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EARLY DIAGNOSIS OF BORON DEFICIENCY IN CHESTNUT

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□ *This study aims to identify an alternative plant tissue to be used in the early diagnosis of boron (B) deficiency in chestnut (*Castanea sativa *Mill.). A B-deficient orchard was selected, and 16 trees were submitted to two levels of B fertilization. When flowers were in bloom, the following tissues were sampled: leaves, androgynous catkins and flowers. There was a significant increase in B content in plant tissues due to B application. In July, the highest B content was observed in flowers in B0, but leaves had the greatest content in B1. Boron content in the tissues collected in July was positively correlated with B contents in leaves sampled in September. Foliar B concentrations, irrespective of the sampling period, were correlated with chestnut productivity, while the other tissues did not. These results suggest that the leaves, sampled in bloom, were the most efficient tissue for the early diagnosis of B deficiency.*

Keywords: boron, chestnut, foliar analysis

INTRODUCTION

In this study the possibility was explored of using alternative tissue to diagnose boron (B) deficiency in an early stage, in order to take remedial actions in time to improve fruit yield. In northeastern Portugal the observation of visible symptoms of boron deficiency in chestnut (*Castanea sativa* Mill.) orchards is not uncommon (Portela et al., 2007). However, it is believed that many orchards are managed under hidden hunger, which obviously limits fruit production. Indeed B application to the soil has been proven to have an obvious effect on fruit set and nut productivity of chestnut (Portela et al., 2010).

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Leaf nutrient analysis is the established method used to diagnose nutritional status of fruit trees and it is an important tool for fertilizer recommendations. However, the normal period for sampling (August–September) is usually too late in the season for application of B amendments in order to improve nut yield. Leaf is generally considered the plant tissue that reflects nutrient supply from the soil, and few studies have systematically examined a range of plant parts to assess their relative efficacy in predicting yield. This study aims to identify an alternative tissue in detecting B deficiency in chestnut orchards earlier in the season. This may provide a tool to decide the correction measures to be applied in time to improve yield.

The use of flower analysis as a tool for early diagnosis of nutrient status or deficiency has been suggested by several authors (Sanz and Montañés, 1995; Pestana et al., 2001; Bouranis et al., 2001) for fruit trees such as peach, orange, and almond. As far as B is concerned, Fregoni (1980) suggested that flowers could be useful to detect nutrient deficiencies, particularly the B in grapevine. More recently Razeto and Castro (2006) considered that inflorescence was a more appropriate tissue than October leaf for diagnosing B status in avocado trees, since this nutrient had a major role in fruit setting, and correction measures were possible by foliar spraying with boron.

MATERIALS AND METHODS

A field trial was established in a 15-year-old chestnut orchard in Jou (41° 28′ 35″ N and 7° 25′ 17″ W) in northern Portugal. The land is gently undulated (5–8% slope) at the 620 m elevation. The mean precipitation is 1079 mm and mean temperature 13.3°C. Compared with average values, the year of 2007–2008 was atypical as far as temperature is concerned: the minimum temperature during bloom period, from middle-June to the beginning of July was 3–4°C lower than the average value of 12–13°C for the same period. In contrast, total rainfall and distribution during 2008 was not a serious constraint. The soils vary from Haplic Dystric Leptosol to Leptic Dystric Cambisol (FAO, 2006).

The chestnut orchard was established on a soil derived from quartzophyllites with abundant coarse fragments, and effective soil depth of 25–40 cm with good drainage. Average soil properties of composite samples are shown in Table 1. The soil B content (soluble in hot water) exhibits small variation (0.34–0.63 mg kg^{-1}) in the orchard.

A group of 16 trees of Judia variety were selected in order to apply the treatments. The trees were separated into two groups with the same mean diameter at breast height and same mean projection area. The B treatment was randomly applied to half the trees (B0 and B1). A basal fertilization was carried out beneath the crown of all trees in the winter of 2006–2007: dolomitic limestone, to correct the soil acidity, and mineral

TABLE 1 Soil physical and chemical properties at two soil depths

Property	0–20 cm	20–40 cm
Gravel, % (v)	35	35
Coarse sand, %	31	32
Fine sand, %	25	22
Silt, %	28	34
Clay, %	16	13
Texture	Loam	Silty loam
Organic matter[a], g kg^{-1}	26	12
pH (H_2O)[b]	4.8	4.6
P_2O_5 extractable[c], mg kg^{-1}	60	13
Exchangeable cations[d], cmolc kg^{-1}		
Ca	0.87	0.18
Mg	0.34	0.10
K	0.37	0.14
Na	0.15	0.15
H+Al	0.83	1.24
$CEC_{effective,}$ cmolc kg^{-1}	2.56	1.81
Base saturation ($CEC_{effective}$), %	66	32
Hot water soluble B[e], mg kg^{-1}	0.44	0.10

a: oxidation with $K_2Cr_2O_7$, by the modified Walkley-Black method; b: 1:2.5 soil solution ratio; c: colorimetric method after extraction with ammonium lactate-acetic acid solution at pH 3.7; d: 1N ammonium acetate at pH 7 and the H+Al by the 1N KCl method; e: spectrophotometry (azomethine H).

fertilizers, applied at adequate rates, to supply nitrogen (N), phosphorus (P), potassium (K), calcium(Ca), magnesium(Mg), sulfur (S), zinc (Zn), copper (Cu) and manganese (Mn). Fertilization with macronutrients was repeated in winter 2007–2008. In the beginning of March 2007, Granubor (Borax Europe Limited, Castéllon, Spain) with 14.6% B was broadcast on the soil surface underneath eight trees, at a rate of 100 g per tree (B1).

Plant sampling was carried out in the beginning of July 2008 in 10 trees randomly selected (B0 and B1) when female flowers were with full floral differentiation (>80%). Four light-exposed branches of fruiting shoots, from the upper part of the middle crown of the four quadrants of trees were picked for sampling: fully developed leaves from the fourth to the eighth position from the tips, androgynous catkins and female flowers. Leaves were sampled again in the beginning of September in the same trees and position as described above. Plant materials were dried at 65°C for 48 h and ground to pass through a 1-mm screen. They were digested as described by van Schouwenburg and Walinga (1978). Calcium, Mg, Fe, Cu, Zn and Mn were determined by atomic absorption spectrophotometry; K by flame photometry. For N and P analysis, the digestion was with sulfuric acid and their concentrations were determined on an autoanalyzer. Boron was measured spectrophotometrically by the azomethine H method.

In the fall of 2008, chestnut fruits were harvested from the ground under the same eight trees from B0 and B1. For the purpose of evaluating nut production, chestnuts which passed through a 24 mm grid were rejected as unmarketable, and the remainder was weighed in fresh.

The data was analyzed by one-way ANOVA followed by the Duncan multiple range test with JMP statistical package (SAS Institute, Cary, NC, USA). Pearson's correlation coefficients between nutrient concentrations in plant tissues and nut production per tree were determined.

RESULTS AND DISCUSSION

Boron application had an evident effect on nut production, which increased fourfold in 2008 (17 kg per tree compared with the 4 kg in the control).

With the exception of B content no differences were found between the concentrations of nutrients under the effect of B fertilization, irrespective of the plant part analyzed (androgynous catkins, female flowers and leaves). Table 2 shows the nutrient concentrations in the various tissues analyzed. As expected, fertilization significantly increased B concentration in all tissues analyzed. In general, the female flowers showed higher nutrient concentrations irrespective of B application. However, as far as B content is concerned, the pattern of variation was different. Boron concentration was highest in the flowers in B0, but leaves had the highest concentration in B1. This means that when B was in short supply, some mobilization of B occurred to the female flowers, reaching the value 13.6 mg kg^{-1}, while the androgynous catkins and leaves had about 9 mg kg. With a larger B supply it was the leaves that had greater B content (27.2 mg kg^{-1}). Relatively higher B contents in flowers have been reported by several authors (Dell and Huang, 1997; Brown and Shelp, 1997), which was interpreted as the

TABLE 2 Nutrient concentrations in chestnut tissues collected in the beginning of July at full bloom

Nutrient	No boron			Boron fertilization		
	Androgynous catkins	Flowers	Leaves	Androgynous catkins	Flowers	Leaves
N, g kg^{-1}	16.1 a	20.2 ab	21.4 b	16.5 A	17.2 AB	20.7 B
P, g kg^{-1}	1.70 a	2.26 a	1.54 a	1.57 A	2.22 B	1.36 A
K, g kg^{-1}	8.1 a	9.6 a	7.0 a	6.5 A	8.4 A	6.9 A
Ca, g kg^{-1}	16.6 b	19.4 b	9.3 a	15.4 B	20.3 C	8.1 A
Mg, g kg^{-1}	3.52 b	3.78 b	2.00 a	3.36 B	4.28 B	1.94 A
Fe, mg kg^{-1}	28 a	28 a	53 b	33 A	51 A	54 A
Mn, mg kg^{-1}	1041 a	890 a	880 a	856 A	925 A	798 A
Zn, mg kg^{-1}	66 b	69 b	25 a	61 B	75 C	25 A
Cu, mg kg^{-1}	10.0 a	8.0 a	8.0 a	12.4 B	10.4 AB	7.8 A
B, mg kg^{-1}	9.0 a	13.6 b	8.8 a	24.0 AB	18.0 A	27.2 B

Means in the same column with a letter in common (lowercase or capital) are not significantly different ($P < 0.05$) by Duncan's multiple range test.

TABLE 3 Foliar nutrient concentrations according to the treatment and sampling period

	No boron		Boron fertilization	
Nutrient	July	September	July	September
N, g kg^{-1}	21.4 a	17.3 a	20.7 A	19.4 A
P, g kg^{-1}	1.54 a	1.28 a	1.36 A	1.48 A
K, g kg^{-1}	7.0 a	6.5 a	6.9 A	7.1 A
Ca, g kg^{-1}	9.3 a	7.5 a	8.1 A	10.6 A
Mg, g kg^{-1}	2.00 a	1.52 a	1.94 A	1.60 A
Fe, mg kg^{-1}	53 a	38 a	54 A	56 A
Mn, mg kg^{-1}	880 a	692 a	798 A	619 A
Zn, mg kg^{-1}	25 b	17 a	25 A	20 A
Cu, mg kg^{-1}	8.0 a	10.6 a	7.8 A	12.0 B
B, mg kg^{-1}	8.8 a	8.3 a	27.2 A	34.1 A

Means in the same column with a letter in common (lowercase or capital) are not significantly different ($P < 0.05$) by Duncan's multiple range test.

apparent B mobility and mobilization of B to more demanding reproductive tissues (Dell and Huang, 1997). It seems that in the chestnut, with sufficient B supply, boron is mobilized to the leaves, the tissue with the highest transpiration rate, rather than to the flowers.

Table 3 shows the mean nutrient concentrations in the leaf samples collected in July and September. Application of B did not affect significantly the concentrations of most nutrients, except the B itself. It can be observed that, in general, the differences in foliar concentrations of nutrients between B0 and B1 are small in July and greater in September, though the increase is not significant. A decrease in concentration of most nutrients in September might have been expected due to a dilution effect, since in that period there is often a nutrient mobilization to the fruit. As mentioned above, the mean nut production was fourfold in B1, so it would be plausible that a higher mobilization of nutrients to the fruit had occurred. However, the tendency was inverse and there was an increase in concentration of most nutrients in September. This led us to hypothesize that the B application to the soil might have created better conditions for development of the root system and therefore a better acquisition of nutrients from the soil. The role of B in root development is well supported by Dell and Huang (1997).

The statistics of B concentration in leaves are shown in Table 4. In September, B content was respectively 8.3 and 34.1 mg kg^{-1} in B0 and B1 ($P < 0.001$). The former corresponds to an obvious B deficiency while the latter is in a range considered adequate for nut trees (Shear and Faust, 1980; Olsen, 2001) or in the range of tentative data for chestnut according Clarke (1987). While the B content did not vary in the control from July to September, it increased in B1 treatment in that period, as occurred with most nutrients.

TABLE 4 B concentration in leaves sampled in the beginning of July and in September, according to the treatment

	Sampling period	
Treatment	July	September
	mg kg^{-1}	
B0	8.8 aA	8.3 aA
B1	27.2 aB	34.1 aB

The capital letters compare lines ($P < 0.001$) and the lowercase letters compare columns ($P < 0.05$). The means with a letter in common are not significantly different.

In order to determine which tissue revealed a better correlation with foliar B concentration in September (the normal period of leaf sampling) and with nut production per tree, the correlation coefficients are displayed in Table 5. Boron concentration in all tissues (sampled in July) is well and positively correlated with leaf picked in September. However, relative to nut production (caliber >24 mm), while the leaf concentrations are significantly correlated with fruit production ($r = 0.70^{*}$), the other tissues (androgynous catkins and flowers) are not statistically correlated.

In summary, the results of the present study show that B mobilization to the flowers occurred when B was in short supply, but leaves reached higher B concentration when B was at adequate levels. They also reveal that leaves, sampled at female bloom, are the most reliable tissue to be used in the early diagnosis of B deficiency, while the alternative tissues are less reliable. These data suggest that the early diagnosis of B deficiency could be possible by sampling the leaves of fruiting shoots when female flowers begin blooming (emergence of stigma). This would still allow foliar spraying with B at full bloom in order to correct B deficiency in chestnut. While this must obviously be confirmed by further studies, foliar spraying at flowering period has been proven (Nyomara and Brown, 1997; Perica et al., 2001) to be very efficient in correcting B deficiency in fruit trees. Further studies are also needed to estimate the reference values for the interpretation of leaf analysis made at bloom stage.

TABLE 5 Correlation coefficients between chestnut production and B concentrations in several tissues and sampling period

	Foliar B in September	Foliar B in July	B in androgynous catkins in July	B in female flowers in July
Nut production	0.701*	0.702*	0.614	0.402
Foliar B in September	-	0.972***	0.892**	0.855**

*,**,***Significant at $P < 0.05$, $P < 0.01$ and $P < 0.001$, respectively.

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REFERENCES

Bouranis, D. L., S. N. Chorianopoulou, G. Zakynthinos, G. Sarlis, and J. B. Drossopoulos. 2001. Flower analysis for prognosis of nutritional dynamics of almond tree. *Journal of Plant Nutrition* 24: 705–716.

Brown, P. H., and B. J. Shelp. 1997. Boron mobility in plants. *Plant and Soil* 193: 85–101.

Clarke, C. J. 1987. Assessing the fertilizer needs of chestnuts. *Growing Today* Aug/Sept: 8–11.

Dell, B., and L. Huang. 1997. Physiological response of plants to low boron. *Plant and Soil* 193: 103–120.

FAO. 2006. World Reference Base for Soil Resources. World Soil Resources Reports No 103. Rome: FAO.

Fregoni, M. 1980. *Nutrition and Fertilization of Vineyard*. Bologna, Italy: Edagricole.

Nyomora, A. M. S., and P. H. Brown. 1997. Fall foliar-applied boron increases tissues boron concentration and nut set of almond. *Journal of American Society of Horticultural Science* 122: 405–410.

Olsen, J. 2001. Leaf analysis important in managing nutrition in the chestnut orchard. *The Western Chestnut* 3: 9–11.

Perica, S., P. H. Brown, and J. H. Connell. 2001. Foliar boron application improves flower fertility and fruit set of olive. *HortScience* 36: 714–716.

Pestana, M., J. P. Correia, A. Varennes, J. Abadia, and E. A. Faria. 2001. The use of floral analysis to diagnose the nutritional status of orange trees. *Journal of Plant Nutrition* 24: 1913–1923.

Portela, E., J. V. Ferreira-Cardoso, and J. Louzada J. 2010. Boron deficiency in chestnut. Effect of boron application on nut yield and quality. *Acta Horticulturae* 866: 315–320.

Portela, E., A. Martins, A. L. Pires, F. Raimundo, and G. Marques. 2007. Management practices in chestnut orchards: Soil management. In: *Chestnuts*, Agro Program 499, eds. J. Gomes-Laranjo, J. Ferreira-Cardoso, E. Portela, and C.G. Abreu, pp. 207–264. Vila Real, Portugal: UTAD.

Razeto, B., and M. J. Castro. 2006. Diagnosis of boron status in avocado trees (*Persea americana* Mill.) using alternative tissue. *Acta Horticulturae* 721: 291–293.

Sanz, M., and L. Montañés. 1995. Flower analysis as new approach to diagnosing the nutritional status of the peach tree. *Journal of Plant Nutrition* 18: 1667–1675.

Shear, C. B., and M. Faust. 1980. Nutritional ranges in deciduous tree fruits and nuts. *Horticultural Reviews* 2: 142–163.

van Schouwenburg, J. C., and I. Walinga. 1978. *Methods of Analysis for Plant Material*. Wageningen, the Netherlands: Agricultural University of Wageningen.