

Evaluation of Two Extraction Boron Methods in Iraqi Calcareous Soils

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Abstract

Two boron (B) extracting methods were evaluated at eight sites varying in chemical and physical characteristics from center and south of Mesopotamia plain (Iraq). Correlations were made between the two extraction methods and plant parameters to find the most appropriate method for Iraqi calcareous soil. A high and significant correlation was found between the extracted B values ($r= 0.903$) ($p\leq 0.01$). The HCl extraction method overtook the hot water method, having the highest correlation coefficient r , determining coefficient r^2 and t value between available B and plant indicators. The mean efficiency of relationship (determination coefficient $r^2 \times 100$) between B extracted with hot water and hydrochloric acid methods in the soils under study with plant indicators was 60.3 and 61.5% for both methods, respectively. These results showed that the efficiency of two methods was closer, which is attributed to the similarity of the values of the aforementioned statistical indicators and the presence of a positive and high significant correlation between them. Therefore, the method of extracting available B with acid can be adopted for ease of implementation, the availability of its requirements and for correlating its results more with plant standards.

Keywords: Boron, Hot water, Hydrochloric acid, Calcareous soil and Wheat.

تقييم طريقتين لاستخلاص البورون من ترب العراق الكلسية

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الخلاصة

قيمت طريقتين لاستخلاص بورون التربة الجاهز في ثمانية مواقع مختلفة في خصائصها الكيميائية والفيزيائية من وسط وجنوب العراق. اجريت علاقات ارتباط بين طريقتي الاستخلاص ومؤشرات النبات لإيجاد الطريقة الانسب للترب العراقية الكلسية. وجد ارتباط كبير ومعنوي لقيم البورون المستخلصة بالطريقتين ($r= 0.903$) ($p\leq 0.01$). تفوقت طريقة الاستخلاص بحامض الهيدروكلوريك على طريقة الماء الحار لامتلاكها اعلى معامل ارتباط r ومعامل تحديد r^2 و قيمة t بين البورون الجاهز ومؤشرات النبات. بلغ متوسط كفاءة العلاقة (معامل التحديد $r^2 \times 100$) بين البورون المستخلص بالماء الحار وحامض الهيدروكلوريك في التربة مع مؤشرات النبات 60.3 و 61.5% لكلا الطريقتين على التوالي. ان تقارب كفاءة الطريقتين يعزى الى تقارب قيم المؤشرين الاحصائيين انفة الذكر ووجود علاقة ارتباط معنوية موجبة وعالية بينهما. لذلك فان طريقة استخلاص البورون الجاهز بالحامض يمكن اعتمادها لسهولة تنفيذها وتوافر مستلزماتها ولارتباط نتائجها مع مؤشرات النبات.

كلمات مفتاحية: البورون الجاهز، ماء حار، حامض الهيدروكلوريك، ترب كلسية، حنطة.

Introduction

Boron is a metalloid with its atomic number 5 and its atomic weight 10.811. Its concentration in the Earth's crust is 10 mg kg⁻¹ and located in the second period of the periodic table. B atom is small, trivalent, and its ionic radius is 0.02 nm. Moreover, its acid (boric acid) is very weak, it behaves like Lewis acid. In an aqueous solution with a pH less than 7.0, it is mainly found in the form of non-dissolution. At high pH, boric acid accepts a hydroxyl ion from water and forms a tetrahedral borate ion (Shelp, 1993).



The major minerals that contain B in most soils are tourmaline and the complex of borosilicate. B is released very slowly from these minerals. The stock of B in dryland soils is variable, as borates and Alkaline earth elements predominate. Total boron content in soils ranges from 20-200 mg B kg⁻¹ (Mengel and Kirkby, 1987) and most of it is not available for plant. Generally, the available portion (water-soluble) in the range of 0.4–5.0 mg B kg⁻¹ (Gupta, 1979). (Jonson and Fixen, 1990) found that the critical limit for B extracted with hot water was 1.0 mg kg⁻¹. In Iran, in calcareous soil, B extracted with hot water was found to be 0.65 mg B kg⁻¹ (Balali *et al.*, 2000) (Feiziasl *et al.*, 2003). (Sims and Johnson, 1991) showed that the critical limit for B extracted with hot water was 0.1 – 2.0 mg kg⁻¹ and since B availability decreased with increasing soil pH, the available of it was insufficient in calcareous soils.

B is an essential element for the growth and nutrition of economic plants (Wimmer, 2020) (Vera- Maldonado *et al.*, 2024), but the range between deficiency and toxicity of B concentration is narrower than that of any other nutrients. The most important

factors affecting B availability are soil pH, texture, moisture, temperature, organic matter, and clay content (Goldberg, 1997) (Van Eynde *et al.*, 2020) (Teagasc, 2021) (U. S. Borax, 2022) (ICL Growing Solutions, 2023).

Elements' movement and their biological availability for plant are related to their concentration in soil solution and strongly depend on their specific chemical formulas, exchange and carbonate bonds (Fuentes *et al.*, 2004). Therefore, it is of utmost importance to anticipate the biological availability of elements compared to the total amount of them in order to determine the appropriate fertilizing recommendation and optimal fertilizing management to avoid toxic and pollutant effect of fertilizers.

Choice of extraction solution usually depends on extraction time (since speed measurement is important), ease of preparing the sample for extraction, but at the same time the high efficiency of extraction and molar concentration of the extraction solution and ratio of soil to the solution of the sample are among the important additional indicators that are taken into consideration (Meers *et al.*, 2007). In the practical application of soil and environmental studies that include soil analysis usually depends on either on use single extraction methods (extraction one element) or methods that extract more than one element which enable to measure several formulas or phases (such as bioavailable or phytoavailable) (Gupta and Aten, 1993).

The number of extracts different in their efficacy was assumed to estimate the available soil B (Choudhari *et al.*, 2021) (Zhang *et al.*, 2021) (Marupaka *et al.*, 2022) (Chen *et al.*, 2024). (Ponnamperuma *et al.*, 1981) had been using hot water and diluted HCl to extract B to estimate its value in order to

assess the B state in soils. Most of these proposed methods have been applied in non-calcareous soils, and there is very little information available about their application in calcareous soils (Al-Ameri, 2013). Therefore, this study was conducted to compare two methods for extracting B from calcareous soils, conducting a biological experiment and making correlations between B concentration in the soil and the number of plant indicators.

Materials and methods

Eight sites were selected from the sedimentary plain (Mesopotamia) from central and southern Iraq (three sites in Baghdad governorate, while the other samples were taken from Babil, Wasit, Najaf, Maysan, and Basra governorate). Table 1. Shows some characteristics of these soils which estimated according to (Sparks *et al.*,1996).

Two methods were used to extract soil available B:

a. Hot water method (H. W).

Boron was extracted by boiling water under a reflux condenser 20g of air-dry soil with 40ml of 0.01M CaCl_2 for 5 min., in a fiber digestion apparatus (Berger and Truog, 1939, FAO, 2024). After slight cooling, it was filtered.

b. Dilute hydrochloric acid method.

Boron was extracted by using 0.05N hydrochloric acid solution according to (Ponnamperuma *et al.*,1981) method, which was used by (Kausar *et al.*,1990). Including weight, 10 gm of anaerobically dry soil, to which 20 ml of dilute hydrochloride acid 0.05 N was added, shaking for 5 minutes and filtered.

B extracted by above methods was estimated by using ICP (Inductively Coupled Plasma Atomic Emission Spectrometer model ICPE-9000 SHIMADZU).

To correlate available B with plant B concentration and yield indicators, a biological experiment has been performed in a greenhouse with a randomized complete block design (RCBD). In order to achieve the goals of this experiment, 7.0 Kg plastic pot was used, weighing 5000 g of all soils with three replications, using wheat (cv. Rasheed) as a test plant.

Major elements N P K have been added at 200, 100 and 120 Kg ha⁻¹ levels (Al-Ameri, 2001). At maturity, plants were harvested, and plant parts were dried at 65° C. and digested by dry-ashing according to (Chapman and Pratt,1961). B was measured by using the Inductively Coupled Plasma (ICP).

Results were statistically analyzed using analysis of variance (ANOVA) by using Least Significant Difference value to compare the different treatments averages at 0.05 level (Steel and Torrie, 1980), using the statistical analysis program (SAS, V.6.2) and also used a simple correlation parameter in describing the relationship between plant biological parameters and the two extraction methods.

Table 1. Chemical and physical soils characteristics.

Region	EC dSm ⁻¹	pH	NO ₃ - N	NH ₄ - N	P	K	CaCO ₃	Clay	Sand	Silt	Texture
			mg Kg ⁻¹				gm Kg ⁻¹				
Twaitha (1)	0.79	6.75	12.3 2	7.84	9.86	305.7 0	297.3	459.33	162.6 3	378 .03	Clay
Twaitha (2)	7.84	6.29	32.2	13.44	9.54	426.2	191.7	202.6	428.7	368 .7	Loam
Al-Jadrya	0.27	7.69	18.7 6	11.48	4.64	36.79	159.1	164.33	755.5 3	80. 16	Sandy loam
Wasit	5.14	6.59	15.4 0	5.32	10.9	287.1	239.5	573.49	40.11	386 .4	Clay
Babil	0.76	6.87	16.2 4	12.32	23.1	221.2	206.5	426.55	171.0 4	402 .41	Silty clay
Najaf	6.49	6.02	41.1 6	13.72	9.86	195.6	172.1	416.42	130.0 6	453 .52	Silty clay
Mysan	1.24	7.39	20.7 2	11.48	32.1	285.4	220.0	349.80	202.7 8	447 .42	Clay loam
Basrah	7.61	8.02	11.7 6	1.68	38.98	321.6	285.6	473.80	179.0 2	347 .18	Clay

Results and Discussion

Table 2 showed B extracted by two chemical extraction methods in eight soils used in this study, results indicated effect of extraction methods significantly in the amount of available B extracted in the soils used, amount of B extracted ranged between 0.147 and 4.82 and between 0.676 and 3.06 mg B Kg⁻¹ soil for hot water and hydrochloric acid extracting methods respectively, with average (2.349) and (1.989) mg B Kg⁻¹ Soil for two methods, respectively.

Results in Table 2 shows a significant difference at level ($p \leq 0.05$) between the

two extraction methods, where the hot water method was superior to the hydrochloric acid method, where it extracted a higher amount of B compared to hydrochloric acid, the extraction efficiency reached 85% for acid method compared to the hot water method (100%), values of extracted boron varied in the two methods used for the soils under study due to the different chemical properties of extract solution and extraction conditions for each method as well as the different chemical and physical properties of soils under investigation

Table 2. Boron extracted from soil with different extraction methods.

Governarate	B (H.W.)	B (HCl)	X [−]
	mg B Kg ^{−1} soil		
Baghdad, Twaitha (1)	1.268	1.370	1.319
Baghdad, Twaitha (2)	3.050	2.840	2.947
Baghdad, Al- Jadrya	0.147	0.676	0.411
Wasit	3.760	3.060	3.412
Babil	0.964	1.200	1.082
Najaf	2.520	1.708	2.112
Mysan	2.260	2.340	2.301
Basrah	4.820	2.720	3.768
X [−]	2.349	1.989	2.169
LSD methods (0.05)	0.0899		
LSD soil (0.05)	0.1797		

However, B values extracted by the two methods were highly correlated and significant at the level of 0.01 ($r = 0.903^{**}$) (Fig. 1). Although, the hot water method considers an appropriate test for predicting readiness B in calcareous soils, it was a tediousness and time-consuming method [boiling presence, requiring precision to adjust volume] and there were margins of error [doubt about having a constant boiling time], and needing Boron-free glassware which was not available in most laboratories (Mahler *et al.*, 1984). Therefore, the hydrochloric acid method can be dependent because it was easy and fast, and all its requirements were available, in contrast to the hot water method that was complicated and requires time and caution, and this had pointed out by many studies (Ponnamperuma *et al.*, 1981) (Kausar *et al.*, 1990) (Rashid *et al.*, 1994) (Al-Ameri, 2013).

Results in Table 2 show that there were significant differences at the (0.05) in the level of boron extracted content between the study soils according to the two extraction methods. B extracted values were varied in the eight soils, as it was giving highest value in Basra and Wasit soils in the hot water method, which reached 4.82 and 3.76 mg Kg⁻¹ soil respectively. While the highest values were in Wasit and Twaitha (2) soils, which reached 3.06 and 2.84 mg Kg⁻¹ soil in the hydrochloric acid method, respectively. As for soil that showed a decrease in its content of B extracted by the two extraction methods, it was Al- Jadriya soil, as its value reached 0.147 and 0.676 mg Kg⁻¹ for hot water and acid

methods, respectively, their values were less than the critical limit for boron in the soil (0.1- 2.0 mg Kg⁻¹ with hot water), Hall, (2008) mentioned that for most crops, 1- 4 mg B Kg⁻¹ soil was sufficient to prevent nutrient deficiencies. Less than 0.5 mg B Kg⁻¹ soil is rated as marginal to deficient.

The variation in the B extracted content from study soils by two methods above was attributed to, due to, different chemical properties of the soil, especially its clay content, due to the convergence of the values of other physical and chemical soil properties, such as soil pH and calcium carbonate. Clay content in Basra and Wasit soils reached 473.8 and 573.5 gm Kg⁻¹ soil, respectively, and these soils gave the highest amount of extracted B by two methods, where clay content contributes to B retention through the adsorption process, whether it was physical or chemical. (Niaz *et al.*, 2007) showed that soil B was positively correlated with clay content of soil, also (Osman *et al.*, 2005) and (Shaaban *et al.*, 2006) mentioned that B deficiency usually occurs in plants growing in sandy soils. B adsorption from soil surfaces increased with increasing clay content in the soil (Arora and Chahal, 2009) (Al-Ameri *et al.*, 2019) (Al- Ameri, 2019). B is low in sandy soils of coarse texture and well- drained.

Regression analysis results (Table 4) on the correlation of B with different plant features (Table 3), indicated that there was a significant linear relationship between B extracted with hot water and hydrochloric acid with some wheat plant indicators, which included both straw and grain yield, biological yield, B concentration in straw and grain, total concentration, B uptake in straw and grain and total uptake.

Table 3. Plant parameters.

City	Straw yield	Grain yield	Biological yield	Straw B con.	Grain B con.	Total B con.	Straw B uptake	Grain B uptake	Total uptake
	gm pots ⁻¹			mg Kg ⁻¹			µg pot ⁻¹		
Twaitha (1)	29.4	7.5	42.2	18.0	12.8	30.8	529.2	96.0	625.2
Twaitha (2)	28.46	5.17	43.66	18.3	15.2	33.5	520.818	78.584	599.402
Al-Jadrya	22.96	4.57	23.244	13.45	0.284	13.734	308.812	1.298	310.110
Wasit	19.1	5.1	34.067	15.0	14.967	29.967	286.5	76.332	362.832
Babil	33.7	9.6	58.7	24.0	25.0	49.0	808.8	240.0	1048.8
Najaf	23.0	8.9	38.5	16.0	15.5	31.5	368.0	137.95	505.95
Mysan	24.3	7.6	38.3	16.367	14.0	30.367	397.718	106.4	504.118
Basrah	34.73	7.6	52.73	19.0	18.0	37.0	659.87	136.80	796.67

Extraction methods were evaluated based on two statistical indicators, correlation coefficient (r) and the t value, values of these two indicators were significant at the level ($P \leq 0.05$) of the two extraction methods. The average correlation coefficient and t value for all plant indicators for hot water method were 0.775 and 3.068, respectively, while 0.784 and 3.126 for the hydrochloric acid method. The mean efficacy of relationship (determination coefficient $r^2 \times 100$) between B extracted with hot water and hydrochloric acid method in the soil under study with plant indicators was 60.3 and 61.5% for both hot water and hydrochloric acid, respectively. From these results, it appears that the efficiency of two methods (hot water and hydrochloric acid methods) was close in extracting available B for the plant due to the convergence of values of two statistical indicators and their efficiency ratio, and the presence of a positive and high significant correlation between them (Figure 1).

Biological yield and total uptake exceed the rest of the plant indicators used in the study in obtaining the highest values of the correlation coefficient with the extracted B in both methods (Table 4). The values reached 0.854 and 0.816 respectively in hot water method and 0.831 and 0.827 respectively in HCl method. These results were in line with Rafique *et al.*, 2002, they showed that B content in the calcareous soils used in their studies correlated very well with both methods with B concentration in diagnostic foliar parts, and total boron absorption and boron concentration in grains of mustard and sorghum. Al-Ameri (2013) confirmed that B concentration in grains, total uptake and dry matter exceed the rest of the corn indicators in obtaining the highest values of correlation coefficient with the extracted B in both methods.

Fig 1. correlation between two extraction methods.

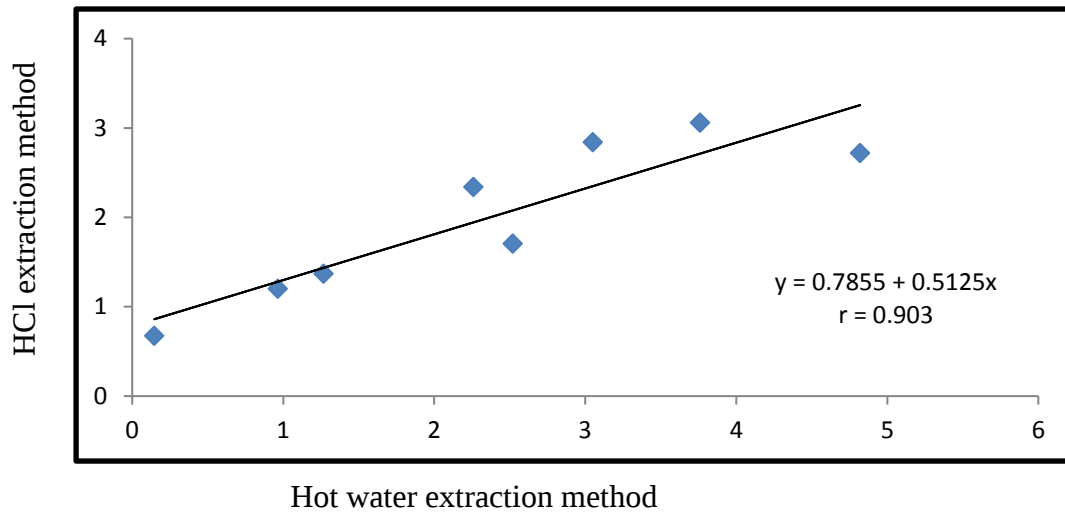


Table 4. Linear correlation between available B and plant parameters.

Plant indicator	Extraction method					
	Hot water			HCl		
	Statically parameter					
	r	r ²	t value	r	r ²	t value
Straw yield	0.806	0.650	3.337	0.759	0.576	2.854
Grain yield	0.786	0.618	3.113	0.805	0.648	3.322
Biological yield	0.854	0.730	4.024	0.831	0.690	3.653
B concentration in straw	0.714	0.510	2.499	0.783	0.613	3.082
B concentration in grain	0.709	0.503	2.465	0.710	0.504	2.468
Total B concentration	0.737	0.543	2.672	0.760	0.578	2.867
B uptake in straw	0.784	0.615	3.096	0.794	0.630	3.197
B uptake in grain	0.770	0.592	2.951	0.784	0.614	3.089
total B uptake	0.816	0.666	3.457	0.827	0.684	3.606
X̄	0.775	0.603	3.068	0.784	0.615	3.126
table r at 0.05= 0.707 and table t value at 0.05= 2.447						

Conclusion

Hydrochloric acid extraction was a fully experimental working method for

simultaneously extracting soils B, and it can be adopted instead of the hot water method because it was a simple and reliable method and its requirements were inexpensive and extracts a greater number of sample in a less time and did not require refluxing equipment, unlike hot water method that considered tedious for routine conditions and requires some special precautions.

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