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Title:
Effects of boron application on yield, foliar B concentration, and efficiency of soil B extracting solutions with boron application in a Xanthic Ferralsol cultivated with banana in Central Amazon

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Abstract:
Boron is known to play an important role for the structure of cell wall, membranes and the integrity and functions of membranes in plants. In soil, the hot water B extraction method has been extensively used for evaluation of soil B status. However, difficulties with this method result in low accuracy and precision of extraction of available boron (B) in soil. The objective of this study was to evaluate the yield, foliar concentration, and efficiency of B extracting solutions and the effect of B fertilization on B uptake in banana leaves and fruits, subgroup Cavendish (AAA), cultivated in a Xanthic Ferralsol (dystrophic Yellow Latosol), in the Amazonas State, Brazil. The experimental design was a completely randomized split plot, comprising four boron rates (0, 4, 8, and 12 kg ha-1), and two harvest cycles (sub-treatments), with four replicates. Available boron was determined with seven extractant solutions: Mehlich 1, Mehlich 3, hot water, HCl 0.05 mol L-1, HCl 0.1 mol L-1, HCl 5.0 mol L-1 and KCl 1.0 mol L-1. The application of B fertilizer increased the yield and B concentration in leaves and fruits. Hot water and KCl 1.0 mol L-1 were the most efficient extracting solutions for the determination of available B in soil. The application of 3.4 kg B ha-1 in first cycle and 1.3 kg B ha-1 in second cycle guarantees an adequate nutritional status in banana plants

Introduction

Banana plantations are an important subsistence agricultural activity in Amazonas State, Brazil (Moreira and Fageria 2009). The cultivation was initially carried out in the floodplain sedimentary fertile soils. After the high incidence of black sigatoka (*Mycosphaerella fijiensis* Morelet), crops moved progressively to the upland, where soils are very poor in most essential nutrients (Moreira and Fageria 2008). This caused, along with the mentioned diseases, low banana crop productivity.

Boron plays very important role in plant growth and development. It is reported to be located in the cell wall and associated with pectin (Loué 1993; Moraes et al. 2002). Boron deficiency was also reported to increase membrane permeability to K^+. However, its primary function in plants has not been clarified yet (Iikura et al. 1997). Boron deficiency causes severe growth inhibition of the banana plant, with negative effect on the pulp consistency of the fruits (Moreira and Almeida 2005).

Most acid soils have low B concentration, which hampers efficiency of B extraction. The testing of a suitable B extraction method for Brazilian acid soils is important to improve evaluation of the nutritional status of crop plants in these soils (Ferreira et al. 2001). For determination of available B in soil, the method currently considered as standard is the hot water method proposed by Berger and Truog (1939), or modified versions in which $BaCl_2$ 0.01 mol L^{-1} or $CaCl_2$ 0.01 mol L^{-1} solutions replace hot water as extractant (Abreu et al. 1994; Ferreira et al. 2001); extraction is carried out in plastic bags and samples are heated in microwave oven or under reflux.

Despite these modifications, some drawbacks still persist. The microwave method demands specific equipment, whereas the difficulty of the reflux methods lies on the need of a precise temperature control, in the procedures of heating and cooling of the soil extractant solutions. Furthermore, the time consumption associated with cleaning glassware and the need of glassware free of borosilicates increase difficulties. In both procedures, the number of samples per batch is restricted, which increases cost significantly; in addition, only a single element is extracted (Sims and Johnson 1991).

Other extractant methods of costing less and easy to handle have been frequently proposed and compared to the hot water under reflux procedure (Bataglia and Raij 1990) or to the heating with microwaves (Ferreira et al. 2002), such as Mehlich 1, Mehlich 3, HCl 0.05 mol L^{-1}, HCl 0.1 mol L^{-1}, $CaCl_2$ 0.01 mol L^{-1} and $CaCl_2$ 0.05 mol L^{-1} solutions. Despite some advantages, the acids extractants can to cause high Fe extraction affecting the determination of the B available.

The objective of this study was to evaluate the effects of increasing rates of B fertilization on its uptake by banana plants and the efficiency of seven B extractants solutions for determination of available B in a Xanthic Ferralsol (40.9% of Brazilian Amazon with 2,097,160 km^2 – Moreira and Fageria 2008).

Materials and Methods

The study was carried in a Xanthic Ferralsol (dystrophic Yellow Latosol) (FAO 1990), with 719 g ha^{-1} clayey texture, bulk density of 0.88 Mg m^{-3}, and the following chemical characteristics: pH (water) = 4.27; organic matter (OM) = 46.9 g kg^{-1}; phosphorus (P) (Mehlich 1) = 2.9 mg kg^{-1}; P (resin) = 9.8 mg kg^{-1}; P (Mehlich 3) = 1.8 mg kg^{-1}; potassium (K) = 47.7 mg kg^{-1}; calcium (Ca) = 2.0 $mmol_c$ kg^{-1}; magnesium (Mg) = 1.2 $mmol_c$ kg^{-1}; aluminum (Al) = 14.5 $mmol_c$ kg^{-1}; acidity (H + Al) = 84.0 $mmol_c$ kg^{-1}; B (hot water) = 0.31 mg kg^{-1}; copper (Cu) = 0.29 mg kg^{-1} (Mehlich 1); iron (Fe) = 333.0 mg kg^{-1} (Mehlich 1);

manganese (Mn) = 5.15 mg kg^{-1} (Mehlich 1) and zinc (Zn) = 0.68 mg kg^{-1} (Mehlich 1). The experimental site is located at the Embrapa (Empresa Brasileira de Pesquisa Agropecuária) in coordinates 3°8' S and 59°52' W, municipality of Manaus, Amazonas State, Brazil.

Natural vegetation in the region is a tropical rainforest. The predominant climate is humid tropical, classified as Afi by the Köppen system, with relatively abundant rainfall throughout the year (mean 2250 mm). The amount of rainfall in the driest months (July to September) is always above 60 mm, and the wettest months are February to April. Average temperature is about 26ºC (Vieira and Santos 1987).

The study site was first cleared from primary forest in 1978, using heavy machinery for clearing and the removal of tree stumps and establishment of rubber, which abandoned. The developing secondary forest was cleared again in January 2002 with heavy machinery.

The experimental design was a completely randomized split plot with four replicates each containing five plants. Treatments consisted of 0, 4, 8 and 12 kg ha^{-1} of B (boric acid, 18% of B) per cycle, and two harvest cycles (sub-treatments). Equal B rates of each treatment were applied in the planting hole (1st cycle) and broadcasted in a semicircle, for the second cycle, after the harvest of the first bunch around the daughter plant. Each plot contained five measurable plant clumps separated by two guard clumps in the row.

Holes (40 cm × 40 cm × 60 cm) were prepared thirty days before planting and refilled with the topsoil layer plus five liters of chicken manure and 400 g of dolomitic limestone (effective calcium carbonate = 78%). At planting, 60 g of P_2O_5 (simple superphosphate – 20% of P_2O_5), 10 g of manganese sulfate (26% of Mn), 20 g of iron sulfate (19% of Fe), 5 g of copper sulfate (13% of Cu), and 30 g of zinc sulfate (20% of Zn) were applied. Spacing adopted was 3 m between rows and 2 m between plants in the rows (1667 plants per hectare). Clones from tissue culture of the cultivar "Nanicão 2001" (triploid AAA of the Cavendish subgroup) were used for the experiment. Plants were managed so that only mothers, daughters and granddaughters were left in the clusters.

Topdressing fertilizations consisted of urea (44% of N) and potassium chloride (58% of K_2O), distributed in four applications: in the second, the fourth, the seventh and the tenth month after planting (Pereira et al. 2002). The first three plots were demarcated around the plant and the others in a semicircle beside the daughter plant.

In the fourth month after planting, 100 g of magnesium sulfate (9% of Mg), 20 g of copper sulfate (13% of Cu), 20 g of iron sulfate (19% of Fe), 10 g of manganese sulfate (26% of Mn) and 30 g of zinc sulfate (20% of Zn) were supplied in broadcast application (Pereira et al., 2002).

At the early flowering stage and at harvesting, a sample of the medial third of the leaf below the apex (leaf blade only) was collected from each treatment; two bananas of the second hand were also collected. Boron concentration in leaves and in fruits (pulp plus rind) was determined according to Malavolta et al. (1997).

Soil samples were collected together with leaf and fruit sampling, at 0 to 20 cm soil depth. Soil samples were collected in two spots (in and between plant rows – ten samples per plot), at a 30 cm distance from plants, and were afterwards homogenized. Analysis of available B in soil was performed using the following extractants: hot water (Abreu et al. 1994), Mehlich 1 (Mehlich 1978), Mehlich 3 (Mehlich 1984), hydrochloric acid (HCl) 0.05 mol L^{-1} (Ponnamperuma et al. 1981), HCl 0.1 mol L^{-1} (Ponnamperuma et al. 1981), HCl 5.0 mol L^{-1} and KCl 1.0 mol L^{-1} (Moreira and Castro 2006). All extracting solutions were filtered through a double layer of low speed filter paper. Determination of B was performed using a spectrophotometer, with the addition of 1.0 mL of buffer solution and 1.0 mL of azomethine-H solution to 4.0 mL of the extractant solution (Abreu et al. 2001), at 420 nm wavelength.

Results were compared by analysis of variance (ANOVA - F test at $p \leq 0.05$) and submitted to regression analysis at 5% and 10% significance and Pearson's relationship, according to procedures described by Pimentel Gomes and Garcia (2002).

Results and Discussion

Boron application increased bunch weight. The yield response in the two harvest cycles was significantly different: 1st cycle, $\hat{y} = 25.76 + 0.30x - 0.052 x^2$ and 2nd cycle, $\hat{y} = 30.83 + 0.32x - 0.019x^2$; $p \leq 0.10$). The maximum yields in the two cycles corresponded to 26.2 Mg ha^{-1} and 32.2 Mg ha^{-1}. As evident from the two equations the yield response to B fertilization was relatively small which may be due to medium levels of B in the soil (0.31 mg kg^{-1}) of experimental area before planting (Alvarez Venegas et al. 1999). Boron levels usually found in Oxisols, Espodosols and Neosols, as determined with the hot water extraction method, are mainly within the range considered as high (B > 0.5 mg kg^{-1}) (Malavolta 1987).

Yield of control plots was higher in the second cycle and this was probably due to the natural yield increase of around 30% from the first to the second cycle and to increased amount of plant-available B in the soil, derived from increased by mineralization of organic matter and by nutrient cycling as a result of decomposition of pseudostems, clusters of terminal bracts and leaves.

Boron application rates caused significantly and linearly increased B foliar concentrations (Figure 1). A decline of 57% in leaf B concentration from the first to the second cycle was observed in control plants. Comparison of B concentrations found in leaves with those considered adequate for the genus *Musa* shows that in the first cycle only the two lower B rates produced foliar B concentrations within the adequate range of 10 to 25 mg kg^{-1} (Malavolta et al. 1997). In the other treatments, except the control in the second cycle, foliar boron concentrations were higher. With 12 kg ha^{-1}, leaf B concentration was about 139% (first cycle) and 1254% (second cycle) higher than in the control (Figure 1). The results showed that the application of 3.4 kg B ha^{-1} in first cycle and 1.3 kg B ha^{-1} in second cycle, respectively, guarantee an adequate nutritional status in banana plants (25 mg B kg^{-1} in medial third of the leaf below the apex; Malavolta et al., 1997).

Despite of B application, the significant increase of the leaf concentration (Figure 1) was also due to cycling of B contained in crop residues. With the well distributed rainfall, these residues were rapidly broken down and mineralized. The lower B concentration found in control and in fruits in the second harvest cycle can be due to a dilution effect (Marschner 1995) and/or decreased soil B level.

Absence of visual symptoms of B phytotoxicity, even at the rate of 12 kg ha^{-1}, and the limited yield response to B application indicate there is no narrow limit in bananas between B deficiency and toxicity. Reuter and Robinson (1988) described that the limit of B toxicity in banana plants is 300 mg kg^{-1}. Salvador et al. (2003), in guava seedlings, and Chapman et al. (1997), in green-house grown rice, lentil and pea, also found no deleterious effects on dry matter production of increasing boron application rates. Another factor is that the B level considered adequate for most crops is highly variable and the demand for this nutrient is ascribed to differences in chemical composition of cell walls of different species and genotypes (Marschner 1995).

Boron concentration in fruits increased significantly with B application rate. The estimated B accumulation was 486 g ha^{-1} and 547 g ha^{-1} in the first and in the second cycle, respectively, with application of 12 kg B ha^{-1} cycle^{-1} (Figure 1). Results showed that export of B was low, a higher proportion being retained in other plant organs or in soil. In the present study, weights of fruits, leaves, terminal bracts and pseudostems, as an average of the two

cycles, corresponded to respectively 27.6%, 9.9%, 2.2% and 60.3% of the total weight of the fresh matter of the banana plant.

The relationship between soil content of available B, obtained with the seven extractants, and foliar B concentration shows that hot water and KCl 1.0 mol L^{-1} extractant solutions gave significant linear coefficients (Figure 2). The adjusted second degree equations of the Mehlich 1, Mehlich 3, HCl 0.05 mol L^{-1} and HCl 0.1 mol L^{-1} extractant solutions were also significant, with coefficients of 0.85, 0.91, 0.77 and 0.73, respectively. The HCl 5.0 mol L^{-1} extractant solution did not show significant correlation with foliar B (Figure 2), probably, due to high Fe level in soil (333 mg Fe dm^{-3}) interfering the B determination with azomethine-H.

Results obtained with the acid solutions, except HCl 5.0 mol L^{-1}, were correlated with the available B extracted with hot water or with KCl 1.0 mol L^{-1} (Table 1). This result partially corroborates Raij and Bataglia (1991) about the efficiency of B extracting solution in predicting available B in soil.

The extractants KCl 1.0 mol L^{-1}, Mehlich 1, Mehlich 3, HCl 0.05 mol L^{-1}, HCl 0.1 mol L^{-1} and HCl 5.0 mol L^{-1} showed a higher recovery capacity than hot water (Figure 2). These results confirm findings of Paula (1995) and Ferreira et al. (2001), who compared the extractive capacity of the extractants Mehlich 1, CaCl$_2$ 0.05 mol L^{-1} and hot water. Presence of the chloride ion in these extractants may have resulted in a higher capacity for recovering of the borate anion adsorbed by the positive changes of the soil colloids (Jin et al. 1987).

One of the advantages using KCl 1.0 mol L^{-1} extractant solution over hot water for predicting available B is that it can be introduced in routine procedures of many laboratories, without the need of an additional extractant. Furthermore it can also be used for determination of exchangeable calcium, magnesium and aluminum, while the hot water extractant can be used only for B determination. Additionally, this method has a wide extraction range. For instance, while the highest record of extracted B with hot water was 1.48 mg kg^{-1}, this value was 3.37 mg kg^{-1} with KCl 1.0 mol L^{-1}. This improves the availability ranges and the interpretation of the results classified as low, medium or high (Figure 2), but more studies are necessary with others type of soils with different chemical and physical characteristics.

Conclusions

Banana plantations are an important subsistence agricultural activity in Amazonian, including the Amazonas State, Brazil. The results showed yields increased with B fertilization and that the application of 3.4 kg B ha^{-1} in first cycle and 1.3 kg B ha^{-1} in second cycle, respectively, in Xanthic Ferralsol (2,097,160 km^2 – 40.9% of Brazilian Amazon soil), guarantees an adequate boron status in banana plants (25 mg kg^{-1}). The application of 12 kg B ha^{-1} cycle^{-1} increased B concentration in leaves and in fruits of banana plants. In the edaphoclimatic conditions studied, KCl 1.0 mol L^{-1} and hot water the extractants solutions were the most efficient for determination of available B in soil than Mehlich 1, Mehlich 3, HCl 0.05 mol L^{-1} and HCl 0.1 mol L^{-1} extractant solutions.

Acknowledgments

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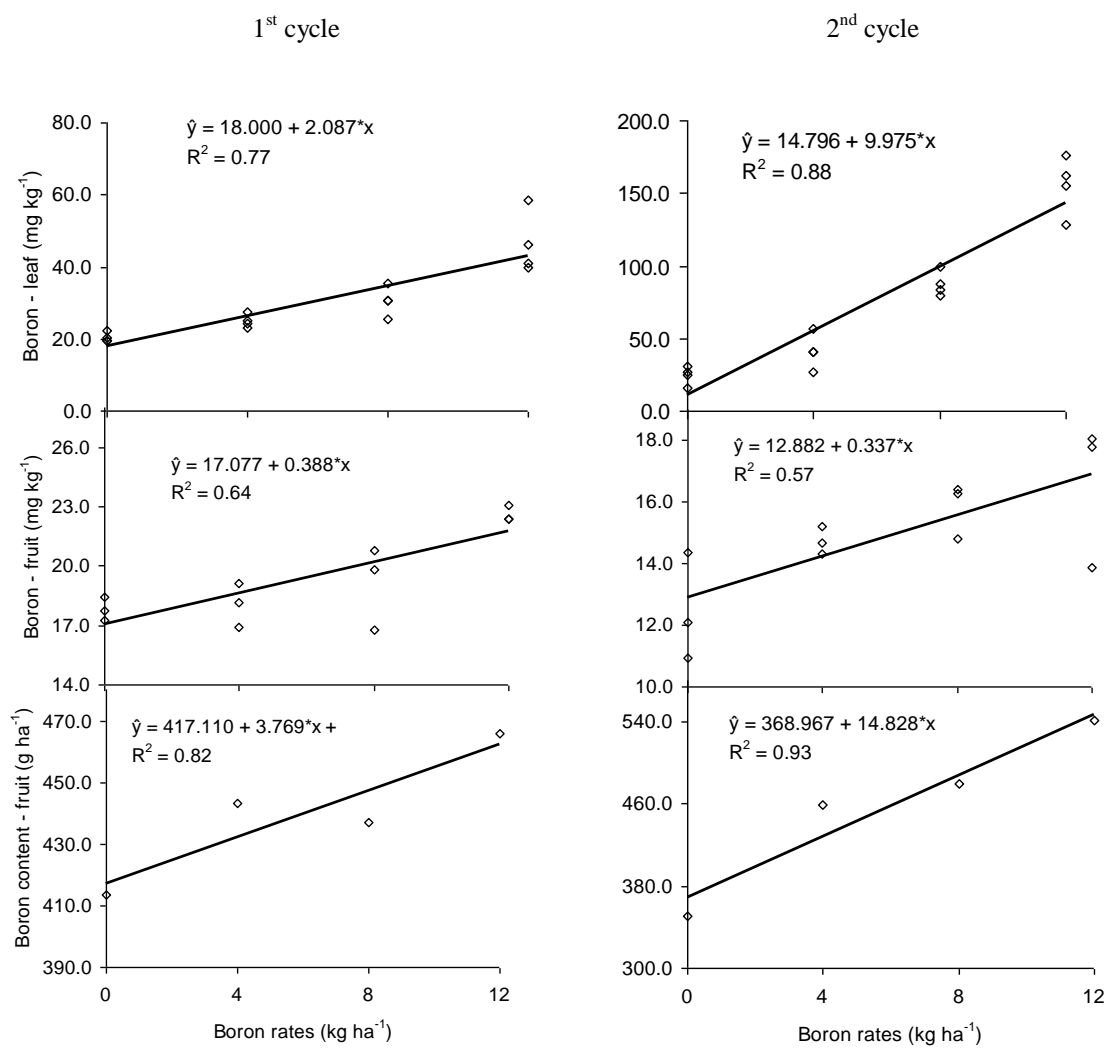
Figure 1: Regression between boron application rates and boron concentration in leaves and in fruits of two yield cycles. Significant at 5% level of probability.
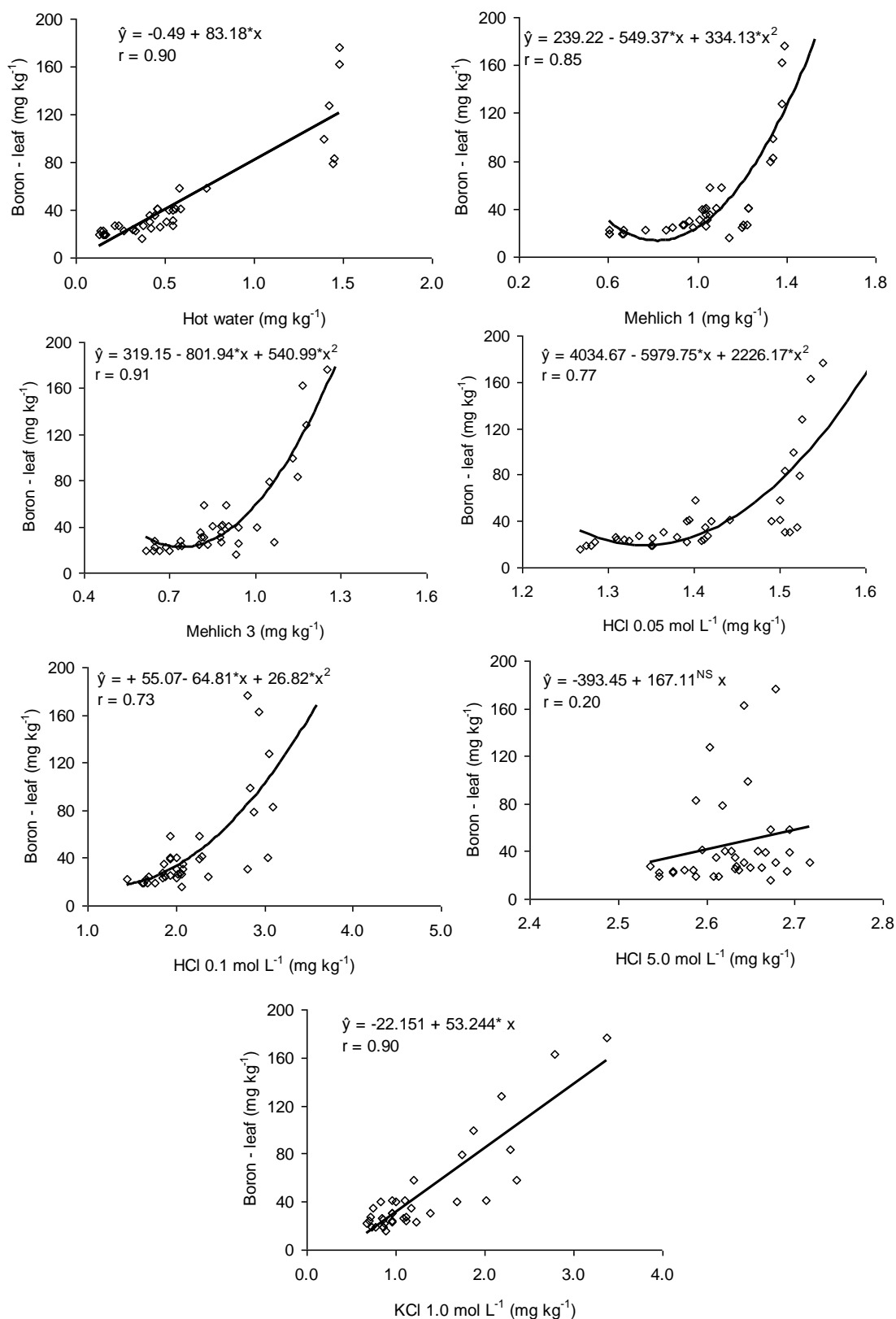
Figure 2: Relationship between B leaf concentration and of B extracted with hot water, Mehlich 1, Mehlich 3, HCl 0.05 mol L^{-1}, HCl 0.1 mol L^{-1}, HCl 5.0 mol L^{-1} and KCl 1.0 mol L^{-1}, recorded in the two yield cycles. *Significant at 5% level of probability. ^{NS}Non-significant.

Table 1: Coefficients of simple linear regression between extraction methods of B from soil[1].

	Hot water	Mehlich 1	Mehlich 3	KCl 1.0 mol L^{-1}	HCl 0.05 mol L^{-1}	HCl 0.1 mol L^{-1}
Hot water	-					
Mehlich 1	0.81*	-				
Mehlich 3	0.89*	0.88*	-			
KCl 1.0 mol L^{-1}	0.83*	0.61*	0.67*	-		
HCl 0.05 mol L^{-1}	0.65*	0.52^{NS}	0.60*	0.78*	-	
HCl 0.1 mol L^{-1}	0.83*	0.82*	0.77*	0.69*	0.64*	-
HCl 5.0 mol L^{-1}	0.26^{NS}	0.39^{NS}	0.38^{NS}	0.33^{NS}	0.37^{NS}	0.27^{NS}

* Significant at 5% level of probability; ^{NS}non-significant.