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Response of chickpea (*Cicer arietinum* L.) yield to zinc, boron and molybdenum application under pot conditions

J. B. Valenciano*, J. A. Boto and V. Marcelo

Department of Agrarian Engineering and Sciences. University of León. Avda. Portugal, 41. 24071 León. Spain

Abstract

Spain is the main chickpea (*Cicer arietinum*) producing country in Europe, despite there are few studies on micronutrient application to chickpea. The response of chickpea to the applications of Zn, B and Mo was studied in pot experiments with natural conditions and acidic soils in northwest Spain from 2006 to 2008 following a factorial statistical pattern (5 × 2 × 2) with three replicates. Five concentrations of Zn (0, 1, 2, 4 and 8 mg Zn pot^{-1}), two concentrations of B (0 and 2 mg B pot^{-1}), and two concentrations of Mo (0 and 2 mg Mo pot^{-1}) were added to the pots. Chickpea responded to the Zn, B and Mo applications. There were differences between soils. The mature plants fertilized with Zn, with B and with Mo had a greater total dry matter production. Harvest Index (HI) improved with the Zn application and with the Mo application. The highest HI was obtained with the $Zn_4 \times B_2 \times Mo_2$ treatment (60.30%) while the smallest HI was obtained with the $Zn_0 \times B_0 \times Mo_0$ treatment (47.65%). The Zn, B and Mo applications improved seed yield, mainly due to the number of pods per plant. This was the yield component that had the most influence on, and the most correlation with seed yield. The highest seed yield was obtained from the $Zn_4 \times B_2 \times Mo_2$ treatment (4.00 g plant^{-1}) while the lowest was obtained from the $Zn_0 \times B_0 \times Mo_0$ treatment (2.31 g plant^{-1}). There was a low interaction between the three micronutrients. The Zn application was more efficient when it was applied with both B and Mo.

Additional key words: dry matter, *Kabuli* type chickpea, micronutrients, yield components.

Resumen

Respuesta del garbanzo cultivado en macetas a las aplicaciones de zinc, boro y molibdeno

España es el principal productor de garbanzo en Europa, pese a ello hay pocos estudios sobre la aplicación de micronutrientes. Se estudió desde 2006 a 2008 la respuesta del garbanzo cultivado en macetas al aire libre a las aplicaciones de Zn, B y Mo, usando tres suelos ácidos, según un diseño factorial (5 × 2 × 2) con tres repeticiones. Cinco concentraciones de Zn (0, 1, 2, 4 y 8 mg Zn maceta^{-1}), dos de B (0 y 2 mg B maceta^{-1}) y dos de Mo (0 y 2 mg Mo maceta^{-1}) fueron añadidas a las macetas. El garbanzo respondió a las aplicaciones de Zn, de B y de Mo, existiendo diferencias entre suelos. En la madurez, las plantas fertilizadas con Zn, B y Mo tuvieron mayor producción de materia seca. El índice de cosecha (IC) mejoró con la aplicación de Zn y de Mo. El IC más alto se obtuvo con el tratamiento $Zn_4 \times B_2 \times Mo_2$ (60,30%) y el IC más bajo con el tratamiento $Zn_0 \times B_0 \times Mo_0$ (47,65%). Las aplicaciones de Zn, de B y de Mo mejoraron el rendimiento de semilla, principalmente debido al número de vainas por planta, componente del rendimiento más influyente y más estrechamente correlacionado con el rendimiento. El rendimiento más alto se obtuvo con el tratamiento $Zn_4 \times B_2 \times Mo_2$ (4,00 g planta^{-1}) y el más bajo con el tratamiento $Zn_0 \times B_0 \times Mo_0$ (2,31 g planta^{-1}). Existió una interacción poco significativa entre los tres micronutrientes, siendo la aplicación de Zn más eficaz cuando se aplicó con B y Mo.

Palabras clave adicionales: componentes del rendimiento, garbanzo tipo *Kabuli*, materia seca, micronutrientes.

Introduction

Chickpea (*Cicer arietinum* L.) is the principal grain legume crop grown in the Mediterranean region, and Spain is the main chickpea-producer in Europe, 31,600 ha and 30,100 t in 2008 (FAO, 2010). Despite its importance, few studies have been conducted to analyse the application of micronutrients to chickpea. Although

* Corresponding author: joseb.valenciano@unileon.es
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All authors are members of the SEA.

Abbreviations used: CV (coefficient of variation), DM (dry matter), DTPA (diethyl triamine penta-acetic acid), DW (dry weight), EC (electrical conductivity), HI (harvest index), PWR (pods weight ratio), RWR (roots weight ratio), SLWR (stems with leaves weight ratio).

the chickpea is a rustic edible plant, widespread deficiencies and/or imbalances of mineral nutrients in the soils along with limited moisture supply are considered major environmental stresses leading toward yield loss in chickpea (Khan, 1998). Chickpea is mainly cultivated as a rainfed crop and water stress often affects both the productivity and the yield stability of the chickpea. Rainfed soils are generally degraded with poor native fertility. Micronutrients play an important role in increasing legume yield through their effects on the plant itself, on the nitrogen-fixing symbiotic process and the effective use of the major and secondary nutrients, resulting in high legume yields. The magnitude of yield losses due to nutrient deficiency also varies among the nutrients (Ali *et al*., 2002). Micronutrient availability for the plant depends, among other factors, texture, organic matter and, mainly, soil pH.

The main micronutrient that limits chickpea productivity is zinc (Zn) (Ahlawat *et al*., 2007). Boron (B) may cause yield losses up to 100% (Ahlawat *et al*., 2007). The availability of molybdenum (Mo) is low in acidic soils. With the exception of Mo, the availability of micronutrients is the greatest in the very slight to medium acid range. In general, each tonne of chickpea grain removes 38 g of Zn from the soil, and it is estimated that 35 g of B and 1.5 g of Mo are removed from the soil as well (Ahlawat *et al*., 2007).

Among micronutrients, Zn deficiency is perhaps the most widespread (Roy *et al*., 2006; Ahlawat *et al*., 2007) and is common among chickpea-growing regions of the world. Chickpea is generally considered sensitive to Zn deficiency (Khan, 1998), although there are differences in sensitivity to Zn deficiency between varieties (Khan, 1998; Ahlawat *et al*., 2007). A comparison between several crop species has shown that chickpea is more sensitive to Zn deficiency than cereal and oil seeds (Tiwari and Pathak, 1982). The critical Zn concentrations in soils vary from 0.48 mg kg^{-1} to 2.5 mg kg^{-1} depending on soil type (Ahlawat *et al*., 2007) and according to Ankerman and Large (1974) soils have low Zn availability when there is less than to 1.1 mg kg^{-1} of Zn (DTPA extraction). Zn deficiency decreases crop yield and delays crop maturity. Also, Zn deficiency reduces water use and water use efficiency (Khan *et al*., 2004) and also reduces nodulation and nitrogen fixation (Ahlawat *et al*., 2007), which contributes to a decrease in crop yield. In neutral to alkaline soils, Zn deficiencies can be encountered (Roy *et al*., 2006), Zn solubility decreases markedly above pH 6.0-6.5 (Sims, 2000). Zn uptake is positively correlated with the amount of organic matter in the soil and negatively correlated with the phosphorus (P) concentration in the soil (Sillanpää, 1972; Hamilton *et al*., 1993; Ahlawat *et al*., 2007). Soils that have a higher concentration of sand and a lower concentration of organic matter produced lower crop yields which lead to poor Zn utilization (Singh and Ram, 1996).

B which also limits chickpea productivity is a less important factor than Zn (Ahlawat *et al*., 2007). B, in acidic soils, has been shown to be a major reducer of chickpea yields in some regions (Srivastava *et al*., 1997). In comparison with others crops, the response of the crop to the application of B is higher in chickpea than in some cereals (Wankhade *et al*., 1996); although differences between chickpea cultivars concerning B deficiency have also been observed (Ahlawat *et al*., 2007). The application of B is important when the concentration of B in the soil is less than 0.3 mg kg^{-1} (Ahlawat *et al*., 2007). According to Ankerman and Large (1974) soils have low B availability when the concentration of B in the soil is less than 0.6 mg kg^{-1} (hot water extraction) and according to Sillanpää (1972) the soil may have a deficiency of B when their concentration in the soil is less than 0.5 mg kg^{-1} depending on the conditions, the extraction time and the soil. B deficiency also causes flower drop and, subsequently, poor podding of chickpeas (Srivastava *et al*., 1997) and poor yields. A B deficiency can be caused by high pH in the soil, the availability of B decreases when the pH is larger than 6.5-7.0 (Sims, 2000), which occur in highly leached sandy soils or in low organic matter soils.

Total Mo content in soil can vary from 0.2 to 5.0 mg kg^{-1} (Sims, 2000) but Mo that is in the soil is largely unavailable, since usually less than 0.2 mg kg^{-1} of Mo has been reported to be soluble (Sillanpää, 1972). According to Ankerman and Large (1974) soils have low Mo availability when the concentration of Mo in the soil is less than 0.11 mg kg^{-1} (ammonium acid oxalate). In Mo-deficient chickpea, the flowers produced are less in number, smaller in size and many of them fail to open or to mature, consequently this leads to lower seed yield (Ahlawat *et al*., 2007). Mo is related directly to N fixation by legumes (Roy *et al*., 2006). The availability of Mo increases as the pH of the soil approaches neutrality (pH 7.0) or is higher than neutral (Sims, 2000). Mo availability is the lowest when the pH of the soil is in the very slight to medium acid range. Mo deficiency is common in very acidic soils especially in crops that are very sensitive to low concentrations of Mo such as legumes (Sims, 2000). High phosphate levels are positively correlated with Mo deficiency.

Foliar fertilization and soil application are effective practices for the implementation of some micronutrients (Roy *et al.*, 2006). Zn, B and Mo application results are controversial according to literature reports (Yanni, 1992; Braga and Vieira, 1998; Johansen *et al.*, 2007; Shil *et al.*, 2007). Also, nutrient interaction in crop plants affects yield of annual crops, this nutrient interaction can be positive, negative or neutral (Fageria *et al.*, 1997). Soil, plant and climatic factors can influence interaction.

This work was conducted to determine the effect of soil Zn and soil B and foliar Mo applications and their possible interactions on growth and seed yield of a *Kabuli* chickpea.

Material and methods

Experimental design

Three experiments were carried out in the province of León on a *Kabuli* chickpea ecotype (cv. Pedrosillano), between 2006 and 2008. The seed of this cultivar is small (the weight of 1,000 seeds is 340 g) cream, rounded and smooth. The experiment was carried out using a factorial statistical pattern (5 × 2 × 2) with three replicates. The first factor was the application of Zn, which had five different levels of Zn, 0, 1, 2, 4 and 8 mg per pot, and the treatment codes were: Zn_0, Zn_1, Zn_2, Zn_4 and Zn_8, respectively. The second factor was the application of B, which had two different levels of B, 0 and 2 mg per pot, and the treatment codes were: B_0 and B_2, respectively. The third factor was the application of Mo, which had two different levels of Mo, 0 and 2 mg per pot, and the treatment codes were: Mo_0 and Mo_2, respectively. Zn was added to the soil as Zn chelate (agent chelating, DTPA, EDTA and HEDTA), 14% (w/v); B was added to the soil as B solution (8% w/v) and Mo was applied foliarly as Mo solution (6.3% w/v).

Plant material and crop management

Plants were grown in Ribas de la Valduerna (León, Spain) (42°18.5'N, 5°57.1'W) under natural environmental conditions in PVC pots. Pots (210 mm diameter × 300 mm deep) were filled with 4 kg of soil. The main physical and chemical properties of the soils which were used are listed in Table 1. The experiments were conduced using acidic soils. These soils have a medium availability of Zn and a very low to a high availability of B according to Ankerman and Large (1974) and high in total Mo according to Gupta (1997). The soils were collected from three sites, located in Ribas

Table 1. Main physical and chemical characteristics of soils used in the experiments, with local names

	Soil (local name)-Year		
	Sotico-2006	**Housa-2007**	**Era-2008**
Texture (Bouyoucus densimeter)	Loam	Loam	Loam
Organic matter (Walkley-Black) (g kg^{-1})	2.1	2.3	2.3
pH (1:2.5, water)	5.6	5.9	6.2
EC[1] (1:5, water) (dS m^{-1})	0.06	0.13	0.06
Calcium carbonate (Bernard calcimeter) (g kg^{-1})	Negligible	Negligible	Negligible
P (Olsen) (mg kg^{-1})	31.6	26.1	8.5
K (1 N NH_4 Ac) ($cmol_c$ kg^{-1})	0.55	0.15	0.19
Ca (1 N NH_4 Ac) ($cmol_c$ kg^{-1})	3.07	4.43	4.63
Mg (1 N NH_4 Ac) ($cmol_c$ kg^{-1})	0.59	0.73	0.92
Na (1 N NH_4 Ac) ($cmol_c$ kg^{-1})	0.09	0.02	0.06
Mn (DTPA)[2] (mg kg^{-1})	9.79	17.50	16.59
Fe (DTPA) (mg kg^{-1})	105.0	160.0	83.7
Cu (DTPA) (mg kg^{-1})	1.12	1.38	1.38
Zn (DTPA) (mg kg^{-1})	1.26	2.03	1.53
B (hot water) (mg kg^{-1})	1.95	0.15	0.40
Mo (nitric acid digestion) (mg kg^{-1})	1.76	2.32	0.61

[1] EC: electrical conductivity. [2] DTPA: diethyl triamine penta-acetic acid.

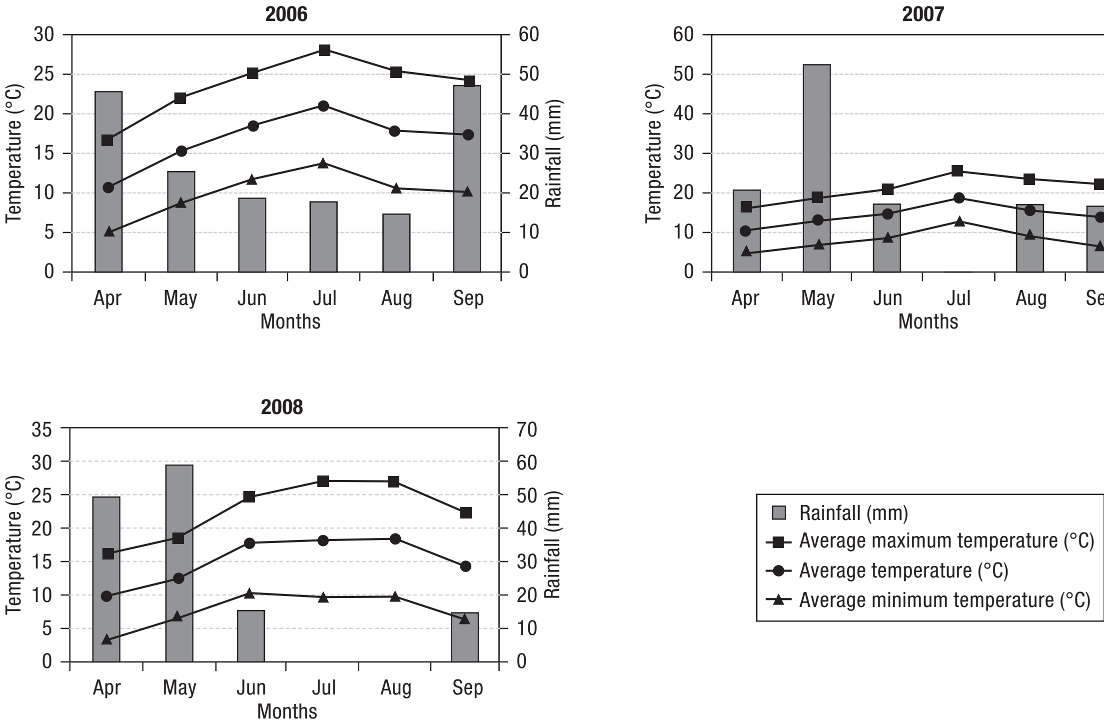

Figure 1. Climatic conditions at Ribas de la Valduerna (León, Spain) during the experimental period. *Source:* Meteorological Station, Sugar Refinery, La Bañeza, Spain.

de la Valduerna (León, Spain), which had not been fertilized for agriculture. The temperature related parameters for this area when the experiment was conducted are shown in Figure 1.

For each experiment, 10 seeds per pot were sown at 3-cm depth in each pot on 6th May 2006, 20th April 2007 and 30th April 2008. One week after emergence, the seedlings were thinned so that there were only two plants per pot.

Three weeks after emergence the five different concentrations of Zn and the two different concentrations of B were added, separately, to each pot, 10th June 2006, 24th May 2007 and 2nd June 2008. The application of Mo was carried out by spraying each pot with a foliar spray 30 days after emergence (Bhanavase and Patil, 1994), 19th June 2006, 2nd June 2007 and 11th June 2008.

Soil moisture was maintained near field capacity by watering the plants every day with deionized water. Chlorothalonil (tetrachloroisophthalonitrile) 50% (w/v) and quinosol (8-hydroxyquinoline sulphate) 50% (w/v) plus thiram (tetramethyl-thiuram sulfide) 80% (w/v) were used to reduce the incidence of disease and for chickpea plant protection (Ondategui, 1996). There were no incidences of pests or diseases during the experiments.

Measurements and statistical analysis

At maturity (19th August 2006, 7th August 2007 and 14th August 2008), all plants were harvested. The roots, the stems with leaves and the pods including seeds were separated and dried in an oven which had a temperature of 80°C to a constant weight, and weighed.

The dry weight (DW) data were used to calculate indices of DW partitioning: root weight ratio (RWR) = root DW/total DW, stem with leaves weight ratio (SLWR) = stem with leaves DW/total DW, pod weight ratio (PWR) = pods DW/total DW and harvest index (HI) = seed DW/total DW.

Also at harvest, plant yield and yield component data (the number of pods per plant, the number of seeds per pod, and the 1,000-seed weight) were collected. The grain yield (g plant^{-1}) was calculated from the yield components.

The data were analysed by conducting an analysis of the variance using SPSS version 15.0.1. The comparison of the means which was conducted was based on Tukey test ($P < 0.01$ and $P < 0.05$) (Steel and Torrie, 1986). Also different correlations were also calculated.

Results

The symptoms of Zn, B and Mo deficiencies (Roy *et al*., 2006) were not observed in any of the pots; however, there were significant differences between the treatments.

The environmental conditions during experiments affected the plant's response differently (Tables 2 and 3), and there were significant differences between environments (soils). At maturity, the production of dry matter (DM) was higher for Era-2008, where the research was conducted using less acidic soils. PWR was smaller for Housa-2007 while SLWR was higher. There was no significant difference in RWR between environments. HI was significantly smaller for Housa-2007 (Fig. 2).

The application of Zn, B and Mo resulted in a higher production of DM (Table 2). The soil Zn application increased growth, due to an increase in the DW of the pods including seeds; however, the root DW and the stems with leaves DW were not affected (Table 2). Treatments influenced DW partitioning between plant organs SLWR decreased until Zn_4, while PWR and HI increased until Zn_4. HI decreased when 8 mg of Zn per pot was applied. The soil B application increased the total DW production, due to an increase in the DW of the pods including seed (Table 2). The B treatments did not influence the DW partitioning between plant organs. The Mo foliar application caused an increase in DM production, due to an increase in the DW of the pods including seeds (Table 2). The Mo treatments influenced the DW partitioning between plant organs, the RWR and SLWR values were higher with the Mo_0 treatment, while PWR value was higher with the Mo_2 treatment.

There was significant interaction between environment and B for HI. The lowest HI was obtained in Housa-2007 × B_0 (Fig. 3). There were low significant interactions between Zn and B for total DW, the highest total DW value was obtained with Zn_4 and B_2 (6.85 g plant^{-1}) and the lowest was obtained with Zn_0 and B_2 (4.98 g plant^{-1}). Also, there was low significant interaction between Zn and B for pods including seeds DW, the highest pods including seeds DW value was obtained with Zn_4 and B_2 (4.59 g plant^{-1}) and the lowest was obtained with Zn_0 and B_2 (2.90 g plant^{-1}). For root DW

Table 2. Effects of concentration of Zn, B and Mo application on dry matter production of chickpea plants at maturity with indication of their significance of analysis of variance and their coefficient of variation (CV)

	Root DW (g plant^{-1})	Stem with leaves DW (g plant^{-1})	Pods including seeds DW (g plant^{-1})	Total DW (g plant^{-1})	HI (%)
Environment (E)	$P \leq 0.01$	$P \leq 0.01$	$P \leq 0.01$	$P \leq 0.01$	$P \leq 0.01$
Zinc application (Zn)	NS	NS	$P \leq 0.01$	$P \leq 0.01$	$P \leq 0.01$
Zn_0	0.38	1.76	2.95	5.10	46.76
Zn_1	0.38	1.77	3.44	5.49	50.88
Zn_2	0.40	1.79	3.68	5.87	50.76
Zn_4	0.41	1.77	4.14	6.31	52.57
Zn_8	0.49	1.79	3.50	5.68	49.43
Boron application (B)	$P \leq 0.10$	NS	$P \leq 0.05$	$P \leq 0.01$	NS
B_0	0.38	1.70	3.39	5.47	49.61
B_2	0.41	1.81	3.69	5.90	50.55
Molybdenum application (Mo)	NS	NS	$P \leq 0.01$	$P \leq 0.01$	$P \leq 0.01$
Mo_0	0.38	1.70	3.25	5.34	48.79
Mo_2	0.40	1.81	3.83	6.04	51.37
Interactions					
Zn × B	NS	NS	$P \leq 0.10$	$P \leq 0.10$	NS
Zn × Mo	NS	NS	NS	NS	NS
B × Mo	$P \leq 0.10$	NS	NS	NS	NS
Zn × B × Mo	NS	$P \leq 0.10$	NS	NS	NS
CV (%)	24.5	25.4	13.8	27.4	14.6

DW: dry weight. HI: harvest index. NS: not significant.

Table 3. Mean yield components and seed yield of the main treatment with indication of their significance of analysis of variance and their coefficient of variation (CV)

	Yield components			Yield (g plant^{-1})
	Pods plant^{-1}	**Seeds pod^{-1}**	**1,000-seed weight (g)**	
Environment (E)	$P \le 0.01$	$P \le 0.01$	$P \le 0.01$	$P \le 0.01$
Zinc application (Zn)	$P \le 0.01$	NS	$P \le 0.05$	$P \le 0.01$
Zn_0	6.48	1.10	362.08	2.55
Zn_1	7.04	1.15	346.01	2.80
Zn_2	7.88	1.15	330.06	2.98
Zn_4	8.24	1.12	350.39	3.23
Zn_8	7.43	1.16	329.78	2.83
Boron application (B)	$P \le 0.10$	NS	$P \le 0.10$	$P \le 0.05$
B_0	7.18	1.14	337.33	2.76
B_2	7.65	1.13	350.00	3.00
Molybdenum application (Mo)	$P \le 0.01$	$P \le 0.05$	NS	$P \le 0.01$
Mo_0	6.74	1.12	347.31	2.63
Mo_2	8.08	1.15	340.03	3.13
Interactions				
Zn × B	NS	NS	NS	$P \le 0.10$
Zn × Mo	NS	NS	$P \le 0.10$	$P \le 0.10$
B × Mo	NS	NS	NS	NS
Zn × B × Mo	NS	NS	NS	$P \le 0.10$
CV (%)	22.2	9.9	17.4	26.4

NS: not significant.

there was low significant interaction between B and Mo, the highest root DW value was obtained when both micronutrients were applied (0.43 g plant^{-1}). Finally, there was low significant interaction between the three nutrients for the stem with leaves DW, the highest stem with leaves DW value was obtained with Zn_2 and B_2 and Mo_2 (2.18 g plant^{-1}) and the lowest was obtained with Zn_1 and B_0 and Mo_0 (1.47 g plant^{-1}).

There were highly significant differences among the environments for yield and for yield components (Table 3, Fig. 2). All the yield components improved for Era-2008 and therefore the highest yield value was

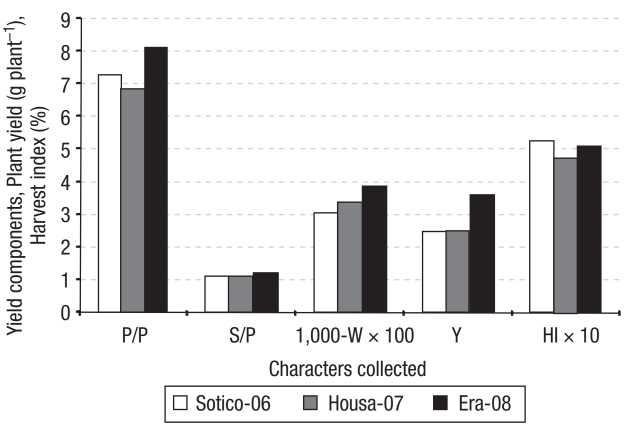

Figure 2. Effect of environment on yield components [P/P: pods plant^{-1}, S/P: seeds pod^{-1}, 1,000-W: 1,000-seed weight (g)], yield (Y, g plant^{-1}) and harvest index (HI). Average of all treatments.

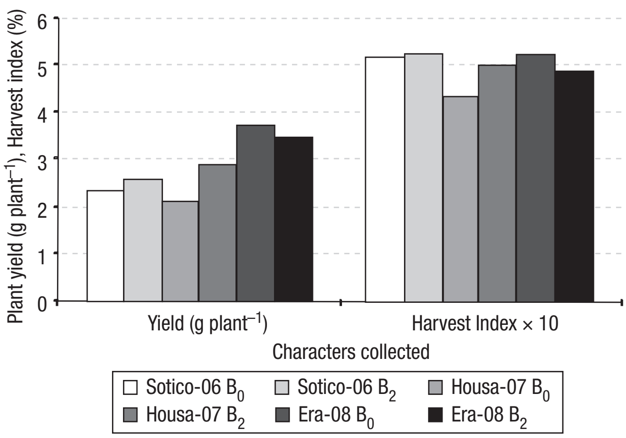

Figure 3. Effect of environment × B interaction on yield and harvest index.

obtained by Era-2008 (3.61 g plant^{-1}). Seed yield was highly correlated with total DW (0.892). HI was highly correlated with the number of pods per plant (0.531) and with seed yield (0.438). The number of pods per plant was closely correlated with seed yield (0.831).

The application of Zn, B and Mo resulted in more seed production (Table 3). There were highly significant differences for soil Zn application on pods per plant and plant yield and significant differences on 1,000-seed weight. The least number of pods per plant (6.48 pods plant^{-1}) and the heaviest 1,000-seed weight (362.08 g) were obtained from the Zn_0 treatment. The lowest plant yield was obtained when the Zn application was not carried out (2.55 g plant^{-1}). Chickpea yield increased with the incremental increases in the application of Zn to until Zn_4 (3.23 g plant^{-1}). The analysis of the variance established that the environment × Zn interaction only had a low significant effect on the number of seeds per pod and interaction on the 1,000-seed weight but there was no significant difference on yield. There were also low significant differences for the soil B application on the number of pods per plant and the 1,000-seed weight and significant differences on yield (Table 3). The fewest number of pods per plant (7.18 pods plant^{-1}) and the lightest 1,000-seed weight (337.33 g) were obtained from the B_0 treatment. The highest yield was obtained when the B application was carried out (3.00 g plant^{-1}). There was a highly significant interaction between the soils and the B application for yield (Fig. 3). Also there was a highly significant interaction between the soils and the B application for number of pods per plant and a low significant interaction for the number of seeds per pod and for the 1,000-seed weight. The biggest differences in yield (35%) for the incremental increases with the soil B application were obtained for Housa-2007 (Fig. 3). The highest overall yield was obtained by Era-2008 × B_0 (3.74 g plant^{-1}). There were significant differences for foliar Mo application on the number of seeds per pod and highly significant differences on the number of pods per plant and yield. The fewest number of pods per plant (6.74 pods plant^{-1}) and lowest number of the seeds per pod (1.12 seeds pod^{-1}) were obtained from the Mo_0 treatment. The highest yield was obtained when the Mo application was carried out (3.13 g plant^{-1}).

There was a low significant Zn × B interaction on seed yield (Table 3). The highest seed yield was obtained from the $Zn_4 \times B_2$ treatment (3.60 g plant^{-1}) followed by the $Zn_2 \times B_0$ treatment (2.98 g plant^{-1}) and $Zn_2 \times B_2$ treatment (2.98 g plant^{-1}). The lowest seed yield was obtained from the $Zn_0 \times B_2$ treatment (2.51 g plant^{-1}) followed by the $Zn_0 \times B_0$ treatment (2.59 g plant^{-1}) (Fig. 4). There was also a low significant Zn × Mo interaction on seed yield (Table 3). The highest seed yield was obtained from the $Zn_4 \times Mo_2$ treatment (3.64 g plant^{-1}) followed by the $Zn_2 \times Mo_2$ treatment (3.37 g plant^{-1}). The lowest seed yield was obtained from the $Zn_0 \times Mo_0$ treatment (2.43 g plant^{-1}) followed by the $Zn_2 \times Mo_0$ treatment (2.58 g plant^{-1}) (Fig. 5). There was finally also a low significant Zn × B × Mo interaction on seed yield (Table 3). The highest seed yield was obtained from the $Zn_4 \times B_2 \times Mo_2$ treatment (4.00 g plant^{-1}) followed by the $Zn_2 \times B_2 \times Mo_2$ treatment (3.53 g plant^{-1}). The lowest seed yield was obtained from the $Zn_0 \times B_0 \times Mo_0$ treatment (2.31 g plant^{-1}) followed by the $Zn_1 \times B_0 \times Mo_0$ treatment (2.41 g plant^{-1}) and by the $Zn_2 \times B_2 \times Mo_0$ treatment (2.41 g plant^{-1}) (Fig. 6).

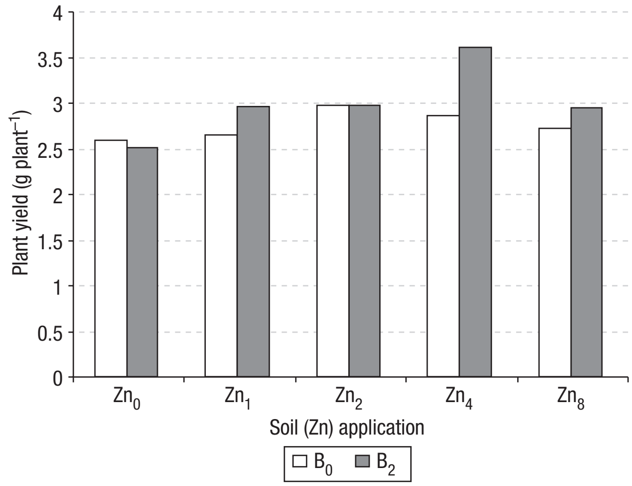

Figure 4. Effect of Zn × B interaction on yield.

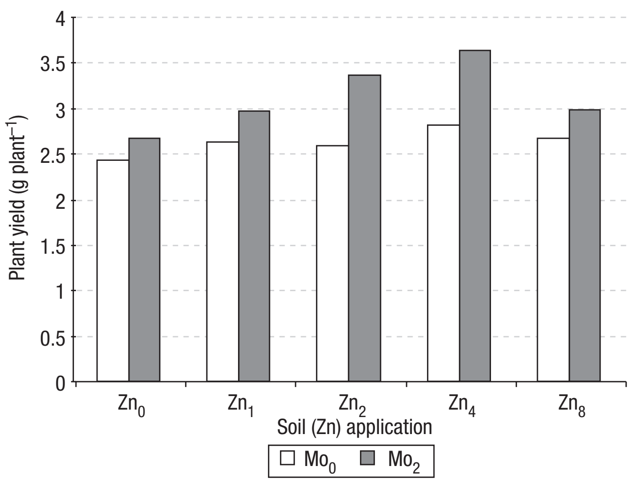

Figure 5. Effect of Zn × Mo interaction on yield.

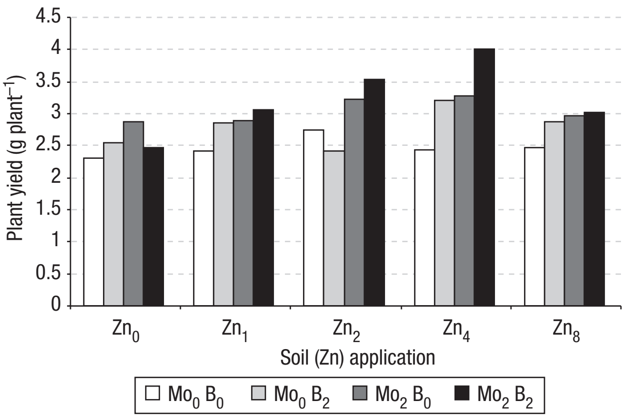

Figure 6. Effect of Zn × B× Mo interaction on yield.

Discussion

The response of the chickpea to the soil micronutrient application varied with environment, this helps to explain their influence on micronutrient deficiency, as Loneragan and Webb (1993) observed for Zn. At maturity, DM production was higher for Era-2008, and since the temperature was not a critical factor (Nielsen, 2001) (it was similar all three years) these differences could be explained, mainly, by the differences in the pH values of the soil because the texture and the organic matter content of the soils were also similar (Singh and Ram, 1996). It has been found that chickpea adapts better to less acidic soils (Ondategui, 1996; Ahlawat *et al*., 2007). This shows that there is a strong environmental influence on chickpea performance, which has also been recorded by other authors (Singh and Sandhu, 2006).

As in other leguminous ones, the application of Zn, B and Mo resulted in a more vegetative growth in acidic soils (Singh *et al*., 1992) this was derived from more production of DM. The growth and yield characteristics were affected by the Zn application. The soil Zn application increased plant growth (Khan *et al*., 2000) and at maturity plants that were fertilized with Zn had a greater total production of DM (Brennan *et al*., 2001). The DM production increase, with increased Zn supply, was mostly due to the increase in the number of pods (including seeds) per plant. The roots treatments influenced the DW partitioning between plant organs. HI increased with an increase in the application of Zn until the Zn_4 level, HI decreased when 8 mg of Zn were applied to each pot, this decrease was also observed by Tripathi *et al*. (1997) where high Zn applications decreased the plant yield slightly. The plant growth was affected by the B application; at maturity plants fertilized with B had greater total DM production, plant growth increases when the availability of B improves (Ahlawat, 1990). Plant growth was affected by the Mo application; at maturity plants fertilized with Mo had greater total DM production, because the Mo foliar application caused an increase in plant growth (Bhanavase and Patil, 1994; Johansen *et al*., 2007). The DM production increase, with increased Mo supply, was mostly due to the increase in the number of pods (including seeds) per plant and also because, according to (Ahlawat *et al*., 2007), there were more flowers produced. The treatments influenced DW partitioning between plant organs. With the increase in the application of Mo, HI increased, the increase in HI can be mainly derived to the increase in seed production (Ahlawat *et al*., 2007).

There was a significant interaction between the environment and B for HI. The lowest HI was obtained by Housa-2007 × B_0, if the availability of B was very low the plant yield decreased a lot; the applications of B were less effective when there was higher availability of B (Ahlawat *et al*., 2007). There were low significant interactions between the micronutrient applications. The interaction between Zn and B on plant growth, when the availability of Zn and B is low, has also been documented in other crops (Hosseini *et al*., 2007). According to Bozoglu *et al*. (2007) on neutral pH soil, the Zn × Mo interaction on the chickpea growth was not recorded. Shil *et al*. (2007) found that there was an interaction between B and Mo but the interaction was only for plant height. Micronutrient application can improve the growth (Johansen *et al*., 2007).

There were highly significant differences among the environments for yield and for yield components. All of the yield components improved for Era-2008 and therefore the highest yield was obtained by Era-2008. These differences could be explained, mainly, by the different soil pH values. For Era-2008 the pH value of the soil was greater than 6 and according to Ondategui (1996) and to Ahlawat *et al*. (2007) chickpea develops better when the pH of the soil is within the 6 to 9 range. Also, the high level of phosphorus in the Sotico-2006 and the Housa-2007 soils, according to Ankerman and Large (1974), could limit yields because of their antagonistic effect on other nutrients. The temperature would have less influence because it was not critical (Nielsen, 2001). This highly significant difference between the environments for all of the characters shows that the environment has a strong influence on chickpea performance, this has also been recorded by other authors (Singh and Sandhu, 2006). The total DW was highly correlated with seed yield (Bhatia *et al*., 1993). According to Kumar *et al*. (2002) HI exhibited the highest significant positive correlation with the number of pods per plant followed by seed yield. The number of pods per plant is the most influential yield component, and is the component that is the most closely correlated with seed yield (Maiti and Wesche-Ebeling, 2001). According to Bhatia *et al*. (1993) the number of pods per plant was the most variable yield component, on the other hand the number of seeds per pod was the least variable yield component and the average of seeds per pod that were obtained in this experiment matched the mean range reported in the literature (Khanna-Chopra and Sinha, 1987). The use of these micronutrients has improved productivity.

Chickpea responded to the soil Zn applications although Zn availability is higher in this pH range (Roy *et al*., 2006) but high P can reduce Zn uptake (Sillanpää, 1972; Hamilton *et al*., 1993; Ahlawat *et al*., 2007). Brennan *et al*. (2001) reported that the relative response of chickpea to applications of Zn is larger than that of other crops. The addition of Zn increased chickpea yield (Brennan *et al*., 2001), but the increases in chickpea yield only occurred until Zn reached the Zn_4 level; the Zn_8 treatment lowered the maximum chickpea yield (Tripathi *et al*., 1997). Zn increased growth and yield (Khan *et al*., 2000). The increase in yield was the result of the increase in the number of pods per plant, which is the same as other leguminous plants (Valenciano *et al*., 2007). In a previous Zn fertilization study involving pots Khan (1998) also reported an increase in grain yield, which was mainly due to an increase in the number of pods per plant, with the application of Zn when the soil had high moisture availability. Valenciano *et al*. (2009) also obtained similar results using the same types of soils but with different environmental conditions. The analysis of the variance established that the environment × Zn interaction only had a low significant effect on the number of seed per pod and on the 1,000-seed weight, but there was no significant difference on yield. The yield response was similar to other work conducted in the past because, although Housa-2007 and Era-2008 had low Zn availability (Ankerman and Large, 1974), in Sotico-2006 the high P level in the soil may induce Zn deficiency by decreasing Zn uptake from the soil (Sillanpää, 1972; Hamilton *et al*., 1993; Ahlawat *et al*., 2007).

Chickpea responded to the soil B application; Ali and Mishra (2001) also found a significant response when foliar applications of B were carried out. The highest yield was obtained when the B application was carried out. The additions of B increased chickpea yield when there was a low availability of B (Roy *et al*., 2006; Shil *et al*., 2007) although Panwar *et al*. (1998) observed a toxic effect of B in high doses; high rates of B application can cause a reduction in crop yield (Sakal *et al*., 1988), mainly in dry conditions (Ahlawat *et al*., 2007). The results of the interactions between the soils and the application of B for yield component and for yield were expected due to the difference in B availability in the different soils. The response to soil B application was higher for Housa-2007, according to Ankerman and Large (1974) the soils with a very low B availability respond better to B application. The largest differences in yield between the incremental soil application of B were obtained by Housa-2007. The response to soil B application is higher in B deficient soil (Wankhade *et al*., 1996; Ahlawat *et al*., 2007).

Chickpea, as in the work conducted by Ali and Mishra (2001), responded well to foliar Mo application due to the low availability of Mo in the acidic soils that were used in the research. There were a significant difference for foliar Mo application on the number of seeds per pod and highly significant differences on the number of pods per plant and yield. Shil *et al*. (2007) also found that yield and these yield components were influenced by application of Mo. The fewest number of pods per plant and the smallest number of seeds per pod were from obtained the Mo_0 treatment. Bozoglu *et al*. (2007) obtained contrary results, in their work the number of pods per plant decreased with the application of Mo; however, their experiments were carried out using neutral pH soil and therefore the availability of Mo is higher. The highest yield was obtained when Mo application was carried out. In previous work (Johansen *et al*., 2007) grain yields were lower in pots without Mo. Although, according to Gupta (1997) even though the total Mo in the soil was high, additions of Mo increased chickpea yield, this is probably due to the fact that there was a low availability of Mo which originated because of a low soil pH (Sillanpää, 1972; Sims, 2000; Ahlawat *et al*., 2007). The total Mo that is in the soil apparently does not always represent the amount of Mo available to the plants. The foliar Mo applications increased chickpea yield, this is in accordance with Bhanavase and Patil (1994) and Singh and Singh (1994), but not in accordance with Braga and Vieira (1998), who did not find any increase in yield but their experiments were carried out using field conditions and the application of Mo was carried out later (57-60 days after emergence). Also, Yanni (1992) found that there was an increase in seed yield when Mo was applied to the soil, but the soil was inoculated with *Rhizobium*.

There was a low significant Zn × B interaction on seed yield. The highest seed yield was obtained from the $Zn_4 \times B_2$ treatment. B should only be applied when Zn is applied as well, if B is applied without an application of Zn the plant yield decreases slightly. Jahiruddin (2008) recorded that the application of Zn had an influence on B supplement on chickpea, although the experiment was conducted using calcareous soil. There was also a low significant Zn × Mo interaction on seed yield which has also been recorded by Bozoglu *et al*. (2007). The highest seed yield was obtained from the $Zn_4 \times Mo_2$ treatment. The crop yield increased when the application of Zn was increased, and the crop yield increased even more when both the Zn and the Mo were applied at the same time. There was no interaction was between B and Mo; this is contrary to the results of Shil *et al*. (2007) in which the combined application of both B and Mo were found superior to their single application. There was also a low significant Zn × B × Mo interaction on seed yield. The highest seed yield was obtained from the $Zn_4 \times B_2 \times Mo_2$ treatment. The beneficial effect of the combined application of these micronutrients (Zn, B and Mo) has been reported with chickpea that grow in calcareous soil (Jahiruddin, 2008) and has also been reported French bean (Kushwaha, 1999). The application of Zn was more efficient when it was applied with B and Mo.

As final conclusions, this study shows that both soil Zn and B applications as well as the foliar application of Mo, under pot conditions with acidic soils at high moisture availability, increase total DM and seed yield due to an increase in the number of pods per plant, principally. High rates of Zn can cause reduction in yield. The combined application of Zn, B and Mo provides a beneficial effect on seed yield; the Zn application was more efficient when it was applied with B and Mo. Soil Zn and foliar Mo applications improve harvest index. Finally, the number of pods per plant is the most influential yield component and the yield component that is most closely correlated with seed yield.

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