

Evaluation of the NH_4HCO_3 -DTPA Soil Test for Assessing Boron Availability to Wheat

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ABSTRACT

The NH_4HCO_3 -DTPA (AB-DTPA), 1 M NH_4HCO_3 , 0.005 M DTPA, pH=7.6, was proposed as a multi-element extractant, for evaluating macro and micronutrients availability to plants. AB-DTPA was also evaluated as a soil test, for assessing boron availability and toxicity to alfalfa. In a pot experiment, ten soils of Northern Greece were used to assess AB-DTPA as an extractant of available boron to wheat (*Triticum aestivum* L., cv. Yecora), in comparison with hot water and saturation extract. Boron (B) was added as borax ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$) to the ten soils, at rates equal to 0, 3, and 5 mg B kg⁻¹. Wheat was grown in pots containing the boron amended soils to the stage of tillering, and dry aboveground biomass, B concentration and B uptake by wheat were determined. AB-DTPA extractable B was significantly greater than saturation extract and similar to hot water at each B application rate, and was correlated significantly with hot water ($r=0.84$), or with saturation extract ($r=0.48$). Extractable boron by all extractants, boron concentration in wheat and boron uptake were significantly affected by the soil x B application rate interaction. In assessing B availability to wheat using AB-DTPA as a soil

test, CEC should be included in the regression equation for B concentration, or pH for B uptake. However, the corresponding adjusted coefficients of determination for B concentration (adjusted $R^2=0.46$) and B uptake (adjusted $R^2=0.48$) were similar or lower to those of hot water (adjusted $R^2=0.45$ and 0.60 , respectively) and the saturation extract (adjusted $R^2=0.70$ and 0.49 , respectively), when the latter two soil tests were used in the regression equations without the inclusion of any soil property.

INTRODUCTION

It is known that plants respond primarily to soil solution boron and according to Gupta et al. (1985), B in the soil solution under field conditions is the ideal soil test in assessing B deficiency, sufficiency or toxicity. However, extracting soil solution from soil, at a moisture content within the range available to plants, is not always feasible. Also the amounts of boron in the saturation extract, which quite frequently is considered an approximation of B in the soil solution, are usually low and consequently the error of the analytical determination might be large. Despite the above, Hatcher et al. (1959) and Aitken and McCallum (1988) successfully used soil solution boron methods for assessing plant available B.

In addition to water soluble B, adsorbed B also need to be included in a successful test in assessing B availability. Thus hot water extractable B (Berger and Truog, 1939) has been proposed as B availability index. Hot water extractable B was found to be a useful index of plant response to B fertilization in deficient soils, and also of the B content of plants (Cartwright et al., 1983).

Other extractants for B, such as, NH_4OAc , mannitol (Aitken et al., 1987), mannitol- CaCl_2 (Cartwright et al., 1983), CH_3COOH , dilute concentrations of HCl (Renan and Gupta, 1991), multi-element extractants such as NH_4HCO_3 -DTPA (Gestring and Soltanpour, 1984, 1987) and the Mehlich 1 and 3 solutions (Shuman et al., 1992), were used, for assessing plant available B. At the present, hot water extractable B appears to be the best index for soil available B (Offiah and Axley, 1993).

The advantages of using a multi-element extractant, such as the NH_4HCO_3 -DTPA, as a soil boron test are obvious, as materials and time are saved. The NH_4HCO_3 -DTPA (AB-DTPA), $1\text{ M NH}_4\text{HCO}_3$ and 0.005 M DTPA , adjusted to $\text{pH}=7.6$, has been proposed by Soltanpour and Schwab (1977) for the simultaneous extraction of available macronutrients phosphorus (P) and potassium (K) and micronutrients zinc (Zn), iron (Fe), copper (Cu), and manganese (Mn). The AB-DTPA test was used to screen soils contaminated with lead (Pb), cadmium (Cd), and Zn (Boon and Soltanpour, 1991), to assess molybdenum (Mo) availability in soils and mine spoils (Wang et al., 1994) and to monitor selected nutrients and heavy metals in a calcareous soil amended with solid waste (Hanlon et al., 1996). Also, Gestring and Soltanpour (1984, 1987) and Al-Mustafa et al. (1993) used

TABLE 1. Some physicochemical characteristics of soils used in study.

Soil	Sand Silt Clay			pH	EC _s	O.M.	CaCO ₃ Fe ₂ O ₃ Al ₂ O ₃			CEC
	g kg ⁻¹						dS m ⁻¹	g kg ⁻¹		
1	860	100	40	6.3	4.0	29	0	11.9	1.08	17.6
2	480	400	120	7.5	1.9	7	15	7.4	0.77	12.4
3	680	280	40	7.7	1.0	14	34	8.7	0.82	21.8
4	610	190	200	7.5	0.9	33	0	39.9	0.97	18.1
5	470	390	140	5.8	0.6	16	0	5.8	0.75	4.8
6	310	450	240	6.3	1.6	7	0	6.8	0.88	9.8
7	180	585	235	6.1	1.2	16	0	10.9	1.04	8.6
8	625	240	135	7.8	0.5	2	6	6.2	0.56	4.2
9	235	550	215	7.4	1.1	18	12	11.1	1.19	14.9
10	270	375	355	7.8	0.5	11	61	15.9	1.55	17.4

AB-DTPA to assess boron availability to alfalfa. The objective of this study was to test AB-DTPA extractable B for assessing B availability to wheat, using ten soils of Northern Greece.

MATERIALS AND METHODS

Soils

Ten soil samples (0-25 cm) were collected, from cultivated fields in Northern Greece, air dried, ground, sieved through a 5-mm sieve and a quantity smaller than 2 mm, was used for the determination of selected physicochemical properties. All analyses were run in duplicate and mean values are reported in Table 1. The pH was determined in a 1:2 soil to water suspension, electrical conductivity in the saturation extract (EC_s), organic matter (O.M.) by wet oxidation (Walkley and Black, 1934), CaCO₃ volumetrically using a calcimeter (Allison and Moodie, 1965) and particle-size analysis by the pipet method (Gee and Bauder, 1986). Cation exchange capacity (CEC) was determined, using CH₃COONa, 1 N, pH=8.2, as saturating solution and CH₃COONH₄, 1 N, pH=7, as extracting solution (Chapman, 1965). Free Fe and aluminum (Al) oxides were determined using dithionite-citrate as extracting solution (Olson and Ellis, 1982).

Boron, as borax (Na₂B₄O₇·10H₂O), was added to subsamples of the ten soils at rates equal to 0, 3, and 5 mg B kg⁻¹. The subsamples were wetted in plastic bags to approximately field capacity and equilibrated for 15 days, air dried and used to the pot experiment. A quantity of soil subsample, smaller than 2 mm, was used for the determination of B in the saturation extract (B_{SE}), hot water extractable B

TABLE 2. Boron concentration extractable by the three extractants.

Boron added mg kg ⁻¹	Soil boron† mg kg ⁻¹		
	Saturation extract	Hot water	AB-DTPA
0	0.57 - 1.04	0.32 - 1.41	0.43 - 2.71
3	0.72 - 1.33	1.48 - 3.68	0.90 - 3.92
5	0.92 - 2.23	2.00 - 3.69	2.02 - 4.00

†All figures represent the range over the ten soils.

(B_{HW}) according to Berger and Truog (1939), and AB-DTPA (1 M NH_4HCO_3 , 0.005 M DTPA, pH=7.6) extractable B ($B_{AB-DTPA}$), according to Soltanpour and Schwab (1977). Analytical boron determination was performed by the azomethine-H method (John et al., 1975). All analyses were run in duplicate and the ranges of the average values, over the ten soils, are reported in Table 2.

Greenhouse Pot Experiment

Two kg of soil subsamples were placed in 2-L plastic pots, contained in plastic dishes. Each pot was sown with 20 seeds of winter wheat (*Triticum aestivum* L., cv. Yecora). The treatments, ten soils x three levels of B addition (0, 3, and 5 mg B kg⁻¹), were replicated three times and all pots were placed on benches in the greenhouse, in a completely randomized design. Randomization was repeated every 15 days. Seven days after seeding, each pot was fertilized with the recommended doses of N, P, and K and after establishment, the plants were thinned out to ten. Plants were grown under natural lighting conditions in the greenhouse at 22±3°C. Aboveground biomass from each pot was harvested at the stage of tillering, dried at 65°C, weighed, and ground to pass a 0.2-mm sieve. Duplicate 0.5 g subsamples were ashed at 550°C for 6 hrs, the ash was dissolved in 10 mL of 0.1 N HCl, filtered, and a portion of the filtrate was used for boron determination, employing the azomethine-H method. Tissue concentration of B, biomass dry weight, and soil weight per pot were used to calculate plant uptake of B per kg of soil. Ranges of the mean values of wheat biomass, B concentration in wheat and B uptake, over the ten soils, are reported in Table 3.

Statistical Analysis

Two types of analysis were performed. First, two-way analysis of variance (ANOVA) was conducted to evaluate effects of soil type and boron application rate to the three soil B tests and to the plant parameters. Also, at each B application

TABLE 3. Boron concentration, boron uptake, and aboveground biomass of wheat, grown in soils after boron additions.

Boron added mg kg ⁻¹	Boron concentration† mg kg ⁻¹	Boron uptake† mg B kg ⁻¹ of soil	Wheat biomass† g pot ⁻¹
0	9.5 - 22.4	0.08 - 0.23	8.9 - 16.4
3	22.7 - 42.5	0.16 - 0.35	9.0 - 16.5
5	23.5 - 64.7	0.22 - 0.52	6.6 - 16.7

†Figures represent the range over the ten soils.

rate two-way analysis of variance was performed to evaluate differences between the amounts of B extracted with the three extractants. Appropriate comparisons between means were performed, using the LSD test, $P=0.05$.

Second, simple and multiple linear regression analysis were used to relate plant response, expressed by biomass, B concentration and boron uptake by wheat, to soil B, determined by the three extractants. The multiple linear regression analysis was conducted utilizing soil B and soil characteristics that affect B availability (Goldberg, 1993), such as pH, CEC, and content of organic matter, clay, Fe_2O_3 , and Al_2O_3 , as the independent variables and the parameters of plant response as the dependent variable. The best subsets utilizing these variables were determined by Mallows' C_p statistic, which is a selection criterion among all possible subsets (Draper and Smith, 1981). The C_p statistic has the form

$$C_p = (RSS_p / s^2) + 2p - n$$

where RSS_p is the residual sum of squares of the subset model containing p parameters, p is the number of parameters in the subset model, s^2 is the residual mean square of the full model and n is the number of cases. The "best" subset is associated with a low C_p value about equal to p (Draper and Smith, 1981).

RESULTS AND DISCUSSION

Extractable boron by the three extractants (saturation extract, hot water, AB-DTPA) was significantly affected by the soil x B application rate interaction, which is in agreement with the findings of Al-Mustafa et al. (1993). Extractable B levels were increased, in many cases significantly, as B application rate increased, regardless of the soil extractant. In agreement with the findings of Gestring and Soltanpour (1984, 1987), for similar boron application rates, the amounts of B_{SE} were significantly smaller in comparison to the other two extracts. The amounts of B_{HW} and $B_{AB-DTPA}$ were similar, in most cases. The amounts of extractable boron with the three extractants were correlated significantly with each other (Table

TABLE 4. Correlation coefficients (r) for soil boron, as determined by the three extractants.

	Saturation extract	Hot water
Hot water	0.58***	
AB-DTPA	0.48**	0.84***

** , ***Significance at the $P \leq 0.01$ and $P \leq 0.001$, respectively.

4). The high correlation observed between the B_{HW} and $B_{AB-DTPA}$ ($r=0.84$) is an indication that hot water and AB-DTPA extract boron from the same pools.

Aboveground biomass of wheat was significantly affected only by the soil type and not by the boron application rate, indicating that wheat biomass yield was affected by productivity factors other than the soil boron. Self (1993) reported that a concentration range of 1-5 mg kg⁻¹ of hot water extractable boron is considered sufficient for normal growth of plants. So, It appears that the ten soils provided the wheat plants with sufficient amounts of boron, even in the case of no boron added (Table 2). Also, If a boron concentration range in wheat tissue of 2-10 mg kg⁻¹ is considered sufficient (Gupta, 1993), the observed boron levels were more than sufficient in all treatments (Table 3). Zada and Afzal (1997) reported that boron application, at rates similar to those used in this study, increased all the yield components and grain yield of wheat plants, but the soil used was low in boron.

Boron concentration in wheat and boron uptake were significantly affected by the soil x B application rate interaction. The adjusted coefficients of determination (adjusted R^2) for the simple regressions of these two plant parameters vs soil boron were significant (Table 5), but for the $B_{AB-DTPA}$ had low values, lower than those reported by Gestring and Soltanpour (1984, 1987). The inclusion of the aforementioned soil properties to the equations did not improve the regressions including B_{SE} as soil boron, but improved those including B_{HW} or $B_{AB-DTPA}$ and the best subsets determined by Mallow's C_p statistic are given in Table 6. Again the adjusted R^2 for the best subsets, included $B_{AB-DTPA}$ as an independent variable, had the lowest values. Gestring and Soltanpour (1984) reported that the addition of pH and clay and organic matter contents of the soils, which they studied, improved the regressions of boron concentration in alfalfa vs soil boron. In agreement with the findings of Gestring and Soltanpour (1984), addition of soil pH improved the regressions of boron uptake by wheat vs soil boron. Also CEC was the soil variable which was included to the multiple linear regression equations for the boron concentration in wheat vs soil boron. Probably because CEC is a soil characteristic dependent on the soil physical and chemical properties, such as texture, clay

TABLE 5. Adjusted coefficients of determination (adjusted R^2), for correlation of boron concentration in wheat or boron uptake with soil boron, as determined by the three extractants.

Extractant	Boron concentration	Boron uptake
Saturation extract	0.70***	0.49***
Hot water	0.45***	0.60***
AB-DTPA	0.27**	0.46***

** , ***Significance at the $P \leq 0.01$ and $P \leq 0.001$, respectively.

mineralogy and organic matter content, which are known that control plant available boron (Goldberg, 1993). Finally It should be noted that even with the addition of soil variables to the regression equations for the AB-DTPA soil test, the adjusted R^2 were similar or lower than the adjusted R^2 for the hot water or the saturation extract soil test, when the latter two soil tests were used in the regression equations without the inclusion of any soil property (Tables 5 and 6).

CONCLUSIONS

Amounts of extractable boron with AB-DTPA and with hot water were similar for the soils studied. AB-DTPA extractable boron was correlated significantly with hot water ($r=0.84$), or with saturation extract ($r=0.48$). Aboveground biomass of wheat was affected significantly only by the soil type, while boron concentration in wheat and boron uptake were affected significantly by the soil x B application

TABLE 6. Best subsets determined by the Mallows' C_p statistic and their corresponding adjusted R^2 , for predicting boron concentration in wheat or boron uptake.

Extractant	Boron concentration		Boron uptake	
	Best subset	Adjusted R^2	Best subset	Adjusted R^2
Hot water	$19.9 + 13.0a^\dagger - 1.3c$	0.68***	$0.42 + 0.10a - 0.05b$	0.71***
AB-DTPA	$19.6 + 10.3a - 1.2c$	0.46***	$0.27 + 0.08a - 0.03b$	0.48***

$^\dagger a$ = soil boron determined by the corresponding extractant (mg kg^{-1}), b = pH, c = CEC ($\text{cmole}_c \text{ kg}^{-1}$).

***Significance at $P \leq 0.001$.

rate interaction. In assessing boron availability using AB-DTPA soil test, CEC should be included in the regression equation for boron concentration in wheat, or pH for boron uptake. But even in this case, the corresponding adjusted coefficients of determination for boron concentration (adjusted $R^2=0.46$) and boron uptake (adjusted $R^2=0.48$) were similar or lower to those of hot water (adjusted $R^2=0.45$ and 0.60 , respectively) and the saturation extract (adjusted $R^2=0.70$ and 0.49 , respectively), when the latter two soil tests were used alone in the regression equations.

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