Seasonal variation of macro- and micro- nutrients in leaves of fig (*Ficus carica* L.) under Mediterranean conditions

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Abstract. Leaves of 'Smyrna' fig (*Ficus carica* L.), variety 'Smyrneiki', were collected at four stages of the annual growth cycle, namely (1) at flowering, (2) during fruit development, (3) at fruit maturity and (4) after fruit harvest, during two consecutive years (2018 and 2019) and the leaf macro-(N, P, K, Ca, Mg) and micro-(Fe, Zn, Cu, Mn, B) nutrient concentrations were determined. Mean concentrations of N, P, K, Ca and Mg ranged between 14.4–28.6, 0.5–1.7, 2–31.2, 22–80.3 and 2.1–6.7 g kg^{-1} (on a dry weight basis-d.w.), respectively, while concentrations of Fe, Zn, Cu, Mn, and B, ranged between 84–280, 11–70, 2–86, 40–206, and 18–39 mg kg^{-1} d.w., respectively. The mean leaf concentration of N decreased significantly at each successive growth stage, whereas those of P, K, Fe and Zn, also decreased progressively, but not always to a statistically significant level between each stage. In contrast, the mean leaf concentration of Ca increased significantly throughout the season, while the concentrations of Mg and Cu also increased, but not to a statistically significant level at each stage. The Mn concentration of fig leaves decreased significantly at fruit maturity, then increased significantly after fruit harvest. Overall, the nutrient concentration of fig leaves varies throughout the period from flowering to fruit harvest, suggesting that trees may need different amounts of nutrients depending on the developmental stage. The seasonal variation of the nutrient concentration in fig leaves confirmed the need for reference values for each phenological stage for leaf analysis interpretation and for developing an efficient fertilization program.

Key words: developmental stages, fig nutrition, reference nutrient values.

INTRODUCTION

The common fig (*Ficus carica* L.) belongs to the Moraceae family; it is native to Asia Minor, Iran and Syria and currently grows as a wild or domesticated species in most of the Mediterranean countries (Therios & Dimassi-Theriou, 2013). Figs are cultivated in the Mediterranean Basin, Iran and northern India, as well as in many regions with similar climatic conditions, such as the USA and Mexico (Flaishman et al., 2008). Its edible fruits are consumed fresh or dried and are used to prepare fig jam and syrup. Greece supplies more than 6% of global exports of dried figs. The mean annual production in Greece in the last 20 years is about 38,200 kg of dried figs from orchards covering an area of approximately 6,000 ha (FAO, 2017). Depending on the fruit

pollination requirements, figs can be from several types. In the current study, the fig belongs to the 'Smyrn' type, and the variety is called 'Smyrneiki'. The fruit of this variety is medium-large, spherical with a small neck, green-yellow, thin peel and has a rich flavor. It is mostly consumed dried but can also be eaten fresh. The tree is deciduous, reaching a height of 2–5 m; it is productive and has a lifespan of 50–60 years (Therios & Dimassi-Theriou, 2013). In Greece, it is cultivated in warm coastal areas, particularly in Evia (207.2 ha) and Messinia (83.3 ha) (https://www.statistics.gr).

In each plant tissue the nutrient concentrations depend on the uptake, vitality, transport, and movement of nutrients within the plant. All these processes may be affected by climatic factors. The content in macro and micronutrients of both leaves and other plant tissues varies with the age of the tissue and the time of sampling (Stylianidis et al., 2002). Leaves are organs in which nutritional elements accumulate and major metabolic processes occur. Thus, leaf analysis is the best means of diagnosing tree nutritional status, as well as indicating nutrient deficiency, toxicity, or imbalance, as reported for other woody crops (Arrobas et al., 2018; Cancela et al., 2018). Smith et al. (1987) reported that deciduous crops show seasonal changes in the mineral composition of the leaves and this can have important implications in relation to the diagnosis of nutrient disorders, the post-harvest storage of the fruit and in the timing of fertilizer applications. The nutrient accumulation curves throughout the growing season are good indicators of fruit tree nutrient demands for any developmental stage. They are also a useful tool to evaluate orchard nutritional status and to estimate the amount of soil nutrient removal (Nachtigall & Dechen, 2006). It is well known that leaf analysis must be based on standardized sampling methods and that results must be compared only with standard values obtained by those procedures. Standard values for figs are scarce and only two studies, namely those of Brown (1994), and Ersoy et al. (2003) are available. These values refer to nutrient concentrations in leaves sampled from the youngest, fully expanded, exposed leaves on non-fruiting branches in the fruit development stage (Brown, 1994) and from the third node, which was counted from the base of the shoots (Ersoy et al., 2003). The seasonal variations in fig leaf-nutrient concentration are necessary to understand and interpret the physiological aspects of fig nutrition. These seasonal changes are not available for all cultivars and varieties of figs. The most detailed nutrition survey of figs was conducted by Proebsting & Warner (1954) in commercial southern California 'Adriatic' and 'Calimyrna' orchards in the 1940s and 1950s. Also, many studies have been carried out in Turkey (Anac et al., 1982; Aksoy et al., 1987; Askin et al., 1998; Hakerlerler et al., 1998).

To achieve maximum production and high quality, the nutritional status of cultivated trees must be optimal throughout the growing season (Chatzissavvidis, 2005). Due to the lack of information on fig nutrition during the growing period in Greece and other countries in the Mediterranean region (except for Turkey), we determined the leaf concentrations of N, P, K, Ca, Mg, Fe, Zn, Cu, Mn, and B in the fig tree variety 'Smyrneiki' over different stages during the annual growth cycle. The objective of this study was to provide valuable information of macro and micro elements in fig leaves at different developmental stages for the design of a more efficient fertilization program under Mediterranean climatic conditions.

MATERIALS AND METHODS

Experimental site
This experiment was carried out in Istiaia (38° 56' 7.96" N 23° 08' 38" E), a province in the North of the Evia island, Greece. The island is characterized by a Mediterranean climate with hot dry summers and cold wet winters. The average annual temperature of the area is 17.2 °C. The hottest and the coldest months are July and January with mean temperatures of 27.4 and 7.5 °C, respectively. The mean annual precipitation is 445 mm. The highest precipitation occurs in December with an average of 60.6 mm. The total dry period lasts for approximately three months (June-August).

Plant sampling and analytical methods
The fig orchard was non-irrigated, occupied an area of about 2.5 ha and consisted of 260 mature (22 years old) trees. The trees were in a 10 m intra-row and 8.5 m inter-row spacing arrangement. The mean yield of marketable fig for this orchard during the last ten years was approximately 40,000 kg ha^{-1} year^{-1}. The yield of marketable fig from the orchard in 2018 and 2019 was approximately 30,000 kg ha^{-1} and 44,000 kg ha^{-1}, respectively. The low yield in 2018 was caused by a high incidence of fruit cracking due to the occurrence of rainfall at fruit maturity. All trees received the same fertilizers for 10 years at least; i.e. each tree received 1.5 kg of mono ammonium phosphate (12–52–0) and 1.5 kg of potassium sulphate (0–0–50) in mid-November in 2017 and 2018, respectively. At the end of the following March in each year, 1.5 kg 'nutrimore winner' (commercial name of fertilizer containing 40% total N (35.5% Uric Nitrogen and 5% Ammoniacal Nitrogen)) was applied to each tree.

Fifteen uniform, healthy trees were selected for sampling. The sampled trees were marked (2018) to be sampled again the next year (2019). Fifty young, fully expanded and exposed leaves on non-fruiting branches were collected from the perimeter of each tree at 1.8 m height, as proposed by Beutel et al. (1983). Leaf samples were collected on mid- to late May, June, late August, and October; these dates corresponded to the flowering, fruit development, fruit maturity and postharvest stages of the growth cycle, respectively. Extremely vigorous or weak shoots were avoided at all samplings. Leaves were stored in paper bags and refrigerated at 15 °C for 1 day. Once in the laboratory, the leaves were washed with deionized water, dried at 55 °C for 48 h, ground in a stainless steel Wiley mill, passed through a 250 μm plastic sieve and stored in covered plastic test tubes until analysis. Total nitrogen was determined by the Kjeldahl method (Bremner & Mulvaney, 1982). For determination of other elements, 0.5 g of the ground material was dry-ashed in a muffle furnace at 550 °C for 3.5 h. Then, the ash was dissolved in 3 mL 6N HCl and diluted to 50 mL with distilled water. The concentrations of Mg, Fe, Mn, and Zn in the clear solution were determined by flame atomic absorption spectrophotometry (Varian, A–300; Varian Techtron Pty. Ltd., Australia), using an air–acetylene flame, while Ca concentration was determined using an acetylene–N$_2$O flame. Potassium was measured using flame photometry (Microprocessor Flame photometer model 1332, Esica Ltd., India). Total P was determined using the Murphy & Riley (1962) method with a PG T60 UV/VIS Spectrophotometer (PG Instruments Ltd., United Kingdom), at 880 nm. Boron was determined using the azomethine–H method (Wolf, 1971) employing the above-mentioned spectrophotometer at 420 nm. The methods of leaf analysis used are described with further details in Klute, (1986).

Soil sampling and analytical methods

Equidistant around the circumference of a circle of 0.5 m radius from the trunk of each selected tree, three samples of soil were taken at depths of 0–30 and 30–60 cm. Then the three samples from each depth and each tree were combined and mixed separately, resulting in 15 mixed soil samples from the depth of 0–30 cm and 15 from the depth of 30–60 cm. Soil samples were taken in October 2017, transferred to the laboratory and dried at room temperature, after which they were ground and passed through a 2 mm sieve (fine earth fraction) for the determination of the following properties: the pH was determined in a soil:water (1:1) suspension (Mclean, 1982); soil texture was determined using the hydrometer method (Gee & Bauder, 1986); organic matter was determined using a modified Walkley–Black method (Nelson & Sommers, 1982); the $CaCO_3$ equivalent by using the quantity of CO_2 produced on reaction with HCl (Nelson, 1982); the cation exchange capacity (CEC) was determined with ammonium acetate method at pH 7.0 (Rhoades, 1982); exchangeable bases were determined using an NH_4OAc (1N, pH 7) method (Thomas, 1982); total nitrogen was estimated by the Kjeldahl procedure (Bremner & Mulvaney, 1982); total P was determined using the Olsen's method (Murphy & Riley, 1962); Fe, Mn, Cu and Zn were estimated by the DTPA method (Linsday & Norvell, 1978); while B was extracted by hot water and determined by colorimetry (420 nm) using the azomethine–H method (Gupta, 1979).

Statistical analysis

According to the Hartley's F max test, variance of leaf nutrient concentrations at a given developmental stage between the two cultivated years were homogeneous so the data of the two years were pooled for each developmental stage and analysis of variance performed. Where a significant difference was found, Duncan's multiple range test at the 5% level of probability was used to compare mean nutrient concentration at different developmental stages. Statistical analysis and graph preparation were carried out with STATISTICA (Statsoft, 2007). The mean leaf nutrient concentration data are expressed as the mean of the two years studied.
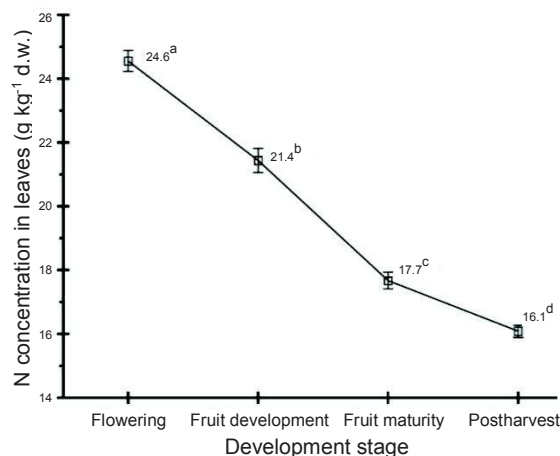
RESULTS AND DISCUSSION

Soil analysis

Table 1 shows the values of the different soil properties determined in the current study. The texture of all soil samples at both depths was clay loam (CL). The pH values were slightly alkaline. Equivalent calcium carbonate content was greater than 100 g kg^{-1}, which characterizes marl soils. Organic matter content was very low, but CEC was greater than 21.5 cmol$_{(+)}$kg^{-1} in the 0–30 cm soil layer, indicating the soil to be fertile. However, the levels of soil N were low, implying that fertilization would be required to replenish N consumed by the trees. Total P concentration was at threshold limits (total P threshold limits are 15 mg kg^{-1}). Potassium was low, but calcium and magnesium concentrations were high: 33.75 cmol$_{(+)}$kg^{-1} soil and 1.95 cmol$_{(+)}$kg^{-1} soil, respectively. Levels of micronutrients (Fe, Zn, Cu, Mn, B) ranged within their corresponding threshold limits. The evaluation of soil nutrient status was based on Landon (1991).

Table 1. Physical and chemical properties of the soil samples collected under the fig trees

Depth	Units	0–30 cm	30–60 cm
Sand	g kg^{-1}	342	381
Silt		312	291
Clay		346	328
Texture		CL*	CL
pH		7.65	7.7
Eq. CaCO$_3$	g kg^{-1}	132.5	165.5
Org. matter		0.8	0.3
Total N		1.25	0.65
P-Olsen		17.8	15.6
Exch. Ca	cmol$_{(+)}$kg^{-1}	33.75	33.3
Exch. Mg		1.95	1.6
Exch. K		0.45	0.3
Exch. Na		0.3	0.3
CEC*		23.8	21.4
Fe$_{(DTPA)}$		2.3	2.05
Zn$_{(DTPA)}$		0.75	0.5
Cu$_{(DTPA)}$	mg kg^{-1}	4.55	2.3
Mn$_{(DTPA)}$		0.55	0.3
B$_{(zeon\ H2O)}$		3.8	2.7

*CEC = cation exchange capacity; CL = Clay loam.

Plant analysis
Macronutrients

Table 2. Mean, minimum, and maximum concentrations of the macronutrients N, P, K, Ca and Mg in the leaves of the fig variety 'Smyrneiki' over the sampling period (2018–2019)

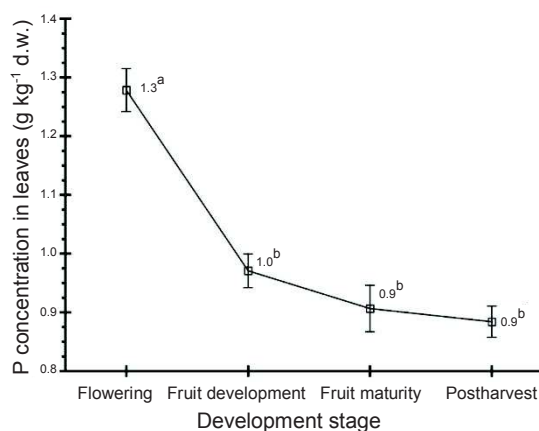|	N	P	K	Ca	Mg
	G kg^{-1} d.w.				
Mean	20.3	1.0	13.1	43.3	4.1
Minimum	14.4	0.5	2.0	22.0	2.1
Maximum	28.6	1.7	31.2	80.3	6.7

Nitrogen (N)

Mean leaf N concentrations were 24.6, 21.4, 17.7, and 16.1 g kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, and the mean value for the total sampling period was 20.3 g kg^{-1} d.w. (Table 2). The mean N in the leaves of the studied fig variety decreased significantly between each stage until the end of the sampling period (Fig. 1), probably due to utilization of N by the trees. Similar patterns of N leaf content in 'Sarilop' and 'Yesilguz' fig leaves were found by Brown (1994) and Ersoy et al. (2003), respectively, while Vemmos et al. (2013) reported that the leaf N concentration of three fig cultivars ('Kalamon', 'Mission' and 'Farkasana') decreased with plant age. In contrast, Cruz et al. (2019) reported that N concentration in fig leaves decreased, but not significantly, throughout the growing season.

Figure 1. Nitrogen (N) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean (± 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.
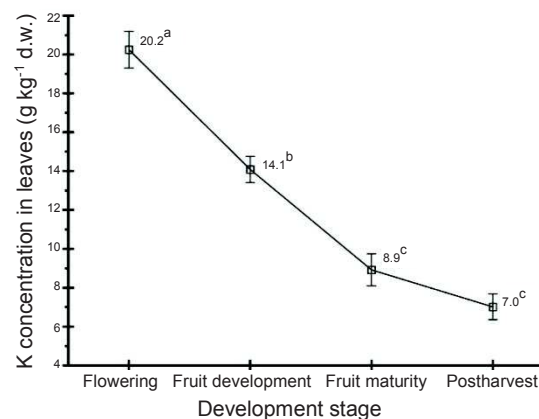
Phosphorus (P)

Leaf P mean concentrations were 1.3, 1.0, 0.9, and 0.9 g kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, and the mean value for the total sampling period was 1.0 g kg^{-1} d.w. (Table 2). Mean leaf P content decreased sharply from flowering to the fruit development stage, but then remained constant (Fig. 2). The mean P values were greater than 0.9 g kg^{-1} in the late spring sampling (flowering) but low compared to most other tree crops (Beutel et al., 1983; Reuter & Robinson, 1986). Other woody crops, such as grapevines, can have even lower P leaf contents (Romero et al., 2014; Cancela et al., 2018). Proebsting & Warner (1954), Ersoy et al. (2003), and Brown (1994) reported that P concentrations in fig leaves decreased

Figure 2. Phosphorus (P) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean (± 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.

over the growing season, whereas Cruz et al. (2019) reported that fig leaf P concentration increased and decreased throughout the growing season.

Potassium (K)

Mean leaf K concentrations were 20.2, 14.1, 8.9, and 7.0 g kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, and the mean value for the total sampling period was 13.1 g kg^{-1} d.w. (Table 2). Mean leaf K concentration decreased markedly, by about 56%, from flowering to fruit maturity, 4–5 months after flowering (Fig. 3). A similar pattern of variation in mean leaf K concentration throughout the growing season was reported by Ersoy et al. (2003) and Brown (1994), as in most deciduous crop species (Smith et al., 1987, Nachtigall & Dechen, 2006; Mirdehghan & Rahemi, 2007; Cruz et al., 2019). Proebsting & Warner (1954) recorded similar mean K concentrations in fig leaves at fruit maturation.

Figure 3. Potassium (K) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean (± 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.
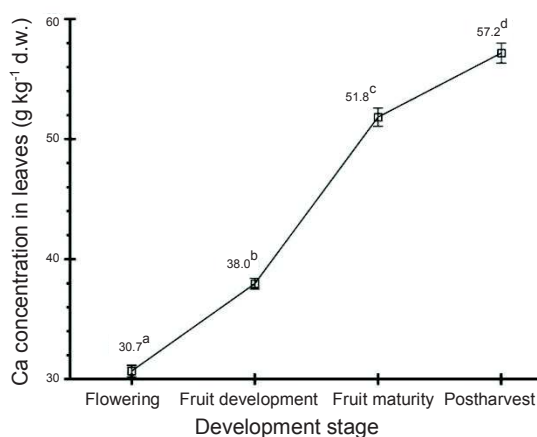
Calcium (Ca)

Mean leaf Ca concentrations were 30.7, 38.0, 51.8, and 57.2 g kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, and the mean value for the total sampling period was 43.3 g kg^{-1} d.w. (Table 2). The mean leaf Ca concentration increased significantly from flowering to postharvest (Fig. 4). Ersoy et al. (2003), reported that the Ca concentration of leaves of 'Yesilguz' figs increased rapidly until the leaves were 3 to 4 months of age, after which there was very little change.
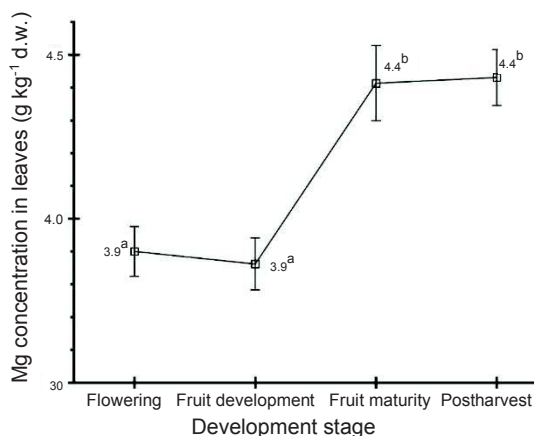
Magnesium (Mg)

Mean leaf Mg concentrations were 3.9, 3.9, 4.4, and 4.4 g kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, with a mean value of 4.11 g kg^{-1} d.w. for the total sampling period (Table 2). Mean leaf Mg concentration increased from fruit development to fruit maturity, but then remained constant (Fig. 5). The same pattern for Mg in other fig cultivars was reported by Ersoy et al. (2003) and Brown (1994).

The leaf concentrations of N, P, K in the studied fig variety decreased during the growth cycle. This reduction should be related to a dilution effect occurred with leaf growth and to the nutrient redistribution to other plant organs (shoots, fruits) throughout the end of cycle. The increase in leaf Ca concentration from flowering to the postharvest stage was probably due to the immobility of Ca in plant tissues and no redistribution to other plant organs. The increase in leaf Mg concentration was likely a consequence of lower K competition since leaf K decreased during the growth period (Nachtigall & Dechen, 2006).

Figure 4. Calcium (Ca) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean ($\pm$ 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.

Figure 5. Magnesium (Mg) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean ($\pm$ 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.

Micronutrients

Iron (Fe)

Mean leaf Fe concentrations were 207, 164, 166, 112 mg kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, and the mean value for the total sampling period was 161 mg kg^{-1} d.w. (Table 3). Mean leaf Fe concentration decreased from flowering to the fruit development stage, then remained constant until fruit maturity, followed by a significant decline (Fig. 6). The Fe concentration at the stage of fruit maturity (July) was greater than 70 μg g^{-1}, which is considered adequate for most tree species (Jones, 1998).

Table 3. Mean, minimum, and maximum concentrations of the micronutrients Fe, Zn, Cu, Mn and B in the leaves of the fig variety 'Smyrneiki' over the sampling period (2018–2019)

	Fe	Zn	Cu	Mn	B
	mg kg^{-1} d.w.				
Mean	161	27	12	91	29
Minimum	84	11	2	40	24
Maximum	280	70	86	206	39

Zinc (Zn)

Mean leaf Zn concentrations were 36, 26, 21, 22 mg kg^{-1} d.w for the flowering, fruit development, fruit maturity and postharvest stages, respectively, with a mean value for the total sampling period of 27 mg kg^{-1} d.w. (Table 3). The highest values of Zn concentration occurred at flowering, then decreased significantly to the fruit development stage and subsequently remained constant (Fig. 7). Similar patterns were obtained by Brown (1994) and Ersoy et al. (2003). The mean leaf Zn concentration was adequate for tree growth, according to Jones (1998).
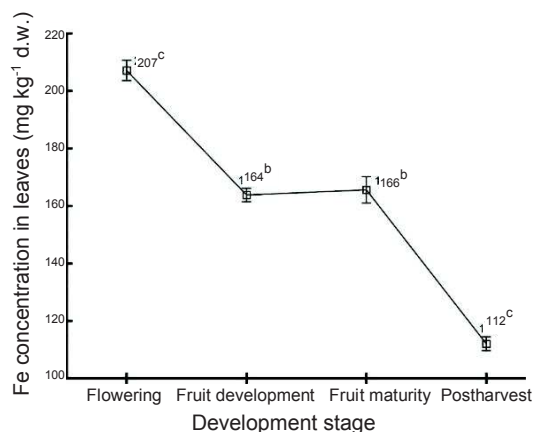
Figure 6. Iron (Fe) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean (± 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.
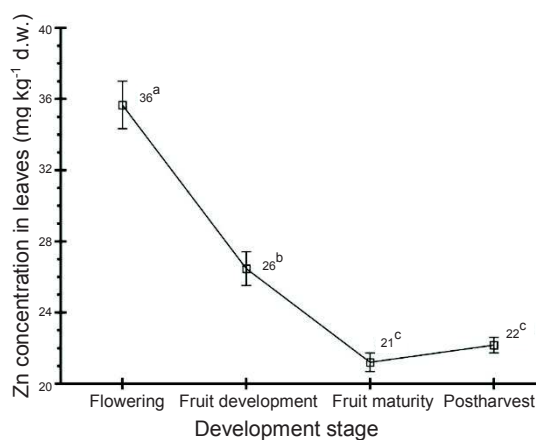
Figure 7. Zinc (Zn) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean (± 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.
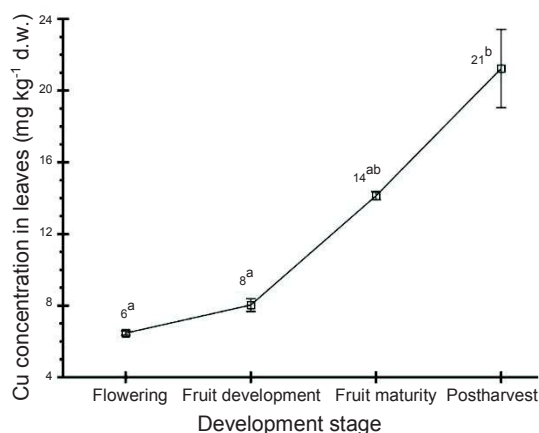
Copper (Cu)

Mean leaf Cu concentrations were 6, 8, 14, and 21 mg kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, with a mean value for the total sampling period 12 mg kg^{-1} d.w. (Table 3). The mean Cu concentration in fig leaves increased progressively with each stage, but only to a statistically significant level between flowering and postharvest (Fig. 8). Ersoy et al. (2003) reported that the Cu concentration in fig leaves decreased with increasing leaf age. Mean leaf Cu concentrations at all stages were higher than 6 mg kg^{-1}, which is considered adequate for most tree species (Jones, 1998).
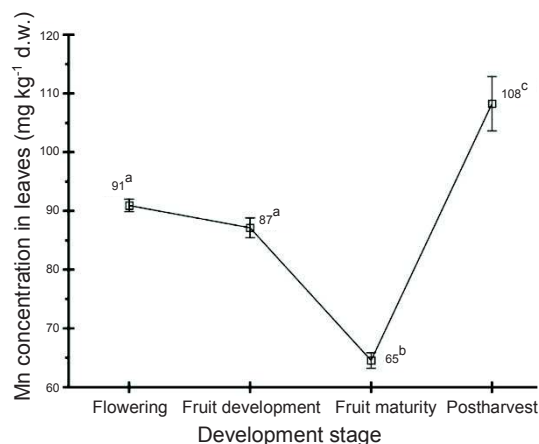
Manganese (Mn)

Mean leaf Mn concentrations were 91, 87, 65, and 108 mg kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively, and the mean value for the total sampling period was 91 mg kg^{-1} d.w. (Table 3). Manganese concentration along the growing cycle follows a unique pattern, decreasing sharply from fruit development to fruit maturity and then increasing sharply to postharvest development stage (Fig. 9).

Boron (B)

Mean leaf B concentrations were 28, 28, 30, and 32 mg kg^{-1} d.w. for the flowering, fruit development, fruit maturity and postharvest stages, respectively and the mean value for the total sampling period was 29 mg kg^{-1} d.w. (Table 3). Mean leaf B concentration did not differ between

Figure 8. Copper (Cu) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean ($\pm$ 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.
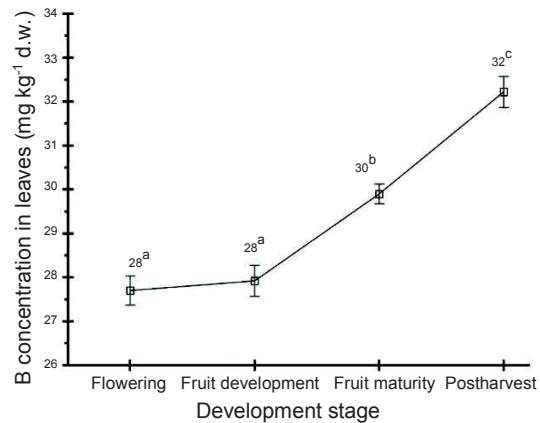

Figure 9. Manganese (Mn) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean ($\pm$ 0.5*SE). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.

flowering and fruit development, but thereafter increased significantly (Fig. 10). Brown (1994) reported that fig leaves are possible B accumulators, like walnut and pistachio. Boron requirements for most plant species are poorly defined because higher amounts

of B are required for flowering and fruit production than for vegetative growth (Hansen et al., 1985).

The mean leaf values of N, P, K, Fe, Ca concentrations at the fruit development stage agree with those reported by Hakerlerler et al. (1998) from 10 fig cultigens. The mean values for macro- and micro-nutrients in fig leaves recorded here were higher than the threshold limits of deficiency (Reuter & Robinson, 1986) probably thanks to the correct time and dose of fertilizers. Mean leaf Fe, Zn, Cu, and B contents showed less intense fluctuation, decreasing (Fe, Zn) or increasing (Cu, B) throughout the sampling period, but not to statistically significantly levels at all stages. No deficiency was observed in any instance.

A classical method for developing a practical basis for fertilizing commercial plants is to define critical nutrient concentrations (reference values) in each species and tissue and relating them to yields (Smith, 1962).

In the studied fig orchard the yields of marketable fig, under the same fertilization treatment applied for 10 consecutive years, were at high levels (40–50 kg tree^{-1}); therefore the macro and micro nutrient concentrations in leaves at different tree developmental stages could be consider critical and hence be used as reference values for developing an efficient fertilization program under Mediterranean climatic conditions.

Figure 10. Boron (B) concentration in leaves of the fig variety 'Smyrneiki' at four stages in the annual growth cycle of the tree on average for two consecutive years. Error bars represent standard error of the mean ($\pm 0.5*SE$). Means at different sampling time followed by the same letter are not significantly different according to Duncan's multiple range test at $P \leq 0.05$.

CONCLUSIONS

Fig leaf nutritional concentrations varied throughout the growing period, indicating that the plants have different nutrient requirements at different developmental stages. The sharp decrease in potassium from flowering until fruit maturity development stage is particularly remarkable and should be considered when developing a fertilization plan. The measured values of each nutrient plotted versus time can be useful in explaining several phenomena during the bearing cycle of the fig tree and can also be used to support decision making for the optimum fertilization of fig trees. Overall, the variation in nutrient contents of fig leaves is similar to that of most deciduous trees. The leaf nutrient concentrations observed at different developmental stages in the current study could be used as standard reference values for leaf analysis interpretation and for developing an optimum fertilization program.

REFERENCES

Aksoy, U., Anac, D., Eryuce, N. & Yoltas, T. 1987. Determination and evaluation of the nutrients status of fig orchards in the Aegean Region. *J. Ege Univ. Fac. Agric.* **24**(2), 21–35.

Anac, D., Aksoy, U., Hakerlerler, H. & Duzbastõlar, M. 1982. *Nutritional status of fig orchard in the small meander basin and relationships between soil and leaf nutrients and some yield and quality attributes*. Taris Research and Development Centre Project no:**4**.

Arrobas, M., Afonso, S. & Rodrigues, M.A. 2018. Diagnosing the nutritional condition of chestnut groves by soil and leaf analyses. *Sci. Hortic*. **228**, 113–121.

Askin, A., Ceyls, S. & Yener, H. 1998. A study on the national status of fig orchards in birgi-irimagzi. *Acta Hort.* **480**, 239–246.

Beutel, J., Uriu, K. & Lilleland, O. 1983. *Leaf analysis for Californian deciduous fruits*. Soil and Plant Tissue Testing in California Bull. 1879. Univ. of California, Berkeley, 17 pp.

Bremner, J.M. & Mulvaney, C.S. 1982. Salicilic acid thiosulfate modification of Kjieldahl method to include nitrate and nitrite. In A.L. Page (ed), *Methods of Soil Analysis*. Part 2. *Chemical and Microbiological Properties*. second ed. Amer. Soc. Agron. No **9** (Part 2) in the Agronomy Series. ASA, SSSA, Madison, WI., pp. 595–562.

Brown, P.H. 1994. Seasonal variations in fig *(Ficus carica* L.) leaf nutrient concentrations. *HortScience* **29**, 871–873.

Cancela, J.J., Fandiño, M., González, X.P., Rey, B.J. & Mirás-Avalos, J.M. 2018. Seasonal variation of macro and micronutrients in blades and petioles of *Vitis vinifera* L. cv. Mencía and Sousón. *J. Plant Nutr. Soil Sci.* **181**, 498–515.

Chatzissavvidis, C., Therios, I. & Molassiotis, A. 2005. Seasonal variation of nutritional status of olive plants as affected by boron concentration in nutrient solution. *J. Plant Nutr.* **28**, 309–21.

Cruz, A.F., Almeida, G.M de, Wadt, P.G.S., Pires, M. de C. & Ramos, M.L.G. 2019. Seasonal variation of plant mineral nutrition in fruit ttrees. *Braz. Arch. Biol. Technol.* **62**, e19180340. Epub October 24, 2019. https://dx.doi.org/10.1590/1678-4324-2019180340

Ersoy, N., Gözlekçi, S. & Kaynak, L. 2003. Seasonal variations in the content of nutrient elements in the leaves of fig (*Ficus carica* L. Yesilguz). *Acta Hortic.* **605**, 269–275.

FAO. 2017. Food and agriculture organization of the United Nations statistical databases. Available online: http://www.fao.org/faostat/en/ (accessed on 2 May 2020).

Flaishman, M.A., Rover, V. & Stover, E. 2008. The Fig: Botany, Horticulture, and Breeding. *Hortic Rev.* **34**, 113–197.

Gee, G.W. & Bauder, J.W. 1986. Particle-size Analysis. In A.L. Page (ed.). *Methods of soil analysis*, Part 1. *Physical and mineralogical methods*. second ed. Amer. Soc. Agron. No **9** (Part 1) in the Agronomy Series. ASA, SSSA, Madison, WI., pp. 383–411.

Gupta, U.C. 1979. Some factors affecting the determination of hot-water boron from potzol soils using azomethine –H. *Can. J. Soil Sci*. **59**, 241–247.

Hakerlerler, H., Saatçi, N., Hepaksoy, S. & Aksoy, U. 1998. Fruit and leaf nutritional status of some Fig clones and cultivars and relationships with some sugar fractions. *Acta Hortic.* **480**, 247–252.

Hansen, E.J., Chaplin, M.H. & Breen, P.J. 1985. Movement of foliar boron out of leaves and accumulation in flower buds and flower parts of 'Italian' prune. *Hortscience* **20**, 747–748.

Jones, B.J. Jr., 1998. *Plant Nutrition Manual*. CRC press, Boca Raton, FL., USA, 149 pp.

Klute, A. 1986. *Methods of Soil Analysis.* Part 1. *Physical and mineralogical methods.* second edition. Amer. Soc. Agron. No **9** (Part 1) in the Agronomy Series. ASA, SSSA, Madison, WI., pp. 1–1188.

Landon, R.J. 1991. *Booker Tropical Soil Manual. A Handbook for Soil Survey and Agricultural Land Evaluation in the Tropics and Subtropics*. Booker Agriculture International Ltd. Longman Publ. New York, 450 pp.

Linsday, W.L. & Norvell, W.A. 1978. Development of a DTPA soil test for zinc, iron, manganese, and copper. *Soil Sci. Soc. Am. J.* **31**, 421–428.

McLean, E. 1982 Soil pH and Lime Requirement. In A.L. Page (ed), *Methods of Soil Analysis*. Part 2. *Chemical and Microbiological Properties*. second ed. Amer. Soc. Agron. No **9** (Part 2) in the Agronomy Series. ASA, SSSA, Madison, WI., pp. 199–223.

Mirdehghan, S.H. & Rahemi, M. 2007. Seasonal changes of mineral nutrients and phenolics in pomegranate (*Punica granatum* L.) fruit. *Sci. Hortic.* **111**, 120–127.

Murphy, J. & Riley, J.P. 1962. A modified single solution method for the determination of phosphate in natural waters. *Anal. Chim. Acta* **27**, 31–36.

Nachtigall, G.R. & Dechen, A.R. 2006. Seasonality of nutrients in leaves and fruits of apple trees. *Sci. Agric.* **63**, 493–501.

Nelson, D.W. & Sommers, L.E. 1982. Total Carbon, Organic Carbon, and Organic Matter. In A.L. Page (ed), *Methods of Soil Analysis*. Part 2. *Chemical and Microbiological Properties*. second ed. Amer. Soc. Agron. No **9** (Part 2) in the Agronomy Series. ASA, SSSA, Madison, WI., pp. 539–577.

Nelson, R.E. 1982. Carbonate and gypsum. In A.L. Page (ed), *Methods of Soil Analysis*. Part 2. *Chemical and Microbiological Properties*. second ed. Amer. Soc. Agron. No **9** (Part 2) in the Agronomy Series. ASA, SSSA, Madison, WI., 181–196.

Proebsting, E.L. & Warner, R.M. 1954. The effect of fertilizers on yield, quality and leaf composition of figs. *Proc Am Soc Hortic Sci*. **63**, 10–18.

Reuter, D.J. & Robinson, J.B. 1986. *Plant Analysis: An interpretation manual*. Inkata Press. Melbourne, pp. 1–218.

Rhoades, J.D. 1982. Cation Exchange Capacity. In A.L. Page (ed), *Methods of Soil Analysis*. Part 2. *Chemical and Microbiological Properties*. second ed. Amer. Soc. Agron. No **9** (Part 2) in the Agronomy Series. ASA, SSSA, Madison, WI., pp. 149–157.

Romero, I., Benito, A., Dominguez, N., Escudero, EG. & Martin, I. 2014. Leaf blade and petiole nutritional diagnosis for *Vitis vinifera* L. cv. 'Tempranillo' by deviation from optimum percentage method. *Span. J. Agric. Res*. **12**(1), 206–214.

Smith, G.S., Clark, C.J. & Henderson, H.V. 1987. Seasonal accumulation of mineral nutrients by kiwifruit. *New Phytol*. **106**, 81–100.

Smith, P.F. 1962. Mineral analysis in plant tissue. Annual Review. *Plant Physiol*. **13**, 81–108.

StatSoft Inc. 2007. STATISTICA, ver. 8. Tulsa, OK.

Stylianidis, D.C., Simonis, A.D. & Syrgiannidis, G.D. 2002. *Nutrition-Fertilization of deciduous fruit trees*. Stamoulis publ., 675 pp. (in Greek).

Therios, J.N. & Dimassi-Theriou, K. 2013. *Pomology Deciduous Fruit Trees*. Published by A.S. Gartagani, Thessaloniki, Greece 844 pp. (in Greek).

Thomas, G.W. 1982. Exchangeable Cations. In A.L. Page (ed), *Methods of Soil Analysis*. Part 2. *Chemical and Microbiological Properties*. second ed. Amer. Soc. Agron. No **9** (Part 2) in the Agronomy Series. ASA, SSSA, Madison, WI., pp. 159–164.

Vemmos, S.N., Petri, E. & Stournaras, V. 2013. Seasonal changes in photosynthetic activity and carbohydrate contents in leaves and fruits of three fig cultivars. *Sci. Hort*. **160**, 198–207.

Wolf, B. 1971. The determination of boron in soil extracts, plant materials, composts, manures, water, and nutrient solutions. *Commun. Soil Sci. Plant Anal*. **2**, 363–374.