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Sclerophylly in *Fraxinus angustifolia* Vahl. subsp. *oxycarpa* (Bieb. ex Willd.) Franco & Rocha Afonso and *Laurus nobilis* L. and Edaphic Relations of These Species

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Abstract: Sclerophylly and foliar nutrient status and interactions between these factors in ash (*Fraxinus angustifolia* Vahl. subsp. *oxycarpa* (Bieb. ex Willd.) Franco & Rocha Afonso) and laurel (*Laurus nobilis* L.) species were examined. In addition, correlations between sclerophyll index and leaf and soil parameters were investigated. It was found that there were statistically significant differences between the mid-growing season and the end of the growing season with respect to leaf N % concentration, N/Ca and N/Mg ratios in ash, and with respect to leaf N % concentration and soil K % concentration in laurel. The sclerophyll index was negatively correlated with leaf P % concentration in both species. Some important correlations were also found between leaf and soil nutrient concentrations.

Key Words: Sclerophyll index, evergreenness and deciduousness, foliar nutrient status.

Fraxinus angustifolia Vahl. subsp. *oxycarpa* (Bieb. ex Willd.) Franco&Rocha Afonso ve *Laurus nobilis* L.'de Sklerofil ve Bu Türlerin Toprak Faktörleriyle İlişkisi

Özet: Dişbudak (*Fraxinus angustifolia* Vahl. subsp. *oxycarpa* (Bieb. ex Willd.) Franco & Rocha Afonso) ve defne'de (*Laurus nobilis* L.) sklerofil ve yaprak besin durumu ve bu faktörler arasındaki ilişkiler incelenmiştir. Buna ek olarak sklerofil indeksi ve yaprak ve toprak parametreleri arasındaki ilişkiler araştırılmıştır. Dişbudak'ta yaprak N % konsantrasyonu, N/Ca ve N/Mg oranları, defnede yaprak N % konsantrasyonu ve toprak K % konsantrasyonu yönünden gelişme mevsiminin ortası ve senesens dönemi arasında istatistiksel yönden önemli farklılık bulunmuştur. Her iki türde de sklerofil indeksi yaprak P % konsantrasyonu ile negatif olarak ilişkilidir. Yaprak ve topraktaki besin elementleri arasında da bazı önemli korelasyonlar bulunmuştur.

Anahtar Sözcükler: Sklerofil indeksi, Herdem yeşil ve yaprak dökən, yaprak besin durumu.

Introduction

The term "sclerophylly" concept refers to the crude fiber contents and hardness in a leaf (1, 2). There is a wide range of species with different degrees of sclerophylly dependent on leaf texture and mineral composition, especially phosphorus availability (3, 4). Loveless (1, 4) suggested that a decrease in phosphorus content below 0.3 % results in an increase in the degree of sclerophylly.

Evergreen and deciduous species represent different adaptive modes, and they are different from each other with respect to mineral composition and sclerophylly (5, 6).

Old leaves of evergreen species play an important role as storage organs. Deciduous species reduce dry season stress through the leaf mortality and through the

elimination of most of the transpiring surface. Additionally, the physiological capacities of deciduous species are limited by low temperatures or drought during the growing season (7). However, evergreen species have lower photosynthetic rates and tend to have higher investment in antiherbivore compounds than deciduous plants (8).

The differences between evergreen and deciduous species in the degree of sclerophylly and the role of foliar chemistry on sclerophylly have long been a topic of discussion for several researchers (3, 6, 7).

In this study N, P, K, Ca and Mg (%) concentrations, sclerophylly index and correlation between these factors were investigated in *Fraxinus angustifolia* Vahl. subsp. *oxycarpa* (Bieb. ex Willd) Franco & Rocha Afonso (deciduous) and *Laurus nobilis* L. (evergreen). N/P, N/K,

N/Ca and N/Mg values were also calculated because these ratios are reported to be species-specific (8, 9). The relationship between the sclerophyll index and each of certain soil variables has been investigated. In addition, correlation, have coefficients, for leaf-soil nutrient concentrations were calculated.

Materials and Methods

Study site and collection of leaf samples

This study was carried out in the Central Black Sea Region around Gelemen (Hacı Osman Forest), Kurupelit (Çakırlar Forest) and Balık Gölü (Galeric Forest), where these species are very widespread. All of these forests have been defined as unique and endangered alluvial ecosystems on a world-wide basis. Such forests, called "flooded forests", constitute the climax phase of the hydrosere, and are under protection all over the world. Hacı Osman and Galeric Forests are included in the 'List of wetlands of international importance', according to the Ramsar Convention (10).

The mean annual precipitation in the study area is 936.9 mm, and the mean annual temperature is 15.1°C. The study area is situated midway between oceanic and Mediterranean climates (11).

Seven 0.05 ha (500 m²) plots were sampled and the plots were established 100 m from each other. At least five individual plants were used in each plot for each species. The trees were randomly chosen. Each selected plot had a closed tree canopy and generally mature individuals were used. Deciduous and evergreen community types appear to be quite distinct in the area, and both species occurred in each plot.

Since sun and shade leaves may differ in foliar nutrient concentrations, only outer sun leaves were collected (12), and upper crown samples at the four cardinal points were taken with an extension tree-pruner. The sampling procedure was repeated twice in order to determine the changes in nutrient concentrations at the end of the growing season. Ash specimens were sampled both in the mid-growing season (July 1996) and at the end of the growing season (mid-November 1996). Similarly, mid-growing season and end-of-growing-season leaves of laurel were sampled in July 1996 and at the beginning of May 1996, respectively. During both growth periods, leaves of the same age were selected.

Healthy mature leaf blades were cut off above the petiole and placed immediately in tightly stoppered, previously weighed bottles. To avoid incorporating large quantities of non-photosynthetic tissue in ash trees, leaf

samples were removed from between large veins (13). The five samples from each individual tree, (about 100 g) from each plot were dried at 70°C, then powdered in a hammer mill and ground in a Wiley mill to pass a 20-mesh sieve. At least five individual tree samples for both species in each plot were selected during the mid-growing season and at the end of the growing season.

Methods of Chemical Analysis

Sieved leaf samples were digested in a mixture of nitric and perchloric acids, with the exception of samples for N % analysis, which were digested with sulphuric acid and selenium with a Kjeldahl apparatus.

P % concentrations were determined with a Hitachi Model 100-40 spectrophotometer by the ammonium molybdate-stannous chloride method. K %, Ca % and Mg % concentrations were determined with a Perkin Elmer 2280 atomic absorption spectrophotometer (14). Reliability of chemical analysis was evaluated by digesting and analyzing standard pine needle material obtained from the U. S. National Bureau of Standards, and digestions were deemed reliable if they were within 5 % of the standard.

Crude fiber (cellulose and lignin) were determined by the acid-detergent method of Van Soest as cited in Monk (5). The degree of sclerophylly was calculated using the sclerophyll index (15, 16). Sclerophyll index was estimated by the method of Loveless (1) and Specht and Rundel (3) using the ratio of crude fiber dry weight x 100/ crude protein weight (N % x 6.25). Any estimate of sclerophylly based on fiber should therefore be expressed in terms of some measure of foliar tissue. Two obvious possibilities present themselves, either to express fiber content on a fresh-weight basis or to express it per unit of leaf protein. Because the moisture content of leaves is subject to diurnal variation, estimates of sclerophylly based on fiber fresh weight will vary for a given leaf according to the time of day at which it is sampled. This source of diurnal variation is eliminated if crude protein is used as a measure of leaf protein.

Three separate soil samples were collected at depths of 0-20 cm in each plot. Composites of these three collections were mixed and analysed for physical and chemical properties. Therefore, there was one value for each soil variable in each plot. Soil samples were air-dried and then passed through a 2 mm sieve. Soil textures were determined by the Bouyoucus hydrometer method. Soil pH, CaCO₃ (%) and total salinity (%) were determined with a Beckman pH meter, Scheibler

calcimeter and conductivity bridge apparatus respectively. Organic matter (%) and N (%) were determined by the Walkley-Black and micro-Kjeldahl methods respectively. P (%) was determined by using the ammonium molybdate-stannous chloride method following the extraction by ammonium fluoride in hydrochloric acid. K (%) and Ca (%) analysis were done with a Perkin Elmer 2280 atomic absorption spectrophotometer following the extraction by ammonium acetate extract. Mg (%) was also determined by atomic absorption spectrophotometer in sodium chloride extract (17).

The results were evaluated by a 2-way ANOVA test. Means and standard errors were calculated using at least five replicates. Seasons and individual plants were used as variables in the 2-way ANOVA test. Statistical analysis were performed with a MINITAB software package (18).

Results

The soil analysis results are shown in Table 1. *Fraxinus* L. and *Laurus* L. usually prefer sandy clay loamy soils. pH ranges are between 6.98 ± 0.20 , 7.34 ± 0.09 in *Fraxinus* and *Laurus* respectively. Both species grow on non-saline soils. CaCO_3 % concentrations ranged from quite low to the medium level in the study area.

At the end of the growing season, leaves have higher total fiber and lignin in *Fraxinus* and *Laurus* (Table 2). The same result was obtained by Cromack & Monk (19) in the majority of deciduous species and Kutbay & Kılınç (6) in *Quercus cerris* L. var. *cerris* and *Phillyrea latifolia* L. N %, P %, K %, Ca % and Mg % concentrations decreased with age in *Fraxinus* leaves. However, N % and P % concentrations in *Laurus* leaves increase with age, while the other element concentrations are still lower in senescent leaves, but the relative decrease was not as strong as in *Fraxinus* leaves.

Two species exhibited differential behaviours according to season (Table 2). Leaf N % concentration and N/Ca and N/Mg ratios between two different growth periods were significantly different at 0.05 level in *Fraxinus*. Leaf N % concentration and soil K % concentration were significantly different between the

two different growth periods at 0.01 and 0.05 levels respectively in *Laurus*. In addition, there are also important differences between individual plants. Leaf Mg % concentration and soil organic matter % concentration were significantly different at levels of 0.01 and 0.05 respectively in *Fraxinus*. Some important differences were obtained for leaf N/K ratio, soil N% concentration, soil P % concentration and soil organic matter % concentration in *Laurus* (Table 2 and 3; $P < 0.05$, $P < 0.05$, $P < 0.05$ and $P < 0.01$ respectively).

Significant correlations were found between leaf P % concentration and the sclerophyll index for both species ($P < .01$; Table 4). N/Mg ratio was negatively correlated with sclerophyll index in *Fraxinus* ($P < .01$, Table 4). However, the other correlation coefficients were not significant.

Discussion

Loveless (1, 4) has concluded that "in terms of mineral matter a decrease in phosphorus content of leaves below about 0.3 per cent results in a proportional increase in degree of sclerophylly". Loveless (1, 4) has also stated that sclerophyll indices above 100-150 indicate that a vegetation type is P deficient. Our sclerophyll index values varied from 109-173 during mid-growing season and 160-473 during at the end of the growing season in *Fraxinus*. These values are 470-1176 and 339-1842 during mid-growing season and at the end of the growing season respectively in *Laurus*. These values were quite different from the maximum value of 350 reported by Loveless (4). In our previous study (6) sclerophyll index values varied from 217-439 and 148-294 in *Quercus cerris* var. *cerris* and *Phillyrea latifolia* respectively. Specht and Rundel (3) also obtained high sclerophyll index values c. 1200 or even as high 2800 in Mediterranean climate plant communities in Southern Australia. Jayasekera (9) has stated that the presence of various organic structural materials with low nutrient contents within scleromorphic leaves dilutes nutrient contents and yields lower concentrations than with less scleromorphic leaves. The higher sclerophyll index values in the present study than in Loveless's (4) study can also

Species	pH	Total salinity (%)	CaCO_3
<i>F. angustifolia</i> subsp. <i>oxycarpa</i>	6.98 ± 0.20	0.087 ± 0.028	3.65 ± 0.56
<i>L. nobilis</i>	7.34 ± 0.09	0.040 ± 0.015	1.80 ± 0.38

Table 1. Mean values and corresponding standard errors of some physical properties of soil.

Leaf parameter	<i>F. angustifolia</i> subsp. <i>oxycarpa</i>	<i>L. nobilis</i>
N %	(G) 1.60 ± 0.27* (S) 0.58 ± 0.06	0.78 ± 0.29** 0.93 ± 0.21
P %	(G) 0.040 ± 3.58 x 10 ⁻³ (S) 0.023 ± 3.13 x 10 ⁻³	0.017 ± 2.69 x 10 ⁻³ 0.029 ± 8.52 x 10 ⁻³
K %	(G) 1.04 ± 0.27 (S) 0.37 ± 0.06	0.55 ± 0.10 0.44 ± 0.07
Ca %	(G) 3.00 ± 0.21 (S) 2.40 ± 0.27	1.46 ± 0.37 1.34 ± 0.27
Mg %	(G) 0.60 ± 0.16 (S) 0.47 ± 0.14	0.14 ± 0.02 0.12 ± 0.02
N / P	(G) 38.83 ± 4.40 (S) 28.31 ± 5.82	61.84 ± 34.05 27.25 ± 8.11
N / K	(G) 1.84 ± 0.48 (S) 1.92 ± 0.51	1.67 ± 0.56 2.14 ± 0.37
N / Ca	(G) 0.53 ± 0.09* (S) 0.39 ± 0.08	0.68 ± 0.27 0.83 ± 0.24
N / Mg	(G) 2.94 ± 0.47* (S) 1.52 ± 0.23	5.75 ± 2.11 9.71 ± 2.97
Sclerophyll index	(G) 142.9 ± 12.5 (S) 256.5 ± 55.9	681.3 ± 169.7 805.8 ± 231.7

Table 2. Mean values and corresponding standard errors of soil N %, P %, K %, Ca %, Mg % concentrations, N/P, N/K, N/Ca, N/Mg ratios and sclerophyll index. Asterisk indicates that there are statistically significant differences between the two growth periods.

G = Mid-growing season S= End of the growing season. * P < 0.05 ** P < 0.01.

been accounted for in this way. Additionally, the sclerophyll index values of *Laurus* are considerably higher than those of *Fraxinus*. Macroelement concentrations of *Laurus* leaves were somewhat lower than those of *Fraxinus* (Table 2).

There were significant differences between seasons with respect to leaf N % concentration at a level of 0.05 in *Fraxinus* and at a level of 0.01 in *Laurus* (Table 2) because leaf nitrogen concentration positively correlated with the rates of photosynthesis. An excessive early withdrawal of nitrogen might reduce the photosynthetic yield of the leaf. At the end of the growing season is interpreted as part of programmed reallocation of resources within the plant. It can also be defined as a complex series of coordinated processes that result in

substantial removal of nutrients from the leaves. Removal of nutrients seems responsible for the decline in photosynthetic capacity and other ecological and physiological processes (20, 21).

The relationship of nitrogen to other elements can be used as a comparative tool because the concentration ratio for a given element pair seems to be more or less species specific (9). Significant differences have been found between seasons in respect to N/Ca and N/Mg ratios in *Fraxinus* and N/K ratio in *Laurus*. Magnesium is vital to the photosynthetic carbon fixation of leaves. Calcium has close metabolic association with magnesium (22). Statistically significant differences may be explained in terms of close relationships between calcium and magnesium.

Leaf parameter	<i>F. angustifolia</i> subsp. <i>oxycarpa</i>	<i>L. nobilis</i>
N %	(G) 0.61 ± 0.06 (S) 0.31 ± 0.04	0.52 ± 0.08 0.62 ± 0.09
P %	(G) 0.0036 ± 1.16 x 10 ⁻³ (S) 0.0048 ± 9.86 x 10 ⁻⁴	0.0074 ± 4.93 x 10 ⁻⁴ 0.0076 ± 6.72 x 10 ⁻⁴
K %	(G) 0.94 ± 0.17 (S) 0.81 ± 0.13	0.4 ± 0.02* 0.27 ± 0.03
Ca %	(G) 0.77 ± 0.12 (S) 0.44 ± 0.08	0.47 ± 0.18 0.59 ± 0.12
Mg %	(G) 0.65 ± 0.13 (S) 0.49 ± 0.12	0.70 ± 0.13 0.48 ± 0.16
Organic matter %	(G) 8.41 ± 1.51 (S) 9.50 ± 1.08	7.33 ± 1.08 7.78 ± 1.13

G = Mid-growing season S= End of the growing season. * P < 0.05.

Table 3. Mean values and corresponding standard errors of soil N %, P %, K %, Ca %, Mg % and organic matter % concentrations. Asterisk indicates that there are statistically significant differences between the two growth periods.

Parameters	Correlation with sclerophyll index	
	<i>F. angustifolia</i> subsp. <i>oxycarpa</i>	<i>L. nobilis</i>
N %	-.036	-.306
P %	-.646**	-.617**
K %	-.153	-.317
Ca %	-.017	.018
Mg %	-.434	-.416
N/P	-.335	-.350
N/K	-.060	-.097
N/Ca	-.387	-.393
N/Mg	-.610**	-.318
Soil N %	-.082	-.211
Soil P %	.476	.085
Soil K %	.167	-.236
Soil Ca %	-.467	-.105
Soil Mg %	.048	.212
Soil organic matter %	.211	.056
PH	.472	-.168

P < 0.01.

Table 4. Pearson correlation coefficients between sclerophyll index and leaf and soil parameters.

Evergreen communities usually occur on soils that are low in potassium (4). However, deciduous *Fraxinus* grows on soils richer in potassium, and potassium concentration is one of the most important parameters in the

distribution of deciduous and evergreen species. Other parameters include pH and other macroelements (20) (Table 3).

As shown in Table 4, the sclerophyll index was negatively correlated with leaf P % concentration in both species ($P < .01$). This offers strong support to Loveless's idea that decrease in phosphorus causes a proportional increase in the degree of sclerophylly (1, 4).

Significant correlation coefficients were obtained for leaf and soil nutrient concentrations in *Fraxinus* and *Laurus* (Table 5). Leaf P %-soil P % and leaf K %-soil K % concentrations were positively correlated ($P < .01$) at the end of the growing season in *Fraxinus*. In addition, leaf K %-soil K % concentrations were positively correlated with each other during mid-growing season. Leaf P % - soil P % and leaf K % - soil K % concentrations were positively correlated with each other during mid-growing season and leaf Ca % - soil Ca % concentrations were positively correlated with each other during both of the growth periods in *Laurus*. A feedback between leaf and soil phosphorus levels was also found by Knops & Koenig (23) in some *Quercus* species in California. However, they found no significant relationships in respect to leaf and soil nitrogen levels. According to the results of the present study the correlations between leaf N % - soil N % concentrations

and leaf Mg % - soil Mg % concentrations during mid-growing season and at the end of the growing season were not significant in either species. Additionally, leaf P % - soil P % concentrations during mid-growing season and leaf Ca % - soil Ca % concentrations during mid-growing season and at the end of the growing season in *Fraxinus* were not significantly correlated with each other. Similarly leaf K % - soil K % concentrations at the end of the growing season and leaf Mg % - soil Mg % concentrations during both of the growth periods were not statistically significant in *Laurus*. In light of these results, it could be concluded that there was no consistent relationship between soil nutrients and corresponding leaf nutrient element levels. However, this is not true for all cases, at least in respect to some nutrients (e.g., leaf phosphorus levels), and the relationships between leaf and soil nutrient levels are caused by species-specific differences, as indicated by Knops and Koenig (23).

Sclerophylly, the interactions between sclerophylly and foliar chemistry, and the role of soil factors on sclerophylly are not yet fully understood. Therefore, additional research is needed on the foliar chemistry of deciduous and evergreen species.

Table 5. Pearson correlation coefficients between leaf and soil nutrient concentrations.

Species	<i>F. angustifolia</i> subsp.		<i>L. nobilis</i>	
Parameters	G	S	G	S
Leaf N % – Soil N %	.0447	– .577	.3586	– .5143
Leaf P % – Soil P %	.4125	.8020**	.7346**	.5478
Leaf K % – Soil K %	.8110**	.8072**	.8810**	–.3901
Leaf Ca % – Soil Ca %	– .1985	– .0201	.7073**	.6316**
Leaf Mg % – Soil Mg %	.5039	.4629	.1037	.3069

G= Mid-growing season S= End of the growing season ** = $P < .01$

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