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ORIGINAL PAPER

MAGNESIUM AND CALCIUM DISTRIBUTION IN MAIZE UNDER DIFFERENTIATED DOSES OF MINERAL FERTILIZATION WITH PHOSPHORUS AND POTASSIUM*

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Abstract

The study was conducted to evaluate the effect of imbalanced mineral fertilization with phosphorus and potassium on calcium and magnesium management in maize at the critical growth stages (BBCH 17 and BBCH 65), as well as the content and accumulation of these nutrients when maize was fully ripe (BBCH 89). A single factor field experiment was established to investigate 8 treatments: the absolute control, without one of the main nutrients (P – WPN or K – WKN), reduced amount of phosphorus and potassium (to 25% – W25 and to 50% – WP50, WK50) and recommended rates of basic nutrients (NPKMg – W100 and NP*KMg, P* – P* as PAPR – W100 PAPR). The results showed no significant effects of mineral fertilization on the calcium and magnesium content in the leaves of maize examined at BBCH 17. Irrespective of mineral fertilization doses applied, Ca and Mg concentrations in maize leaves remained below the standard values. At the next examined maize growth stage (flowering – BBCH 65), magnesium malnutrition was observed as well, whereas the calcium concentration was at the luxury level. Differentiated fertilization with phosphorus and potassium significantly altered Ca and Mg concentrations in the ear leaf of maize at flowering. From the beginning of BBCH 17 stage until the end of maize growth, an antagonism between magnesium and potassium was observed, seen especially distinctively in WKN treatment (no potassium applied). Correlation analysis

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showed significant relationships between grain yield and plant nutrition with magnesium and calcium at maize flowering. Notwithstanding fertilizer treatments, grain yield of fully ripe maize was determined by the total accumulation of magnesium. More than 50% of Mg total uptake was accumulated in grain. On the other hand, calcium was accumulated mostly in maize stems and leaves, which jointly accumulated 87% of this nutrient.

Keywords: imbalanced fertilization, maize parts, magnesium and calcium accumulation.

INTRODUCTION

Among cultivated cereals, maize has the highest yield potential, which can be successfully obtained only if cultivation sites are rich in essential mineral nutrients, including calcium and magnesium. The latter is a key mineral nutrient needed for balanced nutrition in all crop plants. Yield forming functions of magnesium and calcium differ from each other, which results in differentiated effects of these nutrients on the plant growth during the growing period (Maguire, Cowan 2002, White, Broadley 2003). Accumulation of magnesium in plants is closely associated with plant nitrogen management, and a particularly strong relationship as such is manifested after the flowering stage (Barbittin et al. 2005, Hirel et al. 2007). Literature data indicate that inclusion of magnesium into a fertilizer system enhances the utilization of nitrogen by maize (Potarzycki 2010*a*, Szulc 2010). Magnesium is indispensable in the processes of protein hydrolysis in plant vegetative organs as well as for the transfer of assimilation products from leaves to ears. This nutrient takes part in photosynthesis (prolongs the stage of green leaves) and transportation of proteins from plant vegetative organs to seeds or kernels (Cakmak, Kirkby 2008). The yield forming effect of magnesium is particularly evident under the conditions of insufficient supply of nitrogen to plants (Grzebisz 2013). Calcium regulates osmotic and ionic processes in cell membranes, and magnesium works as a cofactor in enzymatic reactions (White, Broadley 2003). The effects of calcium and magnesium deficiency are evident in plants growing on excessively acidic soils, with a low Ca content caused by the leaching of Ca^{2+} cations or with low cation-exchange capacity (CEC), as well as under the conditions of aluminum toxicity to the plant root system (Rengel, Elliot 1992, Ryan et al. 1994).

The aim of the present study was to evaluate the effects of differentiated mineral fertilization with phosphorus and potassium, applied at constant nitrogen and magnesium levels, on calcium and magnesium management in maize at the critical growth stages, as well as to assess the content and accumulation of these nutrients in fully ripe maize.

MATERIALS AND METHODS

A field experiment on maize, variety *Veritis* (FAO: 230-240), was conducted in 2007-2011, on a private farm situated in the village Wieszczyczyn (52°02′ N 17°05′ E) in Wielkopolska Province (western Poland). The trial was performed in a randomized complete block design with 4 replications. The analyses were carried out on 8 fertilizer treatments with different levels of phosphorus and potassium applied, i.e.: the absolute control; without one of the main nutrients (P – WPN or K – WKN); decreased doses of phosphorus and potassium (to 25% – W25 and to 50% WP50; WK50) and recommended doses of basic nutrients (NPKMg – W100 and NP*KMg, P* – P* as PAPR – W100PAPR). During the observation period (2007/2008/2009/2010/2011), the recommended levels of maize mineral fertilization were as follows: kg N ha^{-1} 120/120/150/150/150; kg P ha^{-1} 35/26/26/26/26; kg K ha^{-1} 100/125/133/125/125 and 16 kg Mg ha^{-1}. Phosphorus, potassium and magnesium were applied once in the fall, whereas nitrogen was applied in two doses: 70% before maize sowing and 30% – when maize was at the stage of 4 fully developed leaves. Winter wheat was the crop preceding maize in all the trial years. Details on the then soil and weather conditions as well as maize yields are provided in the paper by Bąk and Gaj (2016).

Assessments of the calcium and magnesium content in maize plants was conducted 3 times during the growing season, when maize was at the following critical growth stages: BBCH 17, BBCH 65 and BBCH 89. During two assessment dates (7 unfolded leaves and flowering), nutrient concentrations were tested in 5 maize leaves, randomly collected from each experimental plot. In the flowering maize (BBCH 65), nutrient concentrations were assessed in the ear leaf. When maize was fully ripe (BBCH 89), calcium and magnesium concentrations were assessed in the following maize organs: leaves, stems, ears, cob cores, husks and grains. Ca and Mg concentrations were examined in plant material after dry mineralization (640°C) followed by dissolving the ash obtained in 33% HNO_3. Magnesium was determined using flame atomic absorption spectrometry, and calcium concentration – by flame photometry. Nutrient uptake was computed from the multiplication of dry weight value by the value of a given nutrient concentration in the plant organs observed (dry weight data available from the authors – unpublished data).

Statistical analysis

The influence of the experimental factor on nutrient accumulation and concentration in maize cultivated under different doses of mineral fertilization with P and K was tested using a 2-way ANOVA (mixed-effects model). To attain more detailed information, a 2-way ANOVA and the Tukey's HSD test were carried out. A detailed description of the model used in the present study was described by Bąk and Gaj (2016), Bąk et al. (2016).

The acquired data on Ca and Mg concentrations in the maize organs examined were compared using the graphical representation in heat maps, where the concentrations of nutrients with reference to the treatment applied (2D variables) were represented in colours. Cluster analysis allowed us to group the treatments based on the concentration of the nutrient tested in a given maize organ, so as to demonstrate the strongest and weakest relationships (within a specified group and among the groups, respectively). The Ward's hierarchical clustering and the Euclidean distance were used to arrange the dendrograms. Causal relationships between Ca and Mg concentrations, uptake, unit uptake and maize grain yield were tested with correlation coefficients.

RESULTS AND DISCUSSION

Magnesium and calcium concentrations at BBCH 17 and BBCH 65

The effect of the experimental factor on the magnesium and calcium content in maize was varied, depending on the nutrient examined and the plant growth stage. At BBCH 17, no significant differences in the Mg and Ca concentrations were observed with regard to the different doses of phosphorus and potassium (Bąk, Gaj 2016). The content of calcium and magnesium ranged narrowly, and amounted to 2.65-2.92 g kg^{-1} and 1.63-1.84 g kg^{-1}, respectively (Figures 1, 2). Notwithstanding the treatment applied, the values obtained were below the standard thresholds (Ca: 5.1-16 g kg^{-1}; Mg: 3-6 g kg^{-1}) determined by Schulte and Kelling (2000). The chemical form of phosphorus applied as fertilizer (treatments: W100 and W100 PAPR) had no significant effect on the differences between the Ca and Mg concentrations in maize leaves. The reasons behind Ca and Mg deficiency in the plant are complex

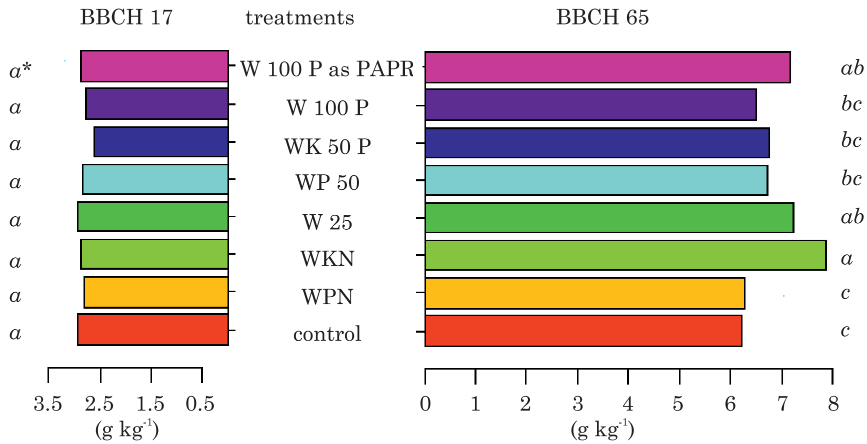


Fig. 1. Calcium concentration in maize leaves at the critical growth stages, depending on differentiated P and K fertilization (g kg^{-1} DM).
* Means with the same letter are not significantly different; $\alpha = 0.05$ (the Tukey's test)

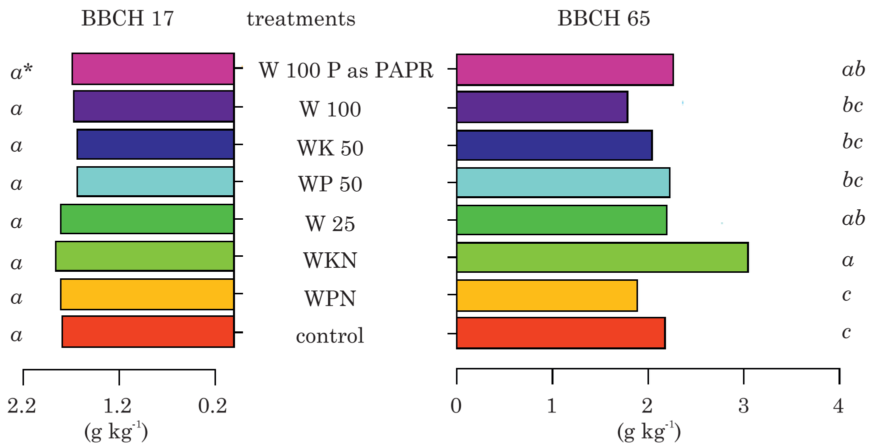


Fig. 2. Magnesium concentration in maize leaves at the critical growth stages, depending on differentiated P and K fertilization (g kg^{-1} DM).
* Means with the same letter are not significantly different; α = 0.05 (the Tukey's test)

and associated with the nutrient shortage in soil, acidic soil reaction, surplus of other cations, as well as malfunctioning processes of nutrient uptake or nutrient transport in growing plants (Fageria 2001, Shaul 2002).

Both calcium and magnesium are taken up by the youngest root tissues, whose growth can be disturbed by toxic aluminum Al^{3+} in the soil (Reid et al. 1995). Negative effects of aluminum on crop plants are aggrevated by the increasing oxidation of Al cations (Bennet, Breen 1991). The initial plant response to toxic effects of Al^{3+} is expressed in the root, which shows inhibited performance of its cap and meristematic cells (Ryan et al. 1994). Calcium is not remobilized in the plant but is gradually accumulated in biomass produced during the plant growth.

During the present study, maize was cultivated on acidic soil (pH 4.9), which resulted in a low Mg and Ca content in the plants at early growth stages. Although the plants showed both Ca and Mg malnutrition at growth stage BBCH 17, significant relationships between maize grain yield and nutrient supply to the plants were observed in a small number of treatments and, depending on a nutrient, these were as follows: for Ca – control, W100 PAPR and for Mg – control, W100 and WK50 (Table 1).

When maize was at the stage of flowering (BBCH 65), different P and K fertilizer doses differentiated the Ca and Mg concentrations in maize ear leaves to a much larger extent than observed in the earlier maize growth stages (Figures 1, 2). Irrespective of the treatment applied, the plants showed luxury concentration of calcium, whereas the Mg concentration was below the lower threshold value, i.e.: 2 g kg^{-1}. Under the conditions of P or K fertilizer deficiency assumed in the present study, maize plants proved that fertilizer absence (especially of K) resulted in an increase of calcium and magnesium in the plant. The highest concentrations of the nutrients ana-

Table 1

Pearson's correlation coefficients between maize grain yield and content of Ca and Mg in maize leaves at the critical growth stages

Stage	nutrients	control	WPN	WKN	W25	WP50	WK50	W100	W100 P as PAPR
		Treatments							
BBCH 17	Ca	0.577*	0.144	0.295	0.443	0.284	0.315	0.368	0.456*
	Mg	0.464*	0.429	0.381	0.297	0.382	0.475*	0.596*	0.296
BBCH 65	Ca	0.647*	0.467*	0.629*	0.064	0.694*	0.540*	0.638*	0.146
	Mg	0.600*	0.482*	0.608*	0.612*	0.445*	0.589*	0.550*	0.548*

* correlation significant at $p < 0.05$

lyzed were observed in the WKN treatment (no potassium applied for 10 years), and this definitely indicates an antagonistic relationship between K and both nutrients analyzed (Ca and Mg). The lack of potassium in fertilizer treatments resulted in a decrease of this nutrient in indicative maize organs (Figure 3) and concurrent compensation of calcium and magnesium. Potassium deficiency in the leaves caused an increase in the magnesium content at both maize growth stages analyzed (Figure 2).

An increase in the plant calcium content at potassium deficiency was observed only in the ear leaf of flowering maize (Figure 1). According to literature data, intensive potassium fertilization can at times not only change the potassium concentration (TAN et al. 2012, NIU et al. 2013), but also incude either excess or scarcity of other nutrients, especially calcium and magnesium, in crop plant yields (KARLEY, WHITE 2009). An interaction between potassium and magnesium has been reported by many authors (DIEM, GODBOLD 1993, PUJOS, MORARD 1997, FAHAT et al. 2013). Magnesium ions inactively

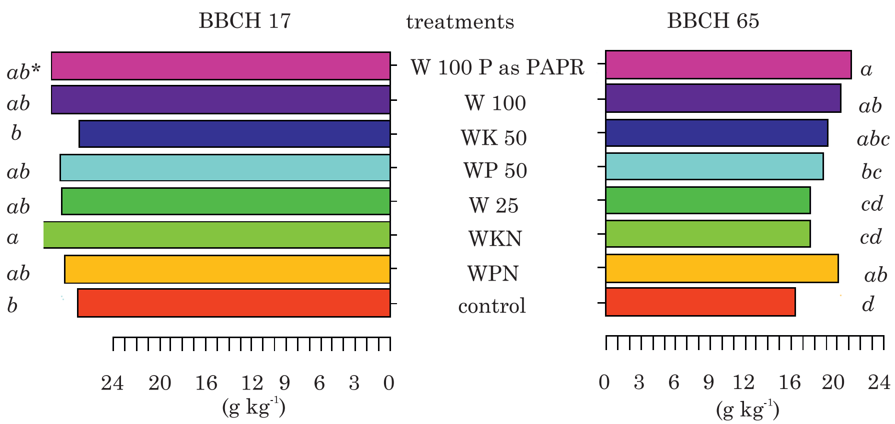


Fig. 3. Potassium concentration in maize leaves at the critical growth stages, depending on differentiated P and K fertilization (g kg^{-1} DM).
* Means with the same letter are not significantly different; $\alpha = 0.05$ (the Tukey's test)

pass through the cell membrane, with the electrochemical gradient of cation concentration, and they are instantly exposed to competition with actively absorbed potassium ions (Gierth, Mäser 2007). Secondly, Mg^{2+} ions, also passively taken in by plants, compete with calcium ions (Pfeiffer, Hager 1993). The results of correlation analyses performed separately for each treatment with regard to relationships between maize grain yield and the Mg and Ca plant nutritional status at BBCH 65, showed significant relationships for both nutrients (Table 1). Only in two treatments (W25 and W100 PAPR), no significant relationships between grain yield and Ca concentrations were observed. A strong relationship between grain yield and plant supply with Ca and Mg at BBCH 65 indicates better suitability of the flowering stage in making predictions of maize grain yield than that of BBCH 17. In view of a plant nutritional status, it is important to maintain appropriate relations between nutrients. The concentrations of individual elements change with the plant's age, and therefore, for yield prediction, it is more useful to employ the knowledge on relations between the nutrients, which provides better information on a plant's physiological status (Santa-Maria, Epstain 2001, Nebolsin et al. 2004). Correlation analyses on relationships between grain yield and nutrient pairs, such as N:K, N:Ca, N:Mg, at maize critical growth stages, for the most part showed significant correlations for N:K and N:Mg, regardless of the fertilizer treatments applied (Table 2). In the case of the N:Mg ratio, negative correlations were observed in some treatments only at BBCH 17, which confirmed earlier observations on the plant Mg nutritional status, which indicated Mg concentrations below the standard level.

Calcium and magnesium concentration at BBCH 89

When maize was fully ripe, calcium and magnesium concentrations were differentiated, depending on a nutrient, the plant organ examined and the effect of the experimental factor (Figures 4, 5). Generally, from the stage of

Table 2

Correlation analysis on relationships between grain yield and nutrient pairs at maize critical growth stages

Growth stages	Parameters	Treatments							
		control	WPN	WKN	W25	WP50	WK50	W100	W100 P as PAPR
		correlation coefficient							
BBCH 17	N:K	0.068	0.707*	0.672*	0.100	0.512*	0.606*	0.661*	-0.057
	N:Mg	-0.504*	-0.259	-0.578*	-0.573*	-0.034	-0.532*	-0.286	-0.156
	N:Ca	-0.188	0.287	0.177	-0.196	0.328	0.207	0.154	0.005
BBCH 65	N:K	0.712*	0.623*	0.355	0.768*	0.239	0.733*	0.648*	0.606*
	N:Mg	0.167	0.361	-0.001	-0.017	0.406	0.295	0.351	-0.048
	N:Ca	0.000	0.340	-0.242	0.605*	-0.254	0.268	0.095	0.396

* correlation significant at $p < 0.05$

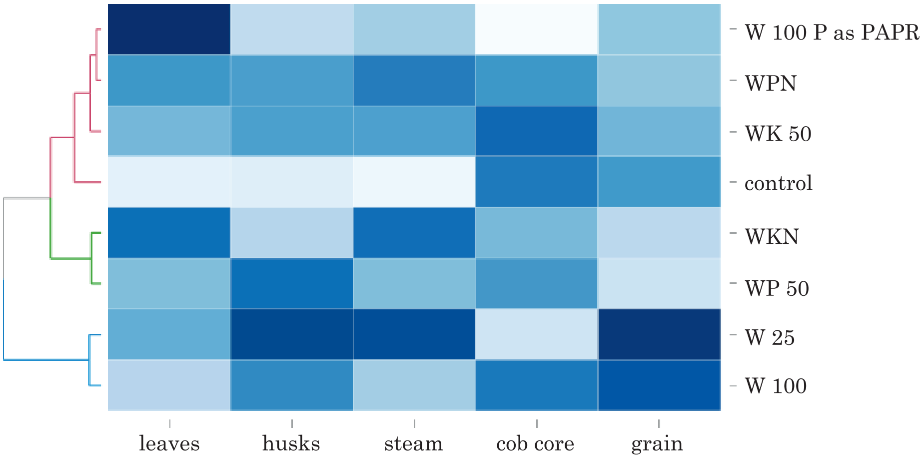


Fig. 4. Effect of P and K fertilization on Ca concentrations in maize organs (g kg^{-1} DM); BBCH 89 – fully ripe

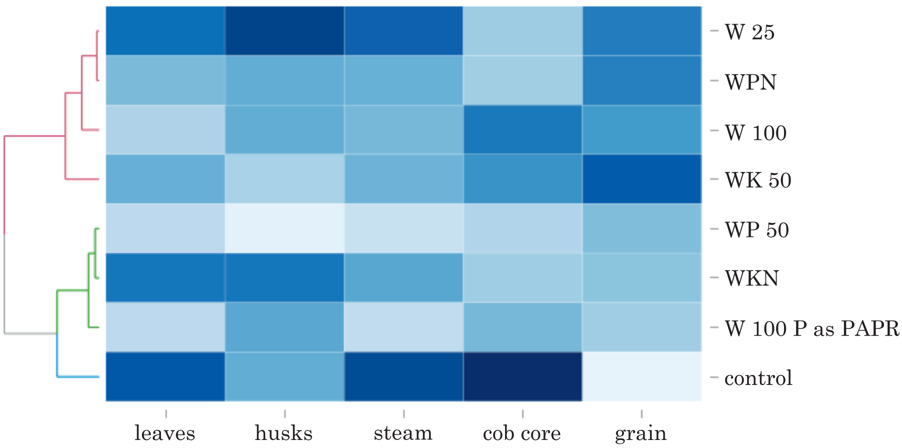


Fig. 5. Effect of P and K fertilization on Mg concentrations in maize organs (g kg^{-1} DM); BBCH 89 – fully ripe

plant vegetative growth to the stage of ripe grain, mineral nutrient concentrations decrease, and the results obtained in the present study confirmed this tendency. The concentration of calcium decreased as follows: leaves>stems>husks>cob cores>grain (Figure 4). When compared to the control, an increase in the Ca concentration owing to fertilization was observed in maize leaves, stems and husks, whereas grain and cob cores showed a Ca decreasing tendency. Significant differences in Ca concentrations as a result of the P form used as fertilizer were observed only in the case of grain. Higher Ca concentrations were observed in the W100 treatment, with single superphosphate (SSP) as a phosphorus source, than in the treatment P as PAPR.

The sequence of Mg concentrations in maize organs examined differed from that observed for Ca, and was as follows: leaves> grain>stems>husks >cob cores (Figure 5). In comparison with the control, mineral fertilization did not result in an increase in the Mg concentration in maize organs analyzed when plants were fully ripe. In each treatment investigated, a tendency was observed towards a decreasing Mg concentration. However, the study by Hejcman et al. (2013) showed no significant effect of differentiated natural as well as mineral fertilization on Ca and Mg concentrations in barley grain. The analysis of maize nutritional status showed (Figures 4, 5) that the Ca concentration in leaves of fully ripe plants ranged from 4.3 g $kg^{-1}$to 5.5 g kg^{-1}, whereas that of Mg was below the standard values (2-8 g kg^{-1}), in the same way as at earlier maize growth stages. Starting from BBCH 17 until the last growth stage, an antagonism was observed between potassium and magnesium, which was first and foremost noticeable in no potassium treatment (WKN). This relationship points to the complexity of potassium and calcium co-occurrence in the plant. Irrespective of the treatment tested, the correlation analysis of K and Mg concentrations showed negative relations, with statistical significance expressed by correlation coefficients obtained: W25 – 0.509, WKN – 0.507, W100 – 0.463. Magnesium may strongly alter Ca^{2+} and K^{+} uptake, whereas K^{+} and Ca^{2+} can limit Mg^{2+} uptake and translocation from the roots to aerial plant organs (Ohno, Gruner 1985).

Magnesium and calcium uptake at BBCH 89

Regardless of the treatment applied, mineral fertilization significantly increased Ca accumulation when compared with the control (Figure 6). On the other hand, no significant differences were found between the treatments tested. The total Ca accumulation in maize on fertilizer treated plots reached 23.5 kg ha^{-1} on average, whereas in the control it was 18 kg ha^{-1}. The experimental factor differentiated Mg accumulation more than calcium uptake (Figure 7). When compared with the control, a statistically significant increase in the total Mg accumulation was observed in the W25 and WK50 treatments (17.6% and 16.4%, respectively). Distribution of nutrients in maize organs is an important factor in the assessment of nutrient accumulation in maize final yield, and nutrient accumulation in grain (accumulation harvest index) should be especially considered. Mineral fertilization with differentiated levels of phosphorus and potassium significantly varied the structure of Mg and Ca accumulation in maize organs, except for cob cores (Figure 6). When compared with other maize organs examined, the largest part of plant magnesium (56%) was accumulated in maize grain. At the same time, the stems and leaves accumulated 20% and 5%, respectively. Comparatively the lowest Mg accumulation was observed in husks (3.5%) and cob cores (2%).

The highest amount of magnesium was accumulated by maize grain (MgHI = 57.7%) harvested from the W100 treatment (optimally balanced with reference to nitrogen), whereas significantly less magnesium (53.4%) was accumulated by grain of maize cultivated under the conditions of the

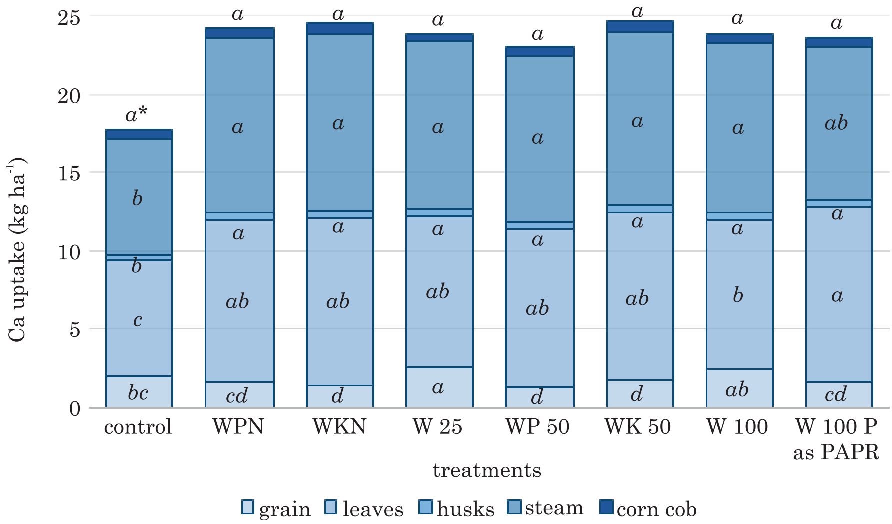
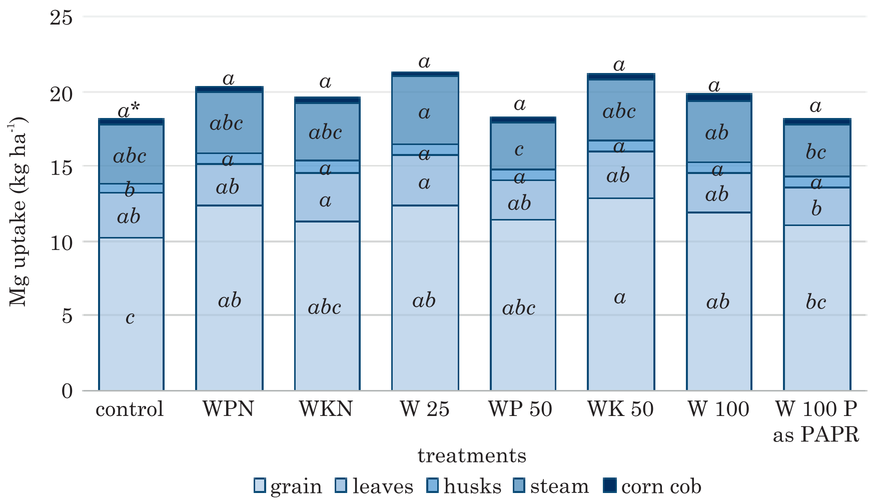


Fig. 6. Structure of Ca accumulation in maize at physiological maturity (BBCH 89), mean 2007-2011.
* Means with the same letter are not significantly different; α = 0.05 (the Tukey's test)



Fig, 7. Structure of Mg accumulation in maize at physiological maturity (BBCH 89), mean 2007-2011.
* Means with the same letter are not significantly different; α = 0.05 (the Tukey's test)

WKN treatment. Considerable accumulation of magnesium in maize grain and leaves is a sign of a strong relationship between this nutrient and the quality of grain yield obtained. Enhanced nutrient distribution among plant parts reflects the efficiency of nutrient use. Correlation analysis showed significant relationships between Mg accumulation in grain and grain yield in all the treatments tested. Irrespective of the treatments applied, a highly

Table 3

Pearson's correlation coefficients between maize grain yield and content and uptake of Ca and Mg

Parameters	Treatments							
	control	WPN	WKN	W25	WP50	WK50	W100	W100 P as PAPR
	correlation coefficient							
Ca grain content	0.233	0.477*	0.075	0.114	0.423	0.396	0.406	-0.006
Mg grain content	0.485*	0.761*	0.568*	0.280	0.746*	0.675*	0.714*	0.348
Ca grain uptake	0.336	0.705*	0.414	0.349	0.688*	0.553*	0.485*	0.334
Mg grain uptake	0.644*	0.811*	0.661*	0.543*	0.814*	0.759*	0.793*	0.516*
Ca total uptake	0.678*	0.394	0.636*	0.434	0.829*	0.453*	0.720*	0.553*
Mg total uptake	0.861*	0.939*	0.783*	0.778*	0.905*	0.880*	0.855*	0.765*

* correlation significant at $p < 0.05$

significant dependency was also found between maize grain yield and the total Mg accumulation (Table 3).

Significant relationships between grain yield and Ca accumulation in grain were observed in the WPN, WP50, WK50 and W100 treatments. The relationship between maize yield and the total accumulation of Ca looked differently, as in the majority of the treatments tested significant relationships were observed, and only the WPN and W25 treatments did not show similar patterns.

The aforesaid dependency confirms the role of magnesium in shaping maize grain yield, which has also been well documented by other authors (SZULC et al. 2008, SZULC 2010, SZULC, WALIGÓRA 2010). The relationship between nutrient uptake and yields points to the effectiveness of nutrient utilization, which is reflected in a commercial product, e.g. grains (VAN DUIVENBODEN et al. 1996).

The structure of Ca accumulation in maize organs showed a different pattern when compared with that of magnesium (Figure 6). Mean Ca accumulation in maize was decreasing as follows: stems (45%) > leaves (41%) > grain (8%) > cob cores (2.6%) > husks (2%). Grain accumulation ranged from 5.8% to 11% of the total Ca uptake, which is demonstrated by the value of CaHI obtained (Table 4). The lowest CaHI values were observed in the following treatments: WP50 (5.6%) and WKN (5.7%). Application of partially acidulated phosphate rock as a P source resulted in a significant decrease in the Ca accumulation in maize grain when compared to the W100 treatment. The difference between W100 and W 100 PAPR was 34%.

Quantitative nutritional needs of a plantation are determined by the so-called nutrient unit uptake (accumulation of a given nutrient/yield unit). In the present study, this factor was represented by maize grain mass along with the respective mass of by-products such as stems and leaves. No effects

Table 4

Calcium and magnesium harvest index and unite uptake, depending on P and K fertilization applied

Treatments	Unit uptake, nutrient*(kg t^{-1})		Nutrient harvest index (%)	
	Ca	Mg	CaHI	MgHI
Control	3.06*a***	2.89*a*	11.02*a*	55.30*ab*
WPN	2.25*a*	2.59*b*	7.00*b*	55.32*ab*
WKN	3.25*a*	2.54*bc*	7.91*b*	53.44*b*
W25	3.10*a*	2.58*b*	10.48*a*	55.53*ab*
WP50	2.99*a*	2.27*c*	5.89*b*	56.02*ab*
WK50	3.18*a*	2.53*bc*	7.37*b*	56.27*ab*
W100	2.77*a*	2.29*c*	10.32*a*	58.69*a*
W100 P as PAPR	3.17*a*	2.43*bc*	7.33*b*	53.83*ab*

* kg per 1t of grain, including concomitant amount of nutrient in vegetative parts (kg t^{-1});
** Means with the same letter are not significantly different; α = 0.05 (the Tukey's test).

were observed of the experimental factor on the Ca unit uptake, with the mean value being 3 kg Ca t^{-1} (Table 4). A different plant response to PK fertilization was observed in the case of Mg unit uptake, the value of which ranged from 2.3 kg Mg t^{-1} to 2.9 kg Mg t^{-1} (Table 4). When compared to the control, the unit uptake values were reduced in all the treatments tested. Significant differences were also observed between the following fertilizer treatments: W100, WP50 a W25,WPN.

CONCLUSIONS

1. The assessment of Mg and Ca nutritional status of maize at the critical growth stages (BBCH 17 and BBCH 65) showed inadequate Mg nutrition at both growth stages analyzed, regardless of a fertilizer treatment applied. At the same time, Ca malnutrition was observed only at the stage of 7 fully unfolded leaves.

2. The significant dependence of maize grain yield on Ca and Mg concentrations in the ear leaves was observed in maize at the stage of flowering, which indicates better prognostic applicability of BBCH 65 growth stage when compared to BBCH 17.

3. Long-term lack of potassium in fertilization resulted in the Ca and Mg increase in maize leaves, observed at both growth stages analyzed (BBCH 17 and BBCH 65).

4. Regardless of a fertilizer treatment applied, maize grain yield was significantly determined by the total accumulation of Ca and Mg in fully ripe plants.

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