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BORON APPLICATION ON A CHESTNUT ORCHARD: EFFECT ON YIELD AND QUALITY OF NUTS

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□ Although boron (B) deficiency in chestnuts (*Castanea sativa* Mill.) has been identified in Portugal and B fertilization was carried out in some orchards, the post-treatment evaluations have not been made to date. So the objective of this trial was to confirm the B deficiency and to quantify the effect of B applications to the soil on nut yield and quality. In October 2006, a fertilizer trial was established in a 15 year-old orchard in a very acid soil derived from siliceous schists. Soil liming and a basal fertilization were carried out in 16 trees and two levels of sodium tetraborate (Granubor, 14.6% B) were applied to eight trees: control (B0) and 100 g of Granubor per tree (B1). In the beginning of September chestnut leaves were collected from five trees and analyzed for macro- and micronutrients. Nut productivity was measured per tree and some nut quality parameters were evaluated, including the chemical composition of the kernel (soluble sugars, starch, total fiber, crude protein, and crude fat) in 2007, and dry matter, nut caliber, and fruit damage in 2007 and 2008. Boron fertilization of chestnuts significantly increased nut production: 75% in the first year (8 kg and 14 kg per tree, respectively, in the B0 and B1) and, in the second year, was over four times higher (4 kg and 17 kg per tree, respectively, in the B0 and B1). No significant differences were found in dry matter, nut caliber, nut damage and chemical composition of the kernel in the first year, but in the second year lower fruit damage by chestnut tortrix ($P < 0.0089$) was observed. Foliar analyses exhibit relatively low B concentrations in the control trees (average value of 8 mg kg^{-1}) in both years, while in the fertilized trees the foliar B concentration were in average 61 mg kg^{-1} in the first year, and 34 mg kg^{-1} in the second year. The large drop in the foliar B concentration in the second year suggests the need to monitoring the nutrient status of chestnut orchards or more frequent of B application than usual practice.

Keywords: boron, yield, nut quality

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INTRODUCTION

Many soils from northeastern (NE) Portugal are prone to the development of boron (B) deficiencies. A regional survey carried out in this area by Vale (1988) shows that most soils, derived from several geological formations, are liable to B deficiency. Alleviation of B deficiency symptoms and resumption of normal pine tree growth was observed after B application by Vale et al. (1999) in soils derived from quartzitic schists. The compilation of B concentration in leaves of chestnut (*Castanea sativa* Mill.) conducted during 10 years (1996–2006) showed that this micronutrient is often in short supply in many chestnut orchards (Portela et al., 2007), and nearly 50% of the 84 orchards sampled had foliar B concentration lower than 20 mg kg⁻¹. This level was tentatively considered as the critical B concentration differentiating chestnut orchards with potential B deficiencies from orchards unlikely to have B deficiencies. The critical B concentration of 20 mg kg⁻¹, although not obtained by systematic study, was based on B levels in orchards with conspicuous B deficiencies already identified, which had foliar B concentrations between 8 and 16 mg kg⁻¹ (Portela et al., 2007).

Comprehensive listing of standard values for the prediction of B deficiency by plant analysis was prepared for fruit and nut trees by Bergman (1992), Martin-Prével et al. (1987) and Shear and Faust (1980). However, reliable data has not been established for chestnuts. The first reference to B deficiency in chestnuts (*Castanea crenata*) is in 1978 in Korea (Park et al., 1978), which refers the premature dropping of fruit in July and August, could be reduced by borax application. The B concentration in leaves was 9.8 mg kg⁻¹ dw in the control plot rising to values of 40–50 mg kg⁻¹ after borax application. Based on this study, adequate levels of B concentration in chestnuts for France were established by Breisch (1995) in the same range of 40–50 mg kg⁻¹. The typical concentrations of B for chestnuts in New Zealand were presented by Clark (1987) as 33–90 mg kg⁻¹. However, these were regarded as tentative, rather than definitive, since they were obtained by monitoring foliar concentrations in a single growing season. Preliminary reference values of 31–75 mg of B kg⁻¹, based upon hazelnut standards in the USA, were recommended for chestnut by Olsen (2001).

In NE Portugal, although B deficiency in chestnuts has been identified and B fertilization empirically carried out in several orchards, with unequivocal positive effects on tree growth and nut production, post-treatment evaluations have not been carried out to date. It is well known that boron disorders not only affect fruit setting and production, but may also affect fruit quality and its commercial acceptability. Thus, this study attempts to diagnose boron deficiency and to determine its effect on nut production and fruit quality. The results may supply useful data to highlight current chestnut fertilizer recommendations.

MATERIALS AND METHODS

Local Site and Soil Description

The experiment was carried out in Jou (41° 28' 35'' N and 7° 25' 17'' W) at 620 m elevation with undulating topography (5–8% slope). Mean annual precipitation is 1079 mm (778 mm from October to March and 301 mm from April to September) and mean annual temperature 13.3°C.

The chestnut orchard was established on soils derived from quartzophyllites, are quite shallow, and may vary from Haplic Dystric Leptosol to Leptic Dystric Cambisol (FAO, 2006) according to soil depth. The effective soil depth of the orchard varies from 25 to 40 cm, the soils had a loamy texture with abundant coarse fragments at the surface and good drainage. Some physical and chemical properties are indicated in Table 1. The chemical soil data are average values from four composite soil samples, taken from the four quadrants beneath the canopy of eight trees.

The annual precipitation and its distribution around the years of 2006–2007 and 2007–2008 was not a serious constraint. The rainfall in 2006–2007 was 1168 mm (880 mm from October to March and 288 mm from April to September) and in 2007–2008 was 825 mm (432 mm from October to March and 393 mm from April to September); no dry spells occurred in the active growing period. The average minimum temperature during the pollination period (last week of June in 2007 and first week of July in 2008) were 9.2°C and 9.6°C, respectively, while the mean minimum

TABLE 1 Soil physical and chemical properties

Soil depth	0–20 cm	20–40 cm
Gravel, % (v)	35	35
Clay, %	13	13
Texture	Loam	Loam
Organic matter ^a , g kg ⁻¹	26	12
pH (H ₂ O) ^b	4.8	4.6
P ₂ O 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
Base saturation (CEC _{effective}), %	66	32
Hot water soluble B ^e , mg kg ⁻¹	0.53	0.07

^a Oxidation with K₂Cr₂O₇, following 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).

temperatures are 12.2°C in the month of June and 14.1°C in the month of July (means of 30 years, according INMG, 1991).

Experimental Trial and Fertilization

A 15 year-old chestnut orchard, tentatively diagnosed as B deficient by morphological abnormalities and foliar analysis was selected in 2006. The trees more dramatically affected with boron deficiency were not included in this trial. A group of 16 trees of Judia variety, with a mean diameter at breast height (DBH) of 16.7 cm and canopy projection area 19 m², was selected in order to apply the treatment. The 16 trees were classified in two sub-groups (eight trees each) according to the DHB. The B treatment was randomly applied to half the trees in each sub-group (B0 and B1).

A basic fertilization was performed beneath the crown of all trees. In the winter of 2006/2007, dolomitic limestone was used to correct the soil acidity and mineral fertilizers were applied at suitable rates to supply nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), zinc (Zn), copper (Cu), and manganese (Mn). In the beginning of March 2007, sodium tetraborate (Granubor, Borax Europe Limited, Castellón, Spain) with 14.6% B was broadcast on the soil surface under eight trees at a rate of 100 g per tree. Fertilization with the macronutrients was repeated in winter 2007–2008.

Plant Sampling, Fruit Parameters, and Laboratory Procedures

Leaf sampling for foliar analysis was carried out in the beginning of September, and five trees (B0 and B1) were selected at random. Fully developed leaves of fruiting shoots (fourth to seventh from the terminal shoot), light exposed, from the upper part of the middle crown were collected in the four quadrants of the trees and were analyzed for macro- and micronutrients. Leaf samples were dried at 65°C for 48 h and ground to pass through a 1-mm screen. They were digested as described by 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 (Novozamsky et al., 1983) and their concentrations were determined on an autoanalyzer. Boron was measured spectrophotometrically by the azomethine H method (Wolf, 1971).

In the beginning of October 2008, fruit setting was evaluated by picking 30 burs in the middle crown of the four quadrants of the five trees, and the percentage of aborted fruits was determined.

Chestnut fruit was harvested from the ground under five trees each from B0 and B1 in 2007, and under all trees in 2008. For the purpose of evaluating nut production chestnuts which passed through a 24 mm grid were rejected as unmarketable, and the remainder were weighed. Samples of 50 nuts were

used for determination of dry matter, caliber (nuts kg^{-1}), percentage of nuts infested with tortrix (*Cydia splendana* Hübner) and shell cracking.

For the purpose of chemical analysis of the fruit, a sample of shelled chestnut (kernel) of five trees were dried at 65°C for 48 h to determine dry matter and analyzed for neutral detergent fiber (NDF) by the method of Robertson and van Soest (1981); crude protein by the micro-Kjeldahl method ($\text{N} \times 5.3$, according to McCarthy and Meredith, 1988); crude fat by extraction with petroleum ether in a Soxhlet apparatus; starch by enzymatic hydrolysis (Salomonsson et al., 1984). Soluble sugars were determined by the colorimetric method of anthrone after extraction with ethanol, as described by Irigoyen et al. (1992).

The data was analyzed by one-way ANOVA followed by the Duncan multiple range test ($P < 0.05$) with JMP statistical package.

RESULTS AND DISCUSSION

Description of Boron Deficiency

The trees more dramatically affected with boron deficiency showed death of shoot tips; the terminal buds systematically failed, many branches were leafless, flowering did not occur and they never yielded burs. These trees had little growth.

In trees that manifest moderate symptoms of B deficiency the young leaves were small and narrow, sometimes malformed and with irregular shape. Some portions of the branches were leafless and there were shortened internodes. Rosetting arrangement of younger leaves was also seen in some trees. Some burs had a small size, because they failed to set fruit. Many burs displayed undersized or atrophied nuts with empty pericaps (shells). Some burs had premature drop, others did not shed the fruits, which remained on the tree after the normal harvesting period.

Chestnut Production and Nut Quality

The results displayed in Table 2 show that B application significantly increased nut production. The increase was 75% in 2007 and over four times higher in 2008, respectively, 4 and 17 kg tree^{-1} in B0 and B1. The rise in nut production might be ascribed to the increase in fruit setting. Indeed, as shown for 2007–2008, the percentage of fruits that failed was 81% in the control and only 20% when boron was applied. It is worth noting that in both years the temperature was very low during the flowering period (June–July), which most likely affected fruit setting too. No significant differences were observed in such quality parameters as caliber, dry matter, and shell cracking. Nut size was somewhat better in the B0 ($P < 0.065$) in 2008, probably due to the high percentage of fruit abortion (81%), which may have propitiated the

TABLE 2 Chestnut production and some quality parameters

	Fruit setting, %	Yield, kg tree ⁻¹	Caliber, nuts kg ⁻¹	Dry matter, %	Tortrix infestation, %	Shell cracking, %
2007						
No boron	—	8.1a	63a	57a	0.0	0.0
With boron	—	14.1b	64a	57a	0.0	0.0
2008						
No boron	19a	4.1a	74a	58a	5.3a	15.7a
With boron	80b	16.9b	86a	59a	1.5b	9.3a

Means in the same column and year with a different letter are significantly different ($P < 0.001$) by the Duncan multiple range test.

nut enlargement. Tortrix infestation and shell cracking were absent in 2007. In 2008 a significant difference was observed in tortrix infestation, which was lower when boron was applied (Table 2). The degree of infestation led to different chestnut quality classes (B1 – 1.5% corresponds to class ‘extra’ and B0 – 5.3% corresponds to class 2), according to Standard CEE-ONU FFV-39 (United Nations, 1996).

As far as the chemical composition of the chestnut kernel is concerned, the results recorded in Table 3 show that no differences were observed in any of the parameters studied in the first year. Compared with the standard quality parameters indicated by Vasconcelos et al. (2008) for the present variety in NE Portugal our results show lower starch content and higher values of soluble sugars and total fiber (NDF). Results obtained for the other chemical components were similar.

Soil and Foliar Analysis

As shown in Table 1 the soil boron level of 0.53 mg kg⁻¹ cannot be considered too low, according to reference values supplied by Shorrocks (1997) and Sillanpää (1990) for soil B extracted by hot water, which indicate that values >0.5 mg kg⁻¹ of soil are acceptable for most medium textured soils. However, it is worth noting that the low effective soil depth and stoniness, and the high percentage of gravel at both depths, reduce the contact

TABLE 3 Chemical composition of the chestnut kernel

	Dry matter, %	Total fiber NDF, %	Crude protein, %	Crude fat, %	Soluble sugars, %	Starch, %
2007						
No boron	56a	20.3a	5.36a	1.04a	18.9a	52a
With boron	58a	20.4a	5.62a	1.18a	17.1a	56a

Means in the same column with a different letter are significantly different ($P < 0.05$) by the Duncan multiple range test.

TABLE 4 Average foliar nutrient concentrations in chestnut trees of the control (B0) and trees fertilized with boron (B1)

	g kg ⁻¹					mg kg ⁻¹				
	N	P	K	Ca	Mg	Fe	Mn	Zn	Cu	B
2007										
No boron	22.6a	1.9a	7.6a	11a	1.5a	62a	1492a	39a	7a	8a
With boron	23.2a	1.7a	8.5a	10a	1.2a	56a	1248a	35a	9a	61b
2008										
No boron	17.3a	1.3a	6.5a	8a	1.5a	38a	692a	17a	11a	8a
With boron	19.4a	1.5a	7.1a	11a	1.6a	56a	619a	20a	12a	34b

Means in the same column and year with a different letter are significantly different ($P < 0.001$) by the Duncan multiple range test.

between the soil and root surface, which may have contributed to the low boron acquisition by chestnut roots. Thus, the soil boron of 0.53 mg kg⁻¹ is apparently low for the growth of chestnut in this particular case.

Concentrations of nutrients in chestnut leaves are displayed in Table 4. As expected none of the nutrients show significant differences between the treatments, except B concentration, with a significant increase when B was supplied. The application of 14.6 g of B per tree was enough to raise the foliar B concentration from 8 mg kg⁻¹ to 61 mg kg⁻¹ dw in the first year (2007), but in 2008 it dropped to 34 mg kg⁻¹. The foliar B concentration in the control trees is, in fact, very low and has a narrow variation, 6 to 10 mg kg⁻¹. The boron foliar levels reached in B1 are considered adequate both for *Castanea crenata* (Park et al., 1978), and also for several nut and fruit trees (Bergman, 1992; Martin-Prével et al., 1987; Shear and Faust, 1980).

The noticeable drop, almost to half, in the foliar B concentration from 2007 to 2008, suggests that one must be attentive to the need for B application in the following years, since the level of 34 mg kg⁻¹ recorded is becoming marginal. Its redistribution in the tree tissues could have occurred. Moreover, the great mobility of boron in the soil and its likely leaching to deeper horizons was propitiated by the high gravel content of the soil, in addition to the siliceous nature of the parent material. Vale et al. (1999) also draw the attention to the rapid drop to deficient levels of foliar B in needles of *Pinus pinea* in the same type of soil nearby, after two years of B fertilization.

CONCLUSIONS

In conclusion, boron had a clear positive effect on chestnut production, but there was no obvious effect on nut quality. The available data for the chemical composition of the kernel does not show clear differences that could be ascribed to boron application, in the first year of observations. These results will be confirmed by analytical determinations of samples from 2008.

The large drop in the B concentration in the second year suggests the need for monitoring the nutrient status of chestnut orchards or more frequent B fertilization.

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