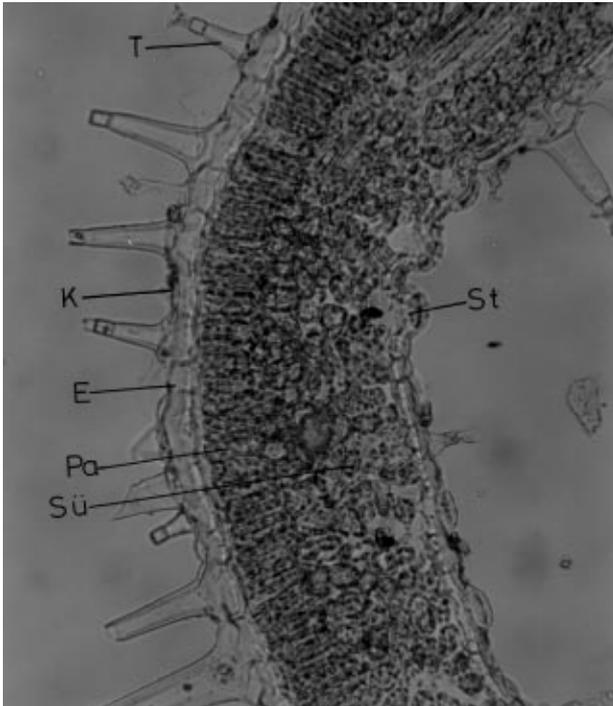
single layer of endodermis are found below the collenchyma. Phloem covers the major part and xylem below it reaches the pith. There are pith rays extending up to the pith from the cambium in the xylem tissue (Figures 5, 6).

The parenchymatous pith cells contain starch which reacts with Lugol and Sartur reactives. No druses or raphides were noticed in the cortex cells. The plant displays secondary growth and the stem anatomy is similar to that of the root.

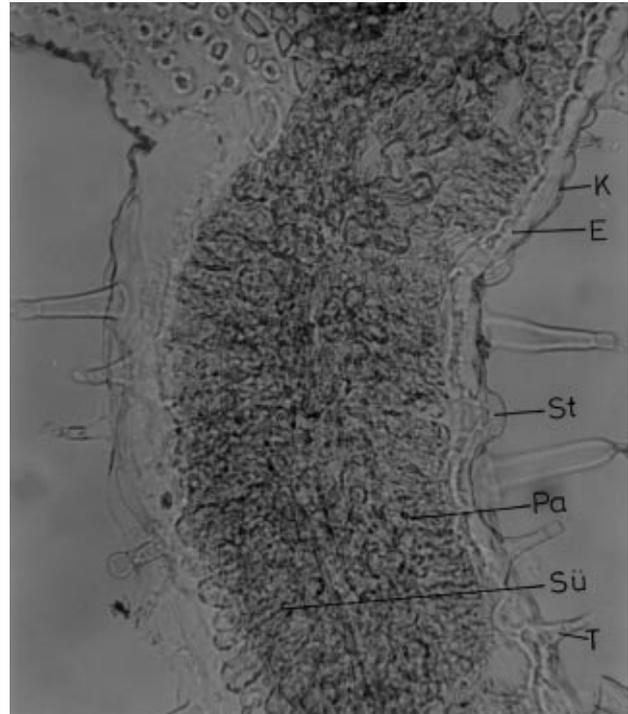
Locality	Altitude (m)	Mean total leaf thickness (μ)	Mean cuticle thickness (μ)
Manisa	180	*195.0 \pm 10.0	*4.56 \pm 0.25
Spil mountain	340	204.0 \pm 7.75	4.88 \pm 0.41
"	700	*225.0 \pm 12.2	*6.73 \pm 0.12
Kemalpaşa	300	228.0 \pm 17.0	4.56 \pm 0.35
Nif mountain	610	246.0 \pm 16.1	4.85 \pm 0.33
"	900	*255.0 \pm 15.8	5.76 \pm 0.55
Salihli Bozdağ	400	*166.5 \pm 11.1	*3.80 \pm 0.11
"	900	246.0 \pm 10.5	*5.61 \pm 0.16
İzmir Çatalkaya	270	*202.5 \pm 7.91	*3.51 \pm 0.18
"	1020	*247.5 \pm 7.91	*5.76 \pm 0.55

Table 5. Leaf and cuticular thicknesses of *O. onites* from different altitudes

*The difference between measured values was found to be significant at $p < 0.05$ level.



Figures 7. Cross sections of the *O. onites* leaves from different altitudes (Spil mountain 180 m -Çatalkaya 1020 m). K= Cuticle, E=Epidermis, KI=Collenchyma, T=Hair, St=Stomata, Pa=Palisade parenchyma, Sü= Spongy parenchyma (10 x 6.3).



Figures 8. Cross sections of the *O. onites* leaves from different altitudes (Spil mountain 180 m -Çatalkaya 1020 m). K= Cuticle, E=Epidermis, KI=Collenchyma, T=Hair, St=Stomata, Pa=Palisade parenchyma, Sü=Spongy parenchyma (10 x 6.3).

Thickness of tissue in the stems was measured micrometrically and the results are given in table 4.

The results clearly show that at higher altitudes the cuticle becomes thicker, collenchyma increases and the xylem, phloem and parenchymatous pith widen.

Anatomy of Leaf

Leaves are bifacial covered by a thick cuticular layer on both upper and lower surfaces and intense papillose and

villose hairs, followed by a single layered epidermis and palisade cells. Palisade cells are rich in chloroplasts. Spongy parenchyma cells are lined-up tightly and occupy a wide area in the mesophyll. There are no broad intercellular spaces in the mesophyll tissue. Stomata are present on both upper and lower epidermis. Vascular bundles are of collateral type. Upper and lower parts of central vessel are surrounded by collenchymatous cells. There are many-layered sclerenchymatous cell bundles

Table 6. Stomatal features in upper epidermal peelings from different altitudes in *O.onites*

Locality	Altitude (m)	Mean Number of Stomata	Mean Number of Epidermal Cells	Stomatal Index	Open /Close (%)	Mean Stomatal Length	Mean Stomatal Width
Manisa	180	7.12 ± 0.16	23.33 ± 0.44	*23.38 ± 1.36	100 Open	*63.89 ± 4.55	*53.95 ± 4.69
Spil mountain	340	6.80 ± 0.70	24.29 ± 0.44	21.87 ± 1.59	"	*66.95 ± 0.00	47.82 ± 0.00
"	700	5.76 ± 0.66	26.22 ± 0.71	18.01 ± 0.92	"	64.66 ± 4.17	*45.53 ± 4.17
Kemalpaşa	300	7.16 ± 0.68	*34.72 ± 0.63	17.09 ± 1.07	"	65.04 ± 3.90	47.82 ± 0.00
Nif mountain	610	6.56 ± 0.65	38.41 ± 0.89	14.58 ± 0.73	"	66.57 ± 1.92	47.44 ± 1.91
"	900	6.36 ± 0.63	*41.17 ± 0.77	*13.38 ± 0.81	"	66.95 ± 0.00	47.82 ± 0.00
Salihli Bozdağ	400	*9.20 ± 0.70	29.14 ± 0.66	23.99 ± 1.06	"	68.49 ± 3.58	49.35 ± 3.57
"	900	*5.52 ± 0.58	32.85 ± 0.59	14.38 ± 0.98	"	*72.69 ± 4.78	47.82 ± 0.00
Yzmir Çatalkaya	270	7.20 ± 0.70	24.34 ± 0.72	22.82 ± 0.97	"	*63.89 ± 4.55	47.82 ± 0.00
"	1020	7.80 ± 0.76	27.07 ± 0.72	22.36 ± 1.05	"	*58.54 ± 3.17	47.82 ± 0.00

*The difference between measured values was found to be significant at p<0.05 level.

Table 7. Stomatal features on the lower epidermis of *O. onites*

Locality	Altitude (m)	Mean Number of Stomata	Mean Number of Epidermal Cells	Stomatal Index	Open-Close (%)	Mean Stomatal Length	Mean Stomatal Width
Manisa	180	12.46 ± 1.35	*43.39 ± 1.48	22.30 ± 0.91	100 Open	*77.15 ± 2.47	56.11 ± 3.36
Spil mountain	340	13.20 ± 0.86	50.17 ± 1.70	20.83 ± 0.50	"	*86.72 ± 1.68	54.20 ± 2.95
"	700	13.46 ± 1.24	*51.90 ± 1.41	20.59 ± 0.87	"	84.17 ± 3.96	54.52 ± 3.96
Kemalpaşa	300	11.00 ± 1.64	*44.31 ± 1.73	19.88 ± 0.94	"	80.34 ± 4.85	57.39 ± 0.00
Nif mountain	610	10.80 ± 2.00	46.42 ± 1.44	18.87 ± 1.38	"	79.07 ± 7.64	*60.58 ± 6.92
"	900	*09.20 ± 1.20	*49.18 ± 1.85	*15.75 ± 0.64	"	81.30 ± 4.96	*49.10 ± 3.37
Salihli Bozdağ	400	13.86 ± 0.91	35.12 ± 1.06	*28.29 ± 0.85	"	*86.08 ± 0.00	*65.68 ± 3.37
"	900	11.20 ± 1.15	37.24 ± 1.42	*23.12 ± 0.80	"	*76.52 ± 0.00	*52.93 ± 4.94
İzmir Çatalkaya	270	*14.26 ± 0.79	*42.13 ± 1.79	25.28 ± 0.44	"	79.59 ± 3.37	*66.95 ± 0.00
"	1020	12.60 ± 1.30	*47.22 ± 1.09	21.06 ± 1.19	"	77.48 ± 2.68	*58.02 ± 2.47

*The difference between measured values was found to be significant at p<0.05 level.

below collenchymatous cells (Figures 7, 8). The thicknesses of different leaf tissues at different altitudes are given in table 5.

Both cuticular as well as total leaf thicknesses increase with increases in altitude. In general, the lower epidermis embodies more stomata and epidermal cells than the upper one. The number of epidermal cells also increases with altitude on both upper and lower surfaces of the leaf (Table 6, 7).

The number of stomata on the upper epidermis of the leaf is lower in high altitude samples such as those from Spil and Bozdağ, but there was no clear difference between the samples from Nif or Çatalkaya (Table 6, Figures 9, 10). It was noted that the number of stomata on the lower epidermis of the leaf is lower in all high altitude samples except those from Spil (Table 7, Figures 11, 12).

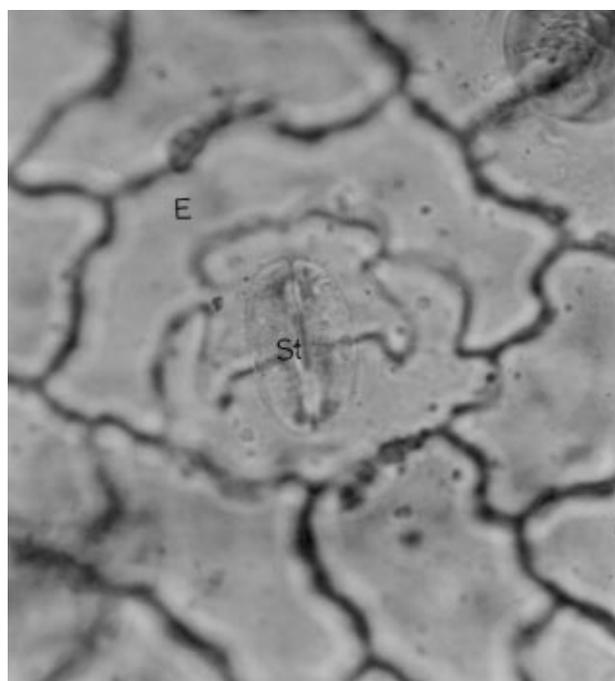
Table 8. Physical analysis of the soils supporting *O. onites*

Locality	Altitude (m) (m)	pH	Sand (%)	Clay (%)	Silt (%)	Texture	Total Salt (%)	CaCO ₃ (%)
Manisa	180	6.94	91.44	5.28	3.28	Sandy	0.30	0.7352
Spil mountain	340	7.02	93.44	5.28	1.28	Sandy	0.42	0.7352
"	700	6.22	83.44	7.28	9.28	Loamy-Sand	0.40	0.6944
Kemalpaşa	300	7.75	85.44	5.28	9.28	Loamy-Sand	0.53	15.889
Nif mountain	610	7.99	55.44	29.3	15.3	Sandy-Clayey-Loam	0.55	21.893
"	900	7.65	75.44	13.3	11.3	Sandy-Loam	0.37	0.0980
Salihli Bozdağ	400	6.39	89.44	7.28	3.28	Sand	N.D.	0.2042
"	900	6.77	93.44	5.28	1.28	Sand	N.D.	0.8162
İzmir Çatalkaya	270	6.03	79.44	9.28	11.2	Loamy-Sand	0.33	0.2451
"	1020	6.54	81.44	7.28	11.3	Loamy-Sand	N.D.	0.6535

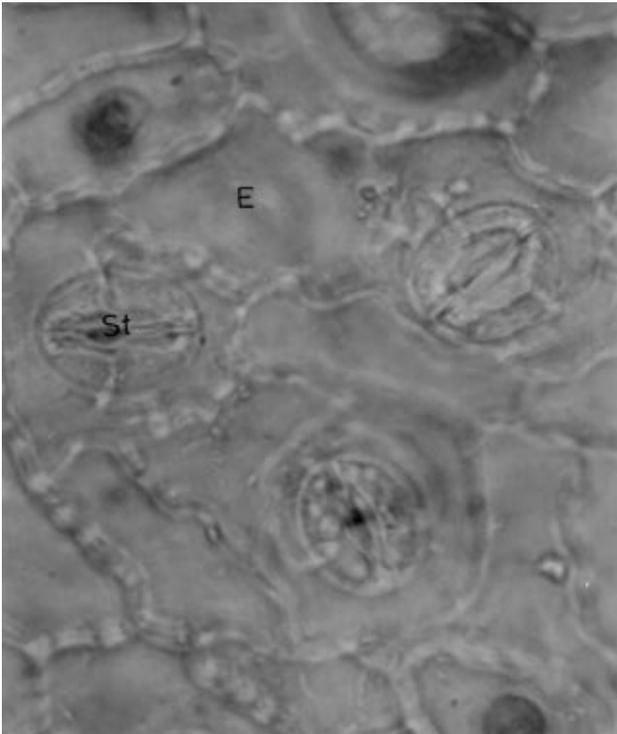
N.D.= Non dedectable.



Figures 9. A view of the stomata from upper epidermal peelings of *O. onites* (Spil mountain 340 m-Bozdağ 900 m). E= Epidermis, St= Stomata (40 x 12.5).



Figures 10. A view of the stomata from upper epidermal peelings of *O. onites* (Spil mountain 340 m-Bozdağ 900 m). E= Epidermis, St= Stomata (40 x 12.5).



Figures 11. A view of the stomata from lower epidermal peelings of *O. onites* (Çatalakaya 270 m-Bozdağ 900 m). E= Epidermis, St= Stomata (40 x 12.5).

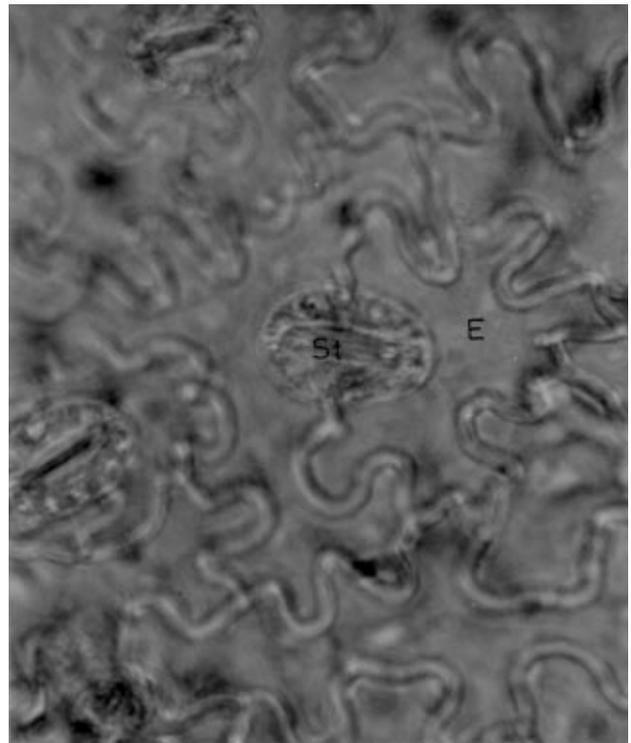
In general, the stomatal indexes in all the high altitude samples are less than those from lower altitudes, on both the upper and lower epidermis except for the samples taken from Çatalakaya. The stomata are longer too in the upper epidermis of high altitude samples from Spil, Nif and Bozdağ, but shorter in those from Çatalakaya, whereas the other ones in the lower epidermis are longer in the high altitude samples from Spil and Nif but shorter in Bozdağ and Çatalakaya samples (Tables 6, 7).

The width of the stomata is less in the upper epidermis of the high altitude samples from Spil and Bozdağ, but no significant difference was observed among the samples from Nif and Çatalakaya. The stomata in general are narrower in the lower epidermis of the high altitude samples from all sites.

These results clearly show that stomata are lower in number on both the lower and upper epidermis of all the high altitude samples and they become narrower but increase in length (Tables 6, 7).

Soil Analysis Data

The physical and chemical analysis of the soil samples taken from the sites where *O. onites* grows are presented in tables 8 and 9.



Figures 12. A view of the stomata from lower epidermal peelings of *O. onites* (Çatalakaya 270 m-Bozdağ 900 m). E= Epidermis, St= Stomata (40 x 12.5).

Table 8 shows that this plant can grow well on sandy clayey loam though it prefers sandy loam. The pH of the soil varies between 6.03 - 7.99 thus it prefers slightly acidic and slightly basic soils. It generally grows on slightly calcareous soils. However, it was also observed at times to grow on extremely calcareous soils like the ones at altitudes of 300-900 m on Nif. The percentage of total salt in the soils is small or moderate, but it grows on non-saline sites mainly.

Soils where *O. onites* occurs are rich in N and P, but poor in K (Table 9). All the samples except that from Bozdağ (900 m) are rich in organic matter.

Plant Analysis Data

The percentages of N, P and K content in the aboveground parts of the plants were analysed during the generative growth period and the results are presented in table 10. According to these results N, P and K contents in all samples exhibit optimal values for plant growth.

Volatile Oil content in the aboveground parts of *O. onites*

The results of volatile oil content of *O. onites* determined during the flowering period are given in table 11.

Locality	Altitude (m)	N (%)	P (%)	K (%)	Organic matter (%)
Manisa	180	0.168	0.0723	0.24	3.801
Spil mountain	340	0.336	0.7230	0.22	4.112
"	700	0.336	0.0723	0.05	3.051
Kemalpaşa	300	0.616	0.0855	0.12	4.939
Nif mountain	610	0.280	0.6570	0.15	5.275
"	900	0.448	0.6570	0.23	3.077
Salihli Bozdağ	400	0.168	0.0921	0.11	2.379
"	900	0.280	0.0921	0.15	1.215
İzmir Çatalkaya	270	0.224	0.0855	0.20	3.723
"	1020	0.280	0.0921	0.24	5.146

Table 9. Chemical analysis of the soils supporting *O. onites*

Locality	Altitude (m)	N (%)	P (%)	K (%)
Manisa	180	0.826	0.1333	0.600
Spil mountain	340	0.588	0.1866	0.850
"	700	0.644	0.1666	0.880
Kemalpaşa	300	0.840	0.1933	0.800
Nif mountain	610	0.336	0.1733	0.350
"	900	0.644	0.1400	0.450
Salihli Bozdağ	400	0.504	0.1800	0.830
"	900	0.700	0.1800	0.790
İzmir Çatalkaya	270	0.602	0.1733	0.650
"	1020	0.854	0.1800	0.800

Table 10. Plant analysis results of *O. onites*

Locality	Altitude (m)	Amount of volatile oil (g / 100 g)
Manisa Spil mountain	180	*2.6 ± 1.02
"	700	3.7 ± 3.44
Kemalpaşa Nif mountain	300	3.4 ± 0.79
"	610	3.9 ± 1.54
"	900	*4.5 ± 0.91
Salihli Bozdağ	400	3.8 ± 0.45
"	900	4.3 ± 1.04
İzmir Çatalkaya	270	3.5 ± 0.16
"	1020	4.1 ± 0.83

Table 11. Volatile oil content of *O. onites* collected from different altitudes

*The difference between measured values was found to be significant at $p < 0.05$ level.

The percentage of volatile oil content varies between 2.6 - 4.5 g / 100 g. The amount of volatile oil content increases with an increase in altitude.

Discussion

The physico-chemical pressures of different environmental parameters are of great importance for the survival of populations under different conditions and

different species react differently to these parameters. In particular the changes in the local environmental conditions play an important role in the expansion and distribution of species as well as their habitats (17). Many environmental factors change due to changes in altitude. For instance, the amounts of precipitation and radiation increase with an increase in altitude together with the effects of wind, daily temperature differences, cloudyness and humidity. On the other hand, evaporation and mean temperature decrease with an altitudinal increase and vegetative phases and the process of pedogenesis shorten (18). All these changes in relation to altitude, affect plant life, in particular the morphological and anatomical characteristics.

A morphological analysis of our plant specimens reveals that the stems of *O. onites* at Spil and Çatalkaya are shorter but no difference exists in Bozdağ samples, but the high altitude samples whereas Nif are longer (Table 2). The samples of *O. onites* with shorter stems are generally of spreading species growing on steep slopes, exposed to the drying effect of the sun and wind and experiencing low water uptake during rainy periods due to the steepness.

Sulkinoja et al. (7) have reported that the stem length of *Betula pubescens* is inhibited with altitude. Our results are in agreement with their findings. The high altitude samples from Nif grow under pine trees close to the trout farm where there is more water and so the stems are longer than the others. The physical characteristics of the soil on Nif mountain differ between high and low altitude sites also.

The high altitude soils are non-calcareous whereas lower ones are extremely calcareous (Texture + Marn) (Table 8).

Lower leaf lengths of *O. onites* are shorter in the high altitude samples from Spil, Nif and Çatalkaya, They are also narrower except in the samples from Nif but no difference exists amongst the Bozdağ samples. In a study by Noitsakis et al. (19), it was reported that the size is reduced in Kermes oak under drought conditions. Similar results were obtained for our species.

The flowers at higher altitudes are fewer in number but larger in size (Table 2), and blossoming periods of *O. onites* start and end later (Table 1) compared with the sites at lower levels. Gibson (20) has stated that there is a strong correlation between the increase in altitude, blossoming period and seed production in the cushion plants, but this has little effect on primary production. In another study by Timoshok et al. (21) it was reported that altitude does not have a significant effect on the seed

production of *Vaccinium myrtillus* L... Our studies support the conclusions of Gibson but differ from those of Timoshok et al.(1990). An increase in the number of flowers clearly indicates that seed production is affected and a decrease in the number of flowers will definitely lead to a decrease in seed production. It emphasizes the idea that ecological factors cause different reactions in different plants.

Anatomical observations show that there is a spatial increase in the root cortex and vascular tissues of the high altitude samples of *O. onites* from all sites (Table 3). In the stem, the cuticle layer grows thicker; collenchyma layers on the corners of the stem increase and also there is a spatial increase in the stem cortex and vascular tissues in all high altitude samples (Table 4). The thicknesses of the cuticle and the whole leaf increase with an increase in altitude at all sites (Table 5).

Jaffe (22) has stated that an increase in the diameter of stems against the effects of wind occurs as a result of the development of small but thick-walled cells. Hunt et al. (23) have observed that in bean plants grown under field conditions an increase in the diameter and a decrease in the internodiums of plants shows a linear correlation as the speed of wind goes up to 4.5 m/h. Retuerto et al. (24) have studied the effect of wind on *Sinapis alba* L. at a speed of 6 m/h for 42 days. They report that little difference is seen during the first days except for an increase in the transpiration rate and stomatal reactions. However after 42 days of continuous wind effect, the ratio of leaf area, root and leaf weights and root/stem ratios increase in comparison with control plants. These findings support our results regarding the diameter increase in both the roots and stems of our samples due to wind at high altitudes. A comparison of the leaf anatomy of 40 species of the genus *Calamagrostis* Adans. from *Poaceae* family has revealed that environmental factors and altitudinal changes result in differences in the number of vascular bundles, epidermal papilla, accumulation of silisium, density of hairs, number and distribution of stomata, arrangement and distribution of sclerenchyma cells and hair lengths (25). An increase in the thickness of leaf and cuticle of our plants in relation to altitude can be ascribed to the variations reported by Escalona (25) too. In our studies we noticed that in general the number of stomata and epidermal cells changes with altitude (Tables 6, 7).

Avissar (26) has noted that in a large potato-producing area in America, there is a wide variation in the stomata between two small homogenous sites where atmospheric conditions vary. He has attributed this situation to the micro-environmental and changing

physiological conditions. Our observations concerning the differences in the number of stomata and epidermal cells in relation to altitude are consistent with those of Avissar (1993).

Since the volatile oil content of *Origanum* species is of great importance for the Turkish economy, the effects of altitudinal variation on this parameter were examined in the samples collected from different altitudes and different sites. Volatile oil content increases in parallel with an increase in altitude (Table 11). The most significant increase was observed in the high altitude samples from Nif. Many workers have investigated this topic. Ceylan (5) stated that the average production of volatile oil varies in samples of *O. smyrnaeum* L. collected during the pre-flowering, flowering and post-flowering stages and grown under Bornova's ecological conditions Vokou et al. (27) emphasized that *O. onites* adapts well to summer drought and the production of volatile oil is high. Avcyata et al. (6) have noted that the distillates of *Origanum* oil known as thyme oil collected from different sites and types vary greatly. Novruzova et al. (28) have reported that the amount of volatile oil in the meso-

xerophytic plants of the *Thymus* L. species is 0.16-0.54 % but this rate goes up to 0.72-0.96 % in xerophytes. Rhizopoulou et al. (29) have stated that volatile oil content increases in *O. majorana* with a decrease in soil moisture.

Although many studies on volatile oil content from various sites have been done none of these deal with the changes in the volatile oil content in relation to altitude. However, relying on the findings of Rhizopoulou et al. (1991) that volatile oil content increases due to a decrease in soil moisture; and of Vokou et al. (1988) that *O. onites* adapts well to summer drought and has high production of volatile oil, we can conclude that volatile oil content increases with altitude in *O. onites* due to similar reasons.

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