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Climate type-related changes in the leaf micromorphological characters of certain landscape plants

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Abstract This research aimed to determine changes to some micromorphological characteristics of certain landscape plants grown in areas with different dominant climate types. Leaf samples were collected from eight woody plant species in areas with terrestrial, Black Sea, and Mediterranean climate types in Turkey. Then, scaled images of the collected leaf samples were obtained using SEM. StomaLength, StomaWidth, PoreLength, PoreWidth, and StomaDensity were determined using measurements made on the images. Data were statistically evaluated, and the changes to these characteristics by climate type and species were determined. The results show that the lowest values were for plants grown in the terrestrial climate, while the highest values were for plants grown in the Mediterranean climate for all characteristics except SDEN. For SDEN, the lowest value was for plants grown in the Mediterranean climate, while the highest value was obtained for plants grown in the terrestrial climate.

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Introduction

Together with rapid development around the world, structural changes in economics, society, culture, and politics have led to the destruction of green areas by accelerating urbanization. This estrangement of people from nature caused by rapid urbanization and industrialization has led people, who are part of nature, to take pieces of nature into their living spaces. Landscape studies have thus become a part of modern life (Cetin 2013, 2015a, b, c, 2016a, b, c, d, e, 2017; Sevik and Cetin 2016; Cetin et al. 2017, 2018).

Unlike other fields, landscape studies foreground the use of plants. Landscape arrangements in outdoor areas are valuable to the extent to which they can go beyond their regular use. In landscaping, people use species that do not exist in the natural flora of a region, which significantly affects the selection of plants in landscape studies (Cetin 2013, 2015a, b, 2016a, b, c, d, e, 2017; Cetin and Sevik 2016a, b, c; Guney et al. 2016, 2017). In landscape studies, plants used outside their natural growth areas usually fail to thrive as they did in their natural growth areas, do not reflect the forms of their natural growth areas, and often face stress factors. The visibility of these stress factors, in addition to the visible morphological features of the plants, also affects micromorphological features (Yigit et al. 2016; Sevik et al. 2017).

Turkey has three main climate types that are quite different from one another. Many plants are grown in areas where all three types of climate prevail in landscape works. These plants generally do not significantly differ morphologically; however, there is not sufficient information on how they differ at a micromorphological level. Changes that occur at a micro level can provide insight on the stress levels, growth, and adaptation levels.

This study aimed to determine changes to micromorphological characteristics in eight woody landscape plants that are grown in areas where different types of Turkish climates prevail.

Materials and methods

This study was conducted on plants collected from areas with the three types of climate in Turkey. The climate types in Turkey are as follows:

(a) The Black Sea climate is rainy in all seasons. The Eastern Black Sea Region receives maximum precipitation in autumn and minimum precipitation in spring. The annual amount of precipitation is 2000–2500 mm. The Western Black Sea Region receives maximum precipitation in autumn and minimum precipitation in spring. The annual amount of precipitation is 1000–1500 mm. The Central Black Sea Region receives maximum precipitation in winter and minimum precipitation in summer. The annual amount of precipitation is 700–1000 mm. The average number of snowy days in areas with the Black Sea climate is 18. The annual average temperature is 13–15 °C. The average temperature in January is 6–7 °C. The average temperature in July is 21–23 °C. The annual temperature difference is 13–15 °C.

(b) In the Mediterranean climate, summers are hot and dry, while winters are mild and rainy. The maximum precipitation is in winter while the minimum precipitation is in summer. The difference between the amounts of precipitation in the summer and winter is quite high. The average annual precipitation ranges from 600 to 1000 mm. The annual average temperature is 18–20 °C. The average in January is 8–10 °C. The average in July is 28–30 °C. The annual temperature difference is 15–18 °C.

(c) The terrestrial climate is hot and dry in summer and cold and snowy in winter. The Central Anatolia Region receives maximum precipitation in spring and minimum precipitation in summer. The average precipitation in the Central Anatolia Region is 300–400 mm. The average winter temperature of Central Anatolia is 1–2 °C, the average summer temperature is 22–23 °C, and the average annual temperature is 10–12 °C. The annual amount of precipitation is 500–600 mm. Figure 1 presents a map of these types of climate in Turkey.

This study was conducted on *Buxus sempervirens* (Authority: Linnaeus, URL1 2018), *Laurocerasus officinalis* (Authority: Linnaeus, URL2 2018), *Fraxinus excelsior* (Authority: Linnaeus, URL3 2018), *Euonymus japonicas* (Authority: Thunberg, URL4 2018), *Prunus cerasifera* (Authority: Ehrhart, URL5 2018), *Aesculus hippocastanum* (Authority: Linnaeus, URL6 2018), *Platanus orientalis* (Authority: Linnaeus, URL7 2018), and *Tilia tomentosa* (Authority: Mönch, URL8 2018) species grown in areas where each of three types of climate prevails. As mentioned in the introduction section of the work, in landscape studies, plants used outside their natural growth areas often fail to thrive as their natural growth areas, do not reflect the form of their natural growth areas, and stress factors. It is aimed to determine how micromorphological characters in species used outside the natural growing area are affected. For this purpose, all types of different climate types were selected. Selected species are different types of leaf structure, tree structure, herbaceous state, and spreading area. In the study, these eight species have various characteristic such as evergreen, deciduous, tall, and shrub. And, some of these species are not native in the study area. Natural distribution of *Euonymus japonicus* is China, Kerean Peninsula and Japan, *Tilia tomemtosa* in Balkans and *Fraxinus excelsior* in northern part of Turkey. Some of the sample individuals are planted artificially. This study is one of the first studies on this subject, and therefore, there is no detailed literature on the study. The reasons for selecting different species are to determine how these species are affected by the conditions of the growing place and to form a base for the next study. If it is possible to determine how the stomatal structure of the individual character is influenced by the climatic condition, it is possible to know how to interpret the results to be obtained in future studies. For example, apart from the natural growing area of *Prunus laurocerasus*, which naturally grows in the black sea region, the stomata are both shrinking and decreasing in number. However, the *Platanus*

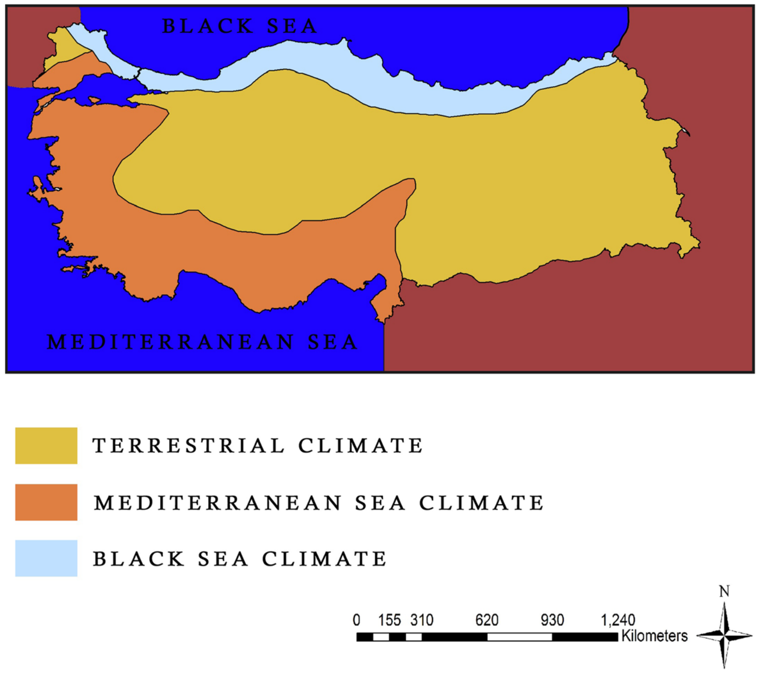

Fig. 1 Map of climate types in Turkey

orientalis grows naturally in the black sea region. Thus, two different species react differently to climate change, possibly related to plant structure. These and similar results can be used in the design of more detailed studies by using them as a basis for further study.

Within the scope of the study, mature leaf samples collected from the above species. Ten leaf samples were collected from each tree at three different regions. Ten tree samples were selected in each species from three different regions. In total, 300 leaf samples were collected from 30 trees. They were selected as sample individuals in the study area; the samples were obtained from individuals used in landscape arrangements in city centers. They are collected from parks away from the traffic in the city center. They were pressed and dried at the end of August 2017 and examined under an electron microscope in the laboratory. Scaled images were obtained from the lower surface and the areas close to the middle parts of the leaf blade using SEM. Files with ".jpeg" extension were created from the images. After completing these processes, the following measurements were made using the "ImageJ" computer measurement program for performing leaf micromorphological measurements:

SDEN Stoma density (in an area of 1 mm^2)
STL Stoma length
STW Stoma width
PORL Pore length
PORW Pore width

Data were assessed using the SPSS package program, and variance analysis and Duncan's test were applied to the data.

Results

Table 1 presents the results of the variance analysis and Duncan's test in determining changes in the micromorphological characteristics of species grown in areas with different types of climate.

Table 1 shows that there are statistically significant differences at a confidence level of at least 95% between the climate types in terms of all characteristics except for PW. Table 1 also shows that the lowest values were obtained for plants grown in the terrestrial climate, while the highest values were obtained for plants grown in the Mediterranean climate, in terms of all characteristics except SDEN. For the SDEN values, the ranking is the exact opposite. The lowest value in terms of SDEN was for plants grown in the Mediterranean climate, which had the highest values for other characteristics, while the highest value was for plants grown in the terrestrial climate, which had the lowest values for other characteristics.

Table 1 Changes in micromorphological characteristics by type of climate

Climate	STL	STW	PL	PW	SDEN
Terrestrial	24.83 a	19.83 a	9.93 a	4.04 a	288.75 c
Mediterranean	27.79 b	20.86 b	12.77 c	4.22 a	202.16 a
Black Sea	23.95 a	18.35 a	11.22 b	4.05 a	250.48 b
F value	9.012***	3.969*	18.070***	0.314 ns	11.707***

*Significant at 0.05 level; ***significant at 0.001 level; *ns* not significant. The letters a, b, c, etc. means, according to Duncan test results, show that the group is located. It is statistically different from the values contained in different groups, starting with the letter a numerical value grows

Eight different species were examined in the study. Table 2 shows the results of the variance analysis and Duncan's test on the presence of differences between the species in terms of micromorphological characteristics.

Table 2 shows that there are statistically significant differences between the species in terms of all characteristics at a confidence level of 99.9%. According to the results of Duncan's test, the lowest values were generally obtained in *Prunus cerasifera* for all characteristics except SDEN. Apart from this, the highest values for STL and STW were obtained in *Laurocerasus officinalis*. These values for STL and STW in *Buxus sempervirens* are lower than *Laurocerasus officinalis*, while the highest values for PL and PW were obtained in *Fraxinus excelsior*. While the lowest values for SDEN were obtained in *Tilia tomentosa*, the highest values for SDEN were obtained in *Fraxinus excelsior* and *Euonymus japonicus*.

Table 3 presents the changes in the micromorphological characteristics based on plant species grown in areas with different prevailing climate types.

Table 3 shows that there are no statistical differences at a confidence level of at least 95% for any micromorphological characteristics except for SDEN in *Fraxinus excelsior*. Furthermore, it was determined that there was no statistical difference at a confidence level of at least 95% for STL and STW in *Euonymus japonicus*, SDEN in *Prunus cerasifera*, STW, PW, and SDEN in *Platanus orientalis*, and PL and PW in *Tilia tomentosa*.

The results show that the lowest values in *Prunus cerasifera*'s characteristics, besides SDEN, were obtained in individuals grown in the Black Sea climate, while the highest values were obtained in individuals grown in the terrestrial climate. The lowest values in *Aesculus hippocastanum* were obtained in the terrestrial climate and the highest values were obtained in the Mediterranean climate. For *Buxus sempervirens*, the lowest values for characteristics besides SDEN and STL were obtained in individuals grown in the Black Sea climate, while the highest values were obtained in individuals grown in the Mediterranean climate. This shows that

Table 2 Change in micromorphological characteristics by plant species

Species	STL	STW	PL	PW	SDEN
Buxus sempervirens	30.37 d	24.34 f	11.41 bc	5.66 d	224.22 b
Laurocerasus officinalis	33.40 e	28.03 g	11.93 cd	2.65 a	212.78 b
Fraxinus excelsior	27.41 c	19.86 d	13.78 e	5.81 d	327.39 c
Euonymus japonicus	25.04 b	20.82 de	10.14 b	4.12 b	459.61 c
Prunus cerasifera	17.12 a	11.00 a	7.81 a	2.76 a	223.06 b
Aesculus hippocastanum	24.29 b	16.23 c	11.06 bc	3.05 a	197.94 b
Platanus orientalis	27.81 c	22.50 ef	13.33 de	4.82 c	212.78 b
Tilia tomentosa	16.94 a	13.27 b	10.65 bc	3.89 b	102.67 a
F value	49.269***	61.601***	13.651***	27.323***	40.783***

***Significant at 0.001 level. The letters a, b, c, etc. means, according to Duncan test results, show that the group is located. It is statistically different from the values contained in different groups, starting with the letter a numerical value grows

Table 3 Changes in micromorphological characteristics by climate type and plant species

Species	Climate	STL	STW	PL	PW	SDEN
Buxus sempervirens	Terrestrial	27.56 a	25.96 c	11.16 ab	6.63 b	256.83 b
	Mediterranean	33.74 c	24.43 b	13.41 b	6.66 b	150.33 a
	Black Sea	29.81 b	22.62 a	9.67 a	3.70 a	265.50 b
	F value	22.972***	38.973***	4.599*	10.135***	39.006***
Laurocerasus officinalis	Terrestrial	33.94 b	29.75 b	8.20 a	2.51 b	222.50 b
	Mediterranean	31.32 a	23.75 a	15.11 c	1.65 a	179.33 a
	Black Sea	34.94 b	30.58 b	12.48 b	3.80 c	236.50 b
	F value	9.842***	18.067***	17.927***	45.714***	9.536***
Fraxinus excelsior	Terrestrial	25.38 a	19.93 a	12.49 a	5.38 a	425.00 b
	Mediterranean	28.37 a	20.87 a	14.08 a	5.82 a	213.17 a
	Black Sea	28.49 a	18.79 a	14.76 a	6.23 a	344.00 ab
	F Value	2.756 ns	1.923 ns	1.924 ns	0.676 ns	4.992 *
Euonymus japonicus	Terrestrial	24.12 a	20.01 a	8.23 a	2.92 a	593.67 c
	Mediterranean	25.98 a	22.39 a	9.51 a	4.44 b	281.00 a
	Black Sea	25.03 a	20.05 a	12.67 ab	5.00 b	504.17 b
	F value	2.788 ns	2.715 ns	6.594**	8.509**	31.313***
Prunus cerasifera	Terrestrial	21.47 b	14.76 c	9.40 b	3.65 b	187.83 a
	Mediterranean	17.83 b	11.21 b	7.99 ab	2.19 a	244.83 a
	Black Sea	12.05 a	7.03 a	6.04 a	2.43 a	236.50 a
	F value	10.031***	9.586***	3.563*	7.056**	2.115 ns
Aesculus hippocastanum	Terrestrial	16.55 a	9.54 a	6.52 a	2.10 a	298.50 c
	Mediterranean	37.28 b	27.02 b	14.88 c	3.70 b	177.17 b
	Black Sea	19.03 a	11.30 a	11.77 b	3.35 b	118.17 a
	F value	61.371***	106.404***	64.428***	7.686**	235.059***
Platanus orientalis	Terrestrial	31.42 b	24.96 a	12.79 a	4.88 a	228.83 a
	Mediterranean	26.72 a	22.06 a	15.15 b	5.49 a	182.17 a
	Black Sea	25.29 a	20.49 a	12.04 a	4.08 a	227.33 a
	F value	4.406*	2.089 ns	6.416**	2.932 ns	1664 ns
Tilia tomentosa	Terrestrial	18.21 b	13.68 b	10.65 ab	4.29 b	96.83 a
	Mediterranean	14.36 a	9.36 a	11.26 b	3.38 a	176.33 b
	Black Sea	16.97 b	14.81 b	10.35 a	3.75 ab	71.67 a
	F value	11.702***	19.538***	2.107 ns	3.061 ns	31.257***

*Significant at 0.05 level; **significant at 0.01 level; ***significant at 0.001 level; *ns* not significant. The letters a, b, c, etc. means, according to Duncan test results, show that the group is located. It is statistically different from the values contained in different groups, starting with the letter a numerical value grows

species react differently to different types of climate. A similar situation was observed for SDEN. The lowest values for SDEN were obtained in *Buxus sempervirens*, *Laurocerasus officinalis*, *Fraxinus excelsior*, and *Euonymus japonicus* grown in areas with the Mediterranean climate, and in *Aesculus hippocastanum* and *Tilia tomentosa* grown in the Black Sea climate. The highest values were obtained in *Buxus sempervirens* and *Laurocerasus officinalis* grown in Black Sea climate, in *Euonymus japonicus* and *Aesculus hippocastanum* grown in the terrestrial climate, and in *Tilia tomentosa* grown in the Mediterranean climate. These results show that the highest and lowest values for all morphological characteristics were obtained in individuals grown in different climate types, depending on the species.

Discussion

The results show that the lowest values for all characteristics except SDEN were obtained in the terrestrial climate, while the highest values were obtained in plants grown in the Mediterranean climate. The opposite results were obtained for SDEN. The lowest values for SDEN were obtained in plants grown in the Mediterranean climate, while the highest values were obtained in plants grown in the terrestrial climate. The stomas were smaller but higher in number in individuals grown in the terrestrial climate, while the stomas were larger but lower in number in individuals grown in the Mediterranean climate. SDEN is regarded as one of the most important indicators of plants' reactions to environmental conditions. Xu and Zhou (2008) reported that stomas control the CO_2 and water vapor input to leaves and that stomas are significantly affected by environmental conditions. Furthermore, the fact that the size and number of stomas are inversely proportional has been indicated by many studies (Hetherington and Woodward 2003; Pearce et al. 2006; Galmés et al. 2007).

Stomatal density is an important ecophysiological parameter in the response of plants to several factors, including drought tolerance (Yang and Wang 2001; Zhang et al. 2006; Liu et al. 2006), light (Sevik et al. 2016a), salt stress (Zhao et al. 2001; Romero-Aranda et al. 2001), hardening (Knecht and Orton 1970), water use efficiency (Ferris et al. 1996), and stomatal conductance (Pearson et al. 1995). Thus, stomatal density can be affected by several environmental variables, such as light, shade, humidity, CO_2, and drought (Beerling et al. 1997; Banon et al. 2004).

It is interesting that stoma density is especially correlated with water stress. In this study, the highest values for SDEN were obtained in the terrestrial climate. While the average annual amount of precipitation in Turkish terrestrial climates is 500–600 mm, it is 600–1000 mm in the Mediterranean climate, and more than 700 mm in areas with the Black Sea climate. Therefore, the number of stomas is related to the amount of precipitation, and, accordingly, water stress. Similar results have also been obtained in studies conducted on different species. Dunlap and Stettler (2001) studied *Populus trichocarpa* and determined that stomas are denser in individuals grown in dry areas. In his study on *Populus alba*, Pearce et al. (2006) achieved similar results. Several other studies yielded similar results (Bosabalidis and Kofidis 2002; Guerfel et al. 2009). Also, isohydric and anisohydric behaviors have been studied in terms of underlying physiological mechanisms (Tardieu and Simmonneau 1998; Schultz 2003).

The results show that there are significant differences between plants in terms of all characteristics. However, it is interesting that each plant reacts differently to the climate conditions in which it is grown. In *Prunus cerasifera*, the lowest values for characteristics besides SDEN were obtained in individuals grown in the Black Sea climate, while the highest values were obtained in individuals grown in the terrestrial climate. The lowest values in *Aesculus hippocastanum* were obtained in the terrestrial climate, while the highest values were obtained in individuals grown in the Mediterranean climate. In *Buxus sempervirens*, the lowest values for characteristics besides SDEN and STL were obtained in individuals grown in the Black Sea climate, while the highest values were obtained in individuals grown in the Mediterranean climate. In terms of SDEN characteristics, the lowest values in most species were obtained in individuals grown in the Mediterranean climate, although for *Aesculus hippocastanum* and *Tilia tomentosa*, the lowest values were obtained in individuals grown in the Black Sea climate. The highest values for *Buxus sempervirens*, *Laurocerasus officinalis*, and *Tilia tomentosa* were obtained in individuals grown in the Black Sea climate, for *Euonymus japonicus* and *Aesculus hippocastanum* in the terrestrial climate, and for *Tilia tomentosa* in the Mediterranean climate. According to these results, the genetic structure of the species is one of the most effective factors for micromorphological characteristics. Phenotypic characteristics generally occur as a result of the interaction between genetics and the environment (Sevik et al. 2012) and are shaped by the effects of genetic factors (Sevik 2012), in addition to many environmental factors (Kaya et al. 2009; Sevik and Cetin 2015; Sevik et al. 2016b). This also explains how different species have different reactions to the same climate conditions. Previous studies have also shown that micromorphological characteristics vary significantly by species (Galmés et al. 2007; Maiti et al. 2016).

Conclusion

This study has determined that plant micromorphological characteristics vary significantly by species and that micromorphological characteristics vary significantly in individuals grown in different climate types. However, while this differentiation varies significantly from one species to the other, it was found that the terrestrial climate has an evident effect on SDEN. This is likely related to water stress. According to these results, SDEN characteristics have significant potential for determining the stress levels of plants. In contrast with other methods, SDEN is easily determined and, most importantly, quickly yields results. Therefore, there is a need for detailed studies to clearly show the relationship of this characteristic with plant stress levels. Furthermore, plant micromorphological characteristics can be used in many areas, from genetic variation to species diagnosis, plant stress level, and determining adaptation ability. Therefore, there is a need for studies to treat this topic in a diversity of fields.

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Compliance with ethical standards

Conflict of interest The authors declare no competing financial, professional, or personal interests from other parties.

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