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GENOTYPE AND LIMING IMPACTS ON BORON AND MOLYBDENUM STATUS IN MAIZE

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Liming experiment with hydrated lime (73% CaO + 2-3% MgO + 21% water) in the amounts 0, 5 and 20 t ha^{-1} was conducted in spring 2006. Six domestic maize hybrids (B1 = Os298P, B2 = Tvrtko303, B3 = Os444, B4 = Os499, B5 = Os552 and B6 = Os596) were sown at beginning of May (basic plot 24 m^2). The ear-leaf of maize was collected at flowering and grain in maturity of the 2006 and 2007 growing seasons. Boron and molybdenum in the samples were determined by the ICP-OES method. The growing season 2006 was favorable for maize growth, while 2007 characterized stress due to drought and high air-temperatures. B and Mo concentrations (2-yr means) in grains were considerably lower for 12-fold (B) and close to two-fold (Mo) in comparison with the leaves. Leaf-Mo in 2007 was about 50% lower in comparison with 2006 (0.310 and 0.149 mg Mo kg^{-1}, respectively). Mean differences among the hybrids (mg kg^{-1}) were as follows: leaf-B 9.3 (B1) to 21.4 (B3) and grain-B 1.00 (B2) to 1.50 (B3); leaf-Mo 0.157 (B5) to 0.361 (B3) and grain-Mo 0.109 (B5) to 0.137 (B1). As affected by liming leaf-Mo were increased compared with the control 2.5-fold and 4.1 fold, grain-Mo 3.8-fold and 4.6-fold, for 5 and 20 t ha^{-1} lime, respectively. Liming effects on B concentrations in maize were considerably lower, because non-significant differences for leaf-B in 2006 and decrease for 36% in 2007 were found.

Key words: boron, molybdenum, grain, leaf, liming, maize hybrids

INTRODUCTION

Maize is the most important crop, which is widely grown under subtropical and temperate climatic regions. In general, climate in Croatia and wide region area is favorable for maize

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growing with adequate heat, mainly unfavorable precipitation distribution, while soils are relative good physical and chemical properties (KOVACEVIC *et al*., 2009; PAVLOV *et al*., 2008). Improvement of soil by liming and adequate fertilization has been resulted mainly by increases of maize yields and affected on nutritional status of maize (JURKOVIĆ *et al*., 2006; KOMLJENOVIĆ *et al.,* 2010; KOVAČEVIĆ and RASTIJA, 2010; KOVAČEVIĆ *et al.,* 2007, 2008; RASTIJA *et al.,* 2006). Maize and other cereals have relative low boron (B) and molybdenum (Mo) requirements. The B and Mo demands of plants are different depending on species and cultivars. B deficiency has been reported worldwide in maize, for example in the United States, China, India, Zimbabwe, Turkey and Switzerland. In B-deficient maize, poor grain- setting can result in barren cobs, and this was attributed to silks being non-receptive (GUNES *et al.,* 2011; LORDKAEW *et al*., 2011). In most plant species the B requirement for reproductive growth is much higher than for vegetative growth. Although maize appears to have a lower requirements for Mo, widespread Mo deficiency were found on Oxisols in Zimbabwe (BERGMANN, 1992; MENGEL and KIRKBY, 2001). Mo is essential component of two major enzymes in plants nitrate reductase and nitrogenase. Mo is also required in the synthesis of ascorbic acid and is implicated in making iron physiologically available in plants (HAQUE, 2012). Aim of this study was testing impacts of liming and genotype on B and Mo status in maize plants, while cadmium (Cd) status were elaborated in the previous study (KOVAČEVIĆ *et al*., 2011).

MATERIALS AND METHODS

The field experiment

Liming experiment with hydrated lime (73% CaO + 2-3% MgO + 21% water) in the amounts 0, 5 and 20 t ha^{-1} was conducted in term April, 19, 2006 on Rakitovica (Osijek-Baranya County) acid soil. Size plot of liming (the factor A) was 414 m^2 (23 x 18 m). Each plot of liming was divided in four sub-plot for receiving four replicates in level of maize genotype. Six domestic maize hybrids originating from Agricultural Institute Osijek (the factor B: B1 = Os298P, B2 = Tvrtko303, B3 = Os444, B4 = Os499, B5 = Os552 and B6 = Os596) was sown at beginning of May (basic plot of the hybrid 24 m^2). Practical details of the experiment were elaborated in the previous study (KOVACEVIC *et al*., 2011).

Sampling, chemical and statistical analysis

The ear-leaf samples of maize (20 leaves in mean sample) were collected at flowering (beginning of silking) and grain (ten cobs) in maturity stages from each basic plot. The total amount of boron and molybdenum in the leaf and grain samples, after microwave digestion using concentrated HNO_3+H_2O_2, was measured by the ICPAES technique by Jobin-Yvon Ultrace 238 ICP-OES spectrometer in the laboratory of the Research Institute for Soil Science and Agricultural Chemistry (RISSAC) of Hungarian Academy of Science and Arts in Budapest, Hungary. The data were statistically analyzed by ANOVA and treatment means were compared using t-test and LSD at 0.05 and 0.01probability levels.

Soil and weather characteristics

The experiment was conducted on acid soil (pH 1n KCl 4.34) and based on the high Hy value (hydrolitical acidity 5.19 cmol kg^{-1}) liming is recommend. This soil was low in humus (1.93%) and moderate supplied by plant available phosphorus and potassium determined by AL-method (13.0 P_2O_5 and 13.8 mg K_2O 100 g^{-1}).

The growing season 2007 was unfavorable for maize growing because of drought and high air-temperatures. In the 4-month period May-August 2007 precipitation in Osijek was only 162 mm or 40% lower than 30-year mean, while, air-temperature was for 2.2 ^{o}C higher, while analogical comparison for 2006 was 319 mm and air-temperature for 0.4 ^{o}C higher. Under these conditions mean grain yield of maize in the experiment was 4.83 t ha^{-1} or 2.4 fold lower than in the favourable 2006 growing season (KOVACEVIC *et al*., 2011).

RESULTS AND DISCUSSION

The B contents of monocotyledons are about 2 to 6 mg kg^{-1} and it is much lower than that of dicotyledons which mainly have B contents of 20-60 mg kg^{-1} (BERGMANN, 1992). For example, B levels (mg kg^{-1}) in different plants growing on the same site were 2.3 (barley), 3.3 (wheat), 5.0 (maize), 13.0 (potato), 25.0 (alfalfa), 37.2 (soybean) and 75.6 (beet) according to BERTRANDT *et al*., cited in BERGMANN, 1992). Also, BERGMANN (1992) evidenced adequate ranges of 6 - 15 mg B kg^{-1} and 0.15 to 0.50 mg Mo kg^{-1} contents in dry matter of leaves opposite ear of maize. The Mo concentrations in plants are mainly low in range between 0.5 and 5.0 mg Mo kg^{-1}. Mo deficient plants contain only 0.02 to 0.20 mg Mo kg^{-1}. HAQUE (2012) reported that Mo concentrations in plants vary from less than 0.1 ppm to more than 30 ppm. However, typical plant concentrations range between 0.1 and 0.2 ppm Mo. It is believed that crops will respond to Mo if they contain less than 0.1 mg Mo kg^{-1}.

According these criteria leaf-B status in our study was in adequate range from 7.9 to 25.6 mg B kg^{-1}. However, leaf-Mo concentrations were inadequate on unlimed treatment in both years, especially under drought conditions of the 2007 growing season (Table 1).

The growing season had especially considerable effects on leaf-Mo concentrations because in 2007 leaf-Mo was about 50% lower in comparison with 2006 (means 0.310 and 0.149 mg Mo kg^{-1}, respectively). However, leaf-B was practically independent on growing season because averages leaf-B were 16.2 and 14.7 mg B kg^{-1}, for 2006 and 2007, respectively. Grain-B and grain-Mo were more stabile properties (means 1.33 and 1.23 mg B kg^{-1}, 0.138 and 0.108 mg Mo kg^{-1}, for 2006 and 2007, respectively) with regards of growing season effects in comparison with status of these elements in the leaves (Table 1).

Mean differences among the hybrids (Table 1) were for leaf-B from 9.3 (B1) to 21.4 (B3) and for grain-B from 1.00 (B2) to 1.50 (B3) mg B kg^{-1}.The B1 hybrid had in both years significant lower leaf-B (2-yr mean 9.3 mg B kg^{-1}) compared with remaining five hybrids (2-yr mean 16.8 mg B kg^{-1}). Also, B3 had in both years the highest leaf-B (2-yr mean 21.4 mg B kg^{-1}) and it was in both years significantly higher from remaining five hybrids (2-yr mean 14.3 mg B kg^{-1}). The B2 hybrid had in both years the lowest grain-B compared with remaining five hybrids (mean 1.35 mg B kg^{-1}), while the highest grain-B were in B3 and B5 were found (1.50 and 1.47 mg B kg^{-1}, respectively).

Differences of Mo status among the hybrids were from 0.157 (B5) to 0.361 (B3) mg Mo kg^{-1}in leaves and from 0.109 (B5) to 0.137 (B1) mg Mo kg^{-1}in grains. In both years the B3 hybrid had the highest leaf-Mo and it was considerably higher compared with remaining five hybrids (mean 0.203 mg Mo kg^{-1}). However, the B5 and B6 had in both year low leaf-Mo (mean 0.106 mg Mo kg^{-1}). Growing season effects had very specific because in 2007 differences among the hybrids from were non-significant. The B3 and B1 hybrids in 2006 had significantly higher grain-Mo (mean 0.156 mg Mo kg^{-1}) than remaining four hybrids (mean 0.129 mg Mo kg^{-1}). The lowest grain-Mo was found in grain of B5 hybrid in both years (Table 1).

Table 1 . Impacts of liming and genotype on boron and molybdenum concentrations in maize

The experiment Rakitovica: hydrated lime (the factor A) and genotype (the factor B) effects									
Maize hybrid (the factor B)		Boron and molybdenum concentrations in dry matter							
		The ear-leaf at silking stage				Grain at maturity			
		Lime (t ha^{-1})			Mean B	Lime (t ha^{-1})			Mean B
		A1 0	A2 5	A3 20		A1 0	A2 5	A3 20	
		Boron (mg B kg^{-1}): the growing season 2006							
B-1	Os298P	7.9	7.9	11.5	9.1	1.53	1.53	1.10	1.39
B-2	Tvrtko 303	17.2	15.6	16.6	16.5	1.27	1.04	0.77	1.03
B-3	Os444	24.4	23.7	23.3	23.8	1.79	1.77	1.21	1.59
B-4	Os499	20.1	16.2	17.7	18.0	1.58	1.23	1.09	1.30
B-5	Os552	17.3	20.2	15.8	17.8	1.77	2.01	0.87	1.55
B-6	Os596	12.9	13.5	10.5	12.3	1.36	1.31	0.78	1.15
Mean A		16.6	16.2	15.9		1.55	1.48	0.97	
	LSD 5%	A: ns	B: 2.0	AB: 3.4		A: 0.11	B: 0.16	AB: 0.27	
	LSD 1%		2.7	ns		0.15	0.21	0.37	
		Boron (mg B kg^{-1}): The growing season 2007							
B-1	Os298P	10.7	9.4	8.5	9.5	1.13	1.15	1.05	1.11
B-2	Tvrtko 303	16.3	12.5	10.6	13.1	0.97	0.78	0.85	0.97
B-3	Os444	25.6	19.1	12.1	18.9	1.26	1.30	1.65	1.40
B-4	Os499	17.8	19.6	11.7	16.3	1.36	1.41	1.16	1.31
B-5	Os552	17.0	18.8	11.0	15.6	1.42	1.36	1.39	1.39
B-6	Os596	15.6	16.3	12.1	14.7	1.36	1.16	1.34	1.29
Mean A		17.2	16.0	11.0		1.25	1.19	1.24	
	LSD 5%	A: 2.0	B: 2.5	AB: ns		A: ns	B: 0.11	AB: 0.19	
	LSD 1%	2.7	3.5				0.15	ns	
		Molybdenum (mg Mo kg^{-1}): the growing season 2006							
B-1	Os298P	0.060	0.323	0.647	0.343	0.043	0.173	0.240	0.152
B-2	Tvrtko 303	0.080	0.317	0.533	0.310	0.040	0.157	0.197	0.132
B-3	Os444	0.233	0.453	0.700	0.462	0.053	0.187	0.240	0.160
B-4	Os499	0.067	0.277	0.463	0.269	0.047	0.150	0.187	0.128
B-5	Os552	0.120	0.320	0.220	0.220	0.050	0.163	0.150	0.121
B-6	Os596	0.113	0.330	0.320	0.254	0.057	0.183	0.167	0.136
Mean A		0.112	0.337	0.481		0.048	0.169	0.197	
	LSD 5%	A: 0.049	B: 0.069	AB: 0.120		A: 0.01	B: 0.02	AB: 0.04	
	LSD 1%	0.067	0.094	0.162		0.02	0.03	0.05	
		Molybdenum (mg Mo kg^{-1}): the growing season 2007							
B-1	Os298P	0.040	0.060	0.217	0.106	0.030	0.190	0.150	0.123
B-2	Tvrtko 303	0.060	0.180	0.450	0.230	0.030	0.120	0.240	0.133
B-3	Os444	0.077	0.180	0.520	0.259	0.030	0.110	0.167	0.102
B-4	Os499	0.070	0.103	0.220	0.131	0.030	0.127	0.163	0.107
B-5	Os552	0.067	0.087	0.130	0.094	0.030	0.120	0.137	0.096
B-6	Os596	0.097	0.067	0.060	0.074	0.030	0.113	0.130	0.091
Mean A		0.068	0.113	0.266		0.030	0.130	0.164	
	LSD 5%	A: 0.028	B: 0.040	AB: 0.069		A: 0.04	B: ns	AB: ns	
	LSD 1%	0.038	0.054	0.092		0.06			

Liming had considerable effects on both leaf-Mo and grain-Mo in maize. As affected by liming leaf-Mo were compared with the control increased 2.5-fold and 4.1 fold, grain-Mo 3.8-fold and 4.6-fold (2-yr averages), for 5 and 20 t ha^{-1} lime, respectively (Table 1). Also, in our earlier study was found considerable effects of liming on leaf-Mo status in maize (2-year means: 0.56 and 1.02 mg Mo kg-1, for 0 and 20 t ha^{-1} lime application, respectively), while leaf-B was independent on liming (ANDRIĆ *et al.*, 2012).

In general, concentrations of B and Mo (2-yr means) in maize grain were considerably lower for 12-fold (B) and close to two-fold (Mo) in comparison with their concentrations in the leaves. Liming effects on B concentrations in maize were considerably lower, because non-significant differences for leaf-B in 2006 and decrease for 36% in 2007 were found. However, for this effects were needed the higher applied lime rate. Also, specific response of liming on grain-B was found in both years because of grain-B decrease for 37% and non-significant differences, for 2006 and 2007, respectively.

In our earlier study (KOVAČEVIĆ *et al.*, 2011) liming effects on decreasing leaf-Cd in maize was found in both years (2-year means: 0.095 and 0.066, for the control and mean of two liming treatments, respectively). Considerable difference of leaf-Cd was found among the maize hybrids and it was in range from 0.040 to 0.160 mg Cd kg^{-1}. Two hybrids (Os298P and Tvrtko 303) separated from remaining four hybrids by the higher leaf-Cd (2-year means 0.141 and 0.043 mg Cd kg^{-1}, respectively). However, these differences are responsible for possible harmful dietary effects only in case of using these hybrids as silage maize because grain-Cd in maize was considerably lower and bellow detection limit of the applied method (<0.02 mg Cd kg^{-1}) and without effects on food contamination . Two high-leaf Cd B1 and B2 hybrids had the lower leaf-B (mean 12.05 mg B kg^{-1}) compared with remaining four hybrids (17.20 mg B kg^{-1}), while leaf-Mo concentrations were similar (means 0.248 and 0.220 mg Mo kg^{-1} respectively). KRALJEVIC-BALALIĆ *et al.*, (2009) tested under field conditions during two growing seasons the variability of Cd content in the leaves at heading stage of 30 wheat cultivars originating from different parts of the world. Differences of Cd contents among the genotypes were from 0.685 (*Kalyan Sona*) to 3.035 (*Timgalen*) mg Cd kg^{-1}.

Soil reaction is one of the most important factors affecting the availability of B in soils. With that regards, under high pH conditions B becomes less available to plants. For this reason, secondary effects of liming including immobilization of soil soluble B. Also, soil moisture and texture, humidity, temperature and genotype are additional factors affecting B uptake by plants (AREF, 2011). B availability is also related to weather characteristics in individual growing season. Deficiency appears to be more prevalent in dry summers (MENGEL and KIRKBY, 2001). In our study, liming considerably affected on decreases of leaf-B for 36% in 2007 and grain-B for 38% in 2006.

Boron is beneficial or essential for humans and acceptable safe range of population mean B intake for adults is 1 to 13 mg day^{-1}. Several reports indicate that many people consume bellow 1 mg day^{-1}, the lower limit of the safe range given (NIELSEN, 2002).

GUNES *et al.,* (2011) reported that B deficiency is widespread in the Anatolia region of Turkey. In 2-yr field experiment by application of five rates of B fertilizer up to 12 kg B ha^{-1}, the authors concluded that addition of 7.7 kg B ha^{-1} was sufficient to elevate soil B to non-deficient levels. This amount of B fertilization resulted by average soil B content 1.02 mg kg^{-1}, maize leaf B content 20.61 kg^{-1} and shoot B content 13.43 mg kg^{-1}. Leaf-B contents in our study were only in the B3 hybrids (the growing season 2006) above this level.

LORDKAEW *et al.,* (2011) tested response of maize to B application in a series of sand culture trials. By addition of 20 μM B (B20 treatment) B20 plants produced 410 grain ear^{-1}, compared with 0.4 for B0 plants (no added B). Pollen from B20 plants applied to B0 silk produced almost no grains, while pollen from B0 on B20 silk increased the number of grains to 37% of the 452 grains $plant^{-1}$ produced from B20 pollen to B20 silk. Tassels, silk and pollen of B0 plants contained 3-4 mg B kg^{-1} compared with twice or more B in these reproductive tissues in B20 plants.

For many species there is only a narrow range in critical tissue concentrations between B deficiency and B toxicity. High levels of B in the environment derive from natural and anthropogenic sources. Industry and waste effluents and irrigation practices are main sources of anthropogenic B pollution (BERGMANN, 1992; MENGEL and KIRKBY, 2001).

BRDAR *et al*. (2008) found considerable hereditary effects on wheat seedlings response to excess of boron as affected by treating with boric acid up to 150 mg l^{-1}. Selection criterion among 12 wheat genotypes was root growth suppression in the presence of boron, which varied between 15.2% (*Apache*) and 46.3% (*Renan*). High concentrations of B are often found in association with saline soils, such as solonchaks and solonetz, which are common in Vojvodina (MILJKOVIĆ, 1960, cited by BRDAR *et al.,* 2008).

Mo availability is also strongly pH dependent and increasing with decreasing pH. As the pH fall the Mo soil solution concentrations decreases. As MoO anion is strongly adsorbed by Fe and Al oxides, which markedly increase at low pH, possible Mo deficiency is normally a problem on acid soils. The Mo concentration in soil increase 100-fold for each unit increase in pH. Besides soil pH, soil organic matter, clay contents and soil wetness are important factors of Mo availability. Wet soils tend to have high organic matter content and large amounts of Mo that may be available. In our study, considerably lower Mo concentrations in maize were found under drought conditions of the 2007 growing season and our results are in accordance with the earlier knowledge regarding soil moisture impacts on Mo uptake in plants. Nutrient interactions are also responsible for Mo availability. Uptake of Mo by plants is usually enhanced by soluble P and decreased by available S and Cu (BERGMANN, 1992; HAQUE, 2012). However, KOMLJENOVIĆ *et al.*, (2006) found significantly decreases of leaf and grain Mo concentrations as affected by ameliorative P fertilization under acid soil conditions in Potkozarje area of Bosnia and Herzegovina. Although maize appears to have a lower requirements for Mo, widespread Mo deficiency were found on Oxisols in Zimbabwe. Seeds treatment with Mo in combination with liming resulted by significant increases of maize yields (MENGEL and KIRKBY, 2001).

CONCLUSION

The growing season, liming and genotype had less or more considerable effects on B and Mo status in maize. Under drought stress of the 2007 growing season leaf-Mo was 50% lower in comparison with normal weather conditions of 2006. As affected by liming B concentrations in maize decreased for 20% and Mo concentrations increased more than 4-fold. In general, hereditary effects on B and Mo status in maize was higher in leaves compared to grain. In the both years, the maize hybrid Os298P had significant lower leaf-B and the hybrid Os444 the highest leaf-Mo compared with remaining five hybrids. These findings could be useful for improvement of food quality for animal and humans with aspects of B and Mo supplies.

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UTICAJ GENOTIPA I KALCIZACIJE NA STANJE BORA I MOLIBDENA U KUKURUZU

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Izvod

Ogled kalcizacije primenom hidratnog kreča (73% CaO + 2-3% MgO + 21% vode) u količinama 0 (kontrola), 5 i 20 t ha^{-1} postavljen je u proleće 2006. Šest domaćih hibrida kukuruza (B1 = Os298P, B2 = Tvrtko303, B3 = Os444, B4 = Os499, B5 = Os552 i B6 = Os596) posejani su početkom maja (osnovna parcela 24 m^2). List ispod klipa je uzet u cvatnji a zrno u fazi zrelosti vegetacije 2006. i 2007. Bor (B) i molibden (Mo) u uzorcima su određeni ICP-OES metodom. Vegetacaija 2006. bila je povoljna za kukuruz, dok je 2007. bila izražena suša u kombinaciji s visokim temperaturama. Koncentracije B i Mo (2-god. prosek) u zrnu bile su za 12 puta (B), odnosno blizu 2 puta (Mo) manje u poređenju s listom. Mo u listu 2007. bio je za 50% niži od onoga 2006. (0.310 i 0.149 mg Mo kg^{-1}). Prosečne razlike između hibrida (mg kg^{-1}) bile su sledeće: B u listu od 9.3 (B1) do 21.4 (B3) i B u zrnu od 1.00 (B2) do 1.50 (B3); Mo u listu od 0.157 (B5) do 0.361 (B3) i Mo zrnu od 0.109 (B5) do 0.137 (B1). Usled kalcizacije s 5, odnosno 20 t ha^{-1}, Mo u listu je povećan prema kontroli za 2,5 puta, odnosno 4.1 puta, a Mo u zrnu 3.8 puta, odnosno 4.6 puta. Uticaj kalcizacije na koncentracije B u kukuruzu bio je značajno slabiji, jer u 2006. nisu ustanovljene signifikantne razlike, a u 2007. je kalcizacijom koncentracija B smanjena u proseku za 36%.

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