

Radkowski A., Radkowska I., Rapacz M., Wolski K. 2017. Effect of foliar fertilization with magnesium sulfate and supplemental L-ascorbic acid on dry matter yield and chemical composition of cv. Egida timothy grass. J. Elem., 22(2): 545-558. DOI: 10.5601/jelem.2016.21.3.1167

ORIGINAL PAPER

EFFECT OF FOLIAR FERTILIZATION WITH MAGNESIUM SULFATE AND SUPPLEMENTAL L-ASCORBIC ACID ON DRY MATTER YIELD AND CHEMICAL COMPOSITION OF CV. EGIDA TIMOTHY GRASS*

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ABSTRACT

In 2011-2013, a field experiment was conducted at the Plant Breeding Station in Polanowice near Krakow (220 m above sea level), on degraded Chernozem developed from loess. The field experiment was set up in a randomized block design with three replications. The experimental material used in this study was cv. Egida timothy grass. The aim of the study has been to test whether L-ascorbic acid supplementing for foliar fertilization with magnesium sulfate enhances magnesium absorption by plants and what effects it has on yielding and dry matter composition of timothy grass cut and used as forage. Three levels of L-ascorbic acid fertilization were applied (10, 20 and 30 g ha⁻¹) with and without magnesium sulfate 7-hydrate. The plants were sprayed until wet, using 300 dm³ liquid per hectare. Spraying was performed 6-8 days from the onset of growth for the first harvest, and a week after cutting for the other harvests. Statistically significant improvement in yielding and forage quality was observed for the combined magnesium/ ascorbic acid fertilization. Fertilization with these two nutrients caused an increase in the total dry matter yield of timothy grass cv. Egida (together from 3 cuts), with differences reaching as much as 26% depending on the year (in the variant of combined magnesium sulfate and ascorbic acid application at 30 g ha⁻¹). Another benefit of ascorbate application was an increase in forage

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^{*} The work has been financed from funds allocated to maintaining the research potential, granted by the Ministry of Science and Higher Education.

quality (the content of crude protein, crude fat, water soluble carbohydrates and macronutrients). Best effects in terms of yielding and forage quality were obtained after a combined application of magnesium sulfate and ascorbic acid (30 g ha⁻¹). Combined fertilization with magnesium sulfate and ascorbic acid tended to produce a stronger effect after periods of dry weather, which is discussed in the study.

Keywords: timothy grass, yield, L-ascorbic acid, magnesium sulfate, forage quality.

INTRODUCTION

Foliar application of mineral and organic compounds with a biostimulatory effect is a way to improve the quantity and quality of forage.

Ascorbic acid, which is commonly found in plants, has a pivotal role in several physiological processes such as antioxidative defence, regulation of key enzymes and thus the control of cell division and expansion, growth, development and senescence (DAVEY et al. 2000, HOREMANS et al. 2000*a*, *b*). In plants, ascorbic acid is responsible for combating the stress caused by drought or excess ultraviolet radiation. Ascorbic acid not only regulates defence and survival of stressed plants by modifying the expression of genes, but it also modulates their growth by acting through phytohormones (PASTORI et al. 2003). In some species of plants, ascorbic acid is also a substrate for biosynthesis of oxalic and tartaric acids (LOEWUS 1999).

As a central element of the chlorophyll molecule, magnesium is crucial for biomass production. It is also essential for photosynthesis, protein synthesis and nuclelotide metabolism. However, plant Mg nutrition has received little attention compared to other essential macronutrients.

Magnesium has a large hydrated radius and sorbs weakly to soil colloids, predisposing it to leaching, particularly in acidic soil with low cation exchange capacity (AITKEN et al. 1999, GRZEBISZ 2011). Magnesium availability in plant crops decreases on alkaline soils (BROADLEY, WHITE 2010). Magnesium deficiency in arable soils may increase when cultivated plants are intensively harvested without Mg fertilization (GRZEBISZ 2011). It is assumed that over 50% of Poland's cropland area has a low content of available magnesium; therefore, mineral fertilization with magnesium is of great importance (FAJEMILEHIN et al. 2008). Magnesium deficiency in plants leads to nutritional disturbances, which have an effect on the yield and quality of agricultural plants (AITKEN et al. 1999), horticultural plants (TROYANOS et al. 2000, RUAN et al. 2012, YANG et al. 2012) and forest trees (Sun et al. 2001, BOXLER-BALDOMA et al. 2006, VACEK et al. 2006). Mg-deficient leaves are reported to be highly photosensitive and an increase in light intensity causes severe chlorosis and photooxidation of thylakoid constituents of these leaves by generation of reactive oxygen species – ROS (CAKMAK, MARSCHNER 1992). Mg deficiency also activates the antioxidative system in plants (TEWARI et al. 2005).

In this paper we hypothesize that combined application of ascorbic acid

and magnesium sulfate may improve forage quantity and quality of timothy grass growing under environmental stress, owing to the additive effect on biomass production and protection against ROS.

MATERIAL AND METHODS

The study, conducted in 2011–2013, was set up at the Plant Breeding Station in Polanowice (20°09' E, 50°20' N) near Kraków (220 m above sea level), on degraded Chernozem developed from loess. The chemical properties of the soil were as follows: $pH_{KCl} - 7.2$, available P – 52.0, K – 124.4 and Mg – 46.3 mg kg⁻¹ soil. Total annual precipitation during the study period (2011-2013) ranged from 390 to 631 mm (Figure 1). Mean total precipitation



Fig. 1. Total monthly precipitation at the Plant Breeding Station in Polanowice in the years 2011, 2012 and 2013

during the growing season (April–September) was in the 240-417 mm range. Mean annual temperature ranged from 4.8 to 6.0°C during the study period, and from 10.8 to 12.2°C from April through September (Figure 2).

The field experiment was set up in a randomized block design with three replications. The experimental material used in this study was timothy grass cv. Egida, which was bred at the Małopolska Plant Breeding Kraków and has been registered in the National Register of Cultivars since 2005.

The experiment was started on 17 September 2010. Timothy grass was sown without a cover crop, at 10.6 kg ha⁻¹ in 12.5-cm row spacing, on 10 m² (6.67 x 1.5) plots.



Fig. 2. Monthly air temperature at the Plant Breeding Station in Polanowice in the years 2011, 2012 and 2013

Timothy was pre-sown with 60 kg N ha⁻¹ of nitrogen fertilizers in the form of ammonium nitrate (NH₄NO₃), 30.5 kg P ha⁻¹ of phosphate fertilizers in the form of triple superphosphate (Ca(H₂PO₄)₂) and 83 kg K ha⁻¹ of potassium fertilizers in the form of potassium salt (KCl). In the next years, nitrogen fertilization was 200 kg N ha⁻¹ in the form of ammonium nitrate: for the first harvest 80 kg N ha⁻¹, and after the second and third harvests 60 kg N ha⁻¹ each; phosphorus fertilizer was applied once in the autumn of the year preceding the harvest at 52.3 kg P ha⁻¹ in the form of triple superphosphate; potassium was supplied as fertilizer at 116.2 kg ha⁻¹, in the form of potassium salt with 57% of K: for the first harvest 66.4 kg K ha⁻¹ and after the second harvest 49.8 kg K ha⁻¹.

Eight fertilizer treatments were tested in the experiment: control plot, foliar application of L-ascorbic acid in 3 doses (10, 20, 30 g ha⁻¹) and plots in which L-ascorbic acid (Standard, Lublin, Poland) was applied together with 10 kg ha⁻¹ of magnesium sulfate heptahydrate (MgSO₄ · 7H₂O; Intermag, Olkusz, Poland).

The experimental plots were denoted as follows:

- treatment AA0 control plot, no foliar application;
- treatment AA10 foliar application of L-AA, 10 g ha⁻¹;
- treatment AA20 foliar application of L-AA, 20 g ha⁻¹;

- treatment AA30 foliar application of L-AA, 30 g ha⁻¹;
- treatment Mg10 foliar application of $MgSO_4$ ·7H₂O, 10 kg ha⁻¹;
- treatment AA10+Mg10 foliar application of MgSO₄·7H₂O, 10 kg ha⁻¹ together with 10 g ha⁻¹ of L-AA, Mg: L-AA molar ratio of 79:1;
- treatment AA20+Mg10 foliar application of MgSO₄·7H₂O, 10 kg ha⁻¹ together with 20 g ha⁻¹ of L-AA, Mg: L-AA molar ratio of 159:1;
- treatment AA30+Mg10 foliar application of MgSO₄·7H₂O, 10 kg ha⁻¹ together with 30 g ha⁻¹ of L-AA, Mg: L-AA molar ratio of 238:1.

The plants were sprayed until wet, using 300 dm³ liquid per hectare. Spraying was performed 6-8 days from the start of growth for the first harvest, and a week after cutting for the other harvests.

The effect of foliar fertilization on the chlorophyll content was studied in different years of the experiment. Leaf greenness index (SPAD) was measured on the upper leaves, prior to each harvest, using a Minolta 502 DL SPAD chlorophyll meter (Minolta, Osaka, Japan). The measurements were taken on thirty fully developed leaves in each plot.

Relative chlorophyll content (M) expressed in SPAD units was calculated from the following formula (UDDLING et al. 2007):

$$M = k \log_{10} \frac{I_{0} (650)}{I_{(650)}} \frac{I_{(940)}}{I_{(650)}} ,$$

where: k – constant of proportionality (40 for 502 DL SPAD);

 $I_{\rm 0(650)}$ and $I_{\rm 0(940)}$ – amount of monochromatic light reaching the leaf at a wavelength of 650 and 940 nm;

 $I_{\rm (650)}$ and $I_{\rm (940)}~-$ amount of light transmission at a wavelength of 650 and 940 nm.

The agricultural and performance traits of the timothy grass submitted to the trials (post-winter condition of plants, regrowth of plants, sward density) were evaluated on a 9-point scale, where 1° was the poorest condition and 9° corresponded to the optimum value for a given trait (DOMAŃSKI et al. 1998).

Timothy grass was harvested at the heading stage (first cut) and after 6-7 weeks (second and third cuts). Directly after harvest, the yield was weighed and 0.5 kg of green mass was randomly sampled for chemical analysis. After drying and grinding, average weighed samples of the plants were analyzed for the content of organic and mineral matter using the Weende method (AOAC 2005).

The results were subjected to the analysis of variance and verified by the Duncan's test with Statistica 10 PL software (StatSoft, Inc., 2011).

RESULTS

Data on the dry matter yield of cv. Egida timothy grass for each harvest during the 3 years are presented in Table 1. The results show that fertiliza-Table 1

Years Treatment 2011 20122013First cut AA0 $80.6b \pm 0.93$ $71.7b \pm 0.73$ $67.3b \pm 0.72$ AA10 $72.8b \pm 0.94$ $67.5b \pm 0.82$ $82.6b \pm 0.71$ AA20 $84.1b \pm 0.82$ $73.8b \pm 0.59$ $70.6b \pm 0.78$ AA30 $84.8b \pm 0.87$ $78.8ab \pm 0.76$ $80.2ab \pm 0.68$ Mg10 $87.5ab \pm 0.65$ $81.7ab \pm 0.85$ $84.6ab \pm 0.79$ AA10+Mg10 $92.8a \pm 0.84$ $87.3a \pm 0.79$ $93.2a \pm 0.83$ AA20+Mg10 $95.8a \pm 0.75$ $88.9a \pm 0.68$ $95.9a \pm 1.47$ AA30+Mg10 $96.6a \pm 1.02$ $89.8a \pm 0.84$ $96.4a \pm 1.16$ SD standard deviation 6.212.57.5V(%) variation coefficient 7.09.3 15.2Second cut $44.8a \pm 0.74$ $31.4a \pm 0.56$ AA0 $23.9b \pm 0.49$ AA10 $24.7b \pm 0.61$ $45.0a \pm 0.63$ $31.8a \pm 0.74$ AA20 $25.1b \pm 0.54$ $46.0a \pm 0.79$ $33.5a \pm 0.63$ AA30 $26.0ab \pm 0.58$ $46.2a \pm 0.68$ $33.1a \pm 0.68$ Mg10 $26.2ab \pm 0.62$ $46.4a \pm 0.81$ $33.3a \pm 0.59$ AA10+Mg10 $46.5a \pm 0.87$ $33.5a \pm 0.47$ $26.5a \pm 0.72$ AA20+Mg10 $27.0a \pm 0.54$ $46.2a \pm 0.76$ $33.6a \pm 0.65$ AA30+Mg10 $27.5a \pm 0.46$ $47.3a \pm 0.83$ $34.3a \pm 0.59$ SD standard deviation 1.20.8 1.0V(%) variation coefficient 4.71.8 2.9Third cut AA0 $21.2b \pm 0.43$ $40.12a \pm 0.45$ $35.0a \pm 0.35$ AA10 $21.2b \pm 0.36$ $41.89a \pm 0.56$ $34.4a \pm 0.57$ AA20 $23.4b \pm 0.47$ $41.93a \pm 0.58$ $33.2a \pm 0.81$ AA30 $23.4b \pm 0.58$ $42.54a \pm 0.62$ $32.5a \pm 0.73$ Mg10 $23.8b \pm 0.62$ $43.03a \pm 0.71$ $33.6a \pm 0.65$ AA10+Mg10 $26.8ab \pm 0.78$ $43.14a \pm 0.87$ $31.2a \pm 0.67$ $35.1a \pm 0.74$ $27.9a \pm 0.82$ AA20+Mg10 $43.38a \pm 0.54$ AA30+Mg10 $28.7a \pm 0.78$ $45.07a \pm 0.62$ $38.2a \pm 0.76$ SD standard deviation 2.91.4 2.1V(%) variation coefficient 11.83.46.1

Dry matter yield of timothy (dt ha') at different harvests depending on the applied fertilization (mean values \pm SEM)

a, b – values in columns separately for each cut marked with different letters differ significantly ($P \le 0.05$) according to the Duncan's multiple range test, SEM – standard error of the mean

tion with L-ascorbic acid alone caused no differences in the dry matter yields of timothy ($P \leq 0.05$). However, there was a tendency for the dry matter yield to increase throughout the growing season in the plots where magnesium

sulfate was applied together with L-ascorbic acid; this effect was particularly evident in treatment AA30+Mg10, where the highest dose of L-ascorbic acid was applied. High dry matter yields were also characteristic for plots F and G. Differences in the other plots in relation to the control plot were not significant (P > 0.05). First-cut yields showed large variation but were higher than second- and third-cut yields.

In the timothy samples obtained from the plot without foliar application (control), the basic nutrients content was lower except for fibre and its fractions (Table 2). Crude protein content ranged from 103.4 to 143.3 g kg⁻¹ d.m. The increasing dose of L-ascorbic acid resulted in a significantly higher the crude protein content in treatment C compared to the control plot. The diffe-Table 2

Treatment	Crude protein	Crude fibre	Crude fat	ADF	ADL	NDF	Water soluble carbohydrates		
	(g kg ¹ d.m.)								
AA0	$103.4c\pm0.61$	$278.9a \pm 2.56$	$20.1c\pm1.93$	$320.4a \pm 4.52$	$26.9ab \pm 1.74$	$659.6a \pm 1.21$	$105.0c\pm2.82$		
AA10	$116.9bc \pm 2.73$	$269.1a \pm 1.45$	$25.3bc\pm3.79$	$314.5a \pm 4.72$	$24.0b \pm 1.31$	$618.3b \pm 1.45$	$107.9c\pm3.78$		
AA20	$120.0b\pm3.78$	$281.6a \pm 3.27$	$26.8b \pm 2.89$	$329.3a \pm 3.84$	$25.6b \pm 1.17$	$635.4ab \pm 2.68$	$112.7bc\pm2.45$		
AA30	$127.3b \pm 1.81$	$270.9a \pm 4.20$	$27.2b \pm 2.84$	$305.8b \pm 3.25$	$27.6ab \pm 2.21$	$622.7b \pm 4.51$	$122.3b\pm2.52$		
Mg10	$122.0b\pm2.79$	$252.4b \pm 2.87$	$27.3b \pm 4.15$	$298.2b \pm 2.71$	$28.0a \pm 1.41$	$598.9b \pm 5.62$	$113.1bc\pm3.02$		
AA10+Mg10	$129.0ba \pm 3.85$	$253.7b\pm2.73$	$27.7b \pm 3.46$	$299.8b \pm 4.26$	$28.1a\pm2.03$	$620.3b \pm 4.67$	$130.4b\pm2.52$		
AA20+Mg10	$132.0a\pm3.71$	$256.7b \pm 3.54$	$31.6a \pm 2.51$	$313.0b\pm5.11$	$28.3a \pm 1.47$	$624.6b \pm 5.71$	$132.0b\pm2.64$		
AA30+Mg10	$143.3a \pm 4.67$	$246.8b \pm 4.05$	$35.3a \pm 2.21$	$308.1b \pm 4.71$	$29.1a \pm 1.21$	$624.4b \pm 4.32$	$143.0a \pm 1.41$		
SD	11.8	13.1	4.4	10.5	1.8	17.1	13.4		
V(%)	9.5	5.0	16.0	3.4	6.6	2.7	11.1		

Nutrient content depending on the applied fertilization (means for three harvests from three years of the study), mean±SEM

a, b, c - values in columns marked with different letters differ significantly ($P \le 0.05$) according to the Duncan's multiple range test, SEM - standard error of the mean

rence was significant in all the plots where magnesium sulfate was applied together with L-ascorbic acid. The highest crude protein values were noted in the treatment where magnesium sulfate was combined with the highest concentration of L-ascorbic acid.

Crude fat content, determined as ether extract, is in the 20-50 g kg⁻¹ d.m. range for roughages. The crude fat content of the analyzed plants ranged from 20.1 g kg⁻¹ d.m. in plants from plot A to 35.3 g kg⁻¹ d.m. in plants from plot H; thus, the content of this component fell within the range of reference values.

The crude fibre content of the studied plants was in the 246.8-281.6 g kg⁻¹ d.m. range, being the highest in treatment C. The three-year average results concerning changes in the mineral content are shown in Table 3. The mineral content varied according to the fertilizer treatment, with the highest re-

Table 3

Treatment	Phosphorus	Potassium	Calcium	Magnesium	Sodium	
AA0	$2.21c\pm0.10$	$20.05c\pm1.21$	$1.40c\pm0.11$	$1.07b\pm0.09$	$0.11b \pm 0.010$	
AA10	$2.24c\pm0.13$	$20.50c \pm 1.44$	$1.42c\pm0.07$	$1.08b\pm0.10$	$0.11b \pm 0.008$	
AA20	$2.45 bc \pm 0.12$	$21.33b \pm 1.07$	$1.49ab\pm0.09$	$1.11b\pm0.08$	$0.11b \pm 0.007$	
AA30	$2.71b\pm0.15$	$21.59b \pm 0.86$	$1.68b\pm0.08$	$1.18a \pm 0.07$	$0.12ab \pm 0.011$	
Mg10	$2.80b\pm0.18$	$21.95b \pm 1.12$	$1.45b\pm0.05$	$1.10ab\pm0.08$	$0.13ab \pm 0.009$	
AA10+Mg10	$2.82b\pm0.21$	$22.65b \pm 1.36$	$1.67b\pm0.07$	$1.20b\pm0.10$	$0.13ab \pm 0.012$	
AA20+Mg10	$2.90a\pm0.19$	$23.51ab \pm 1.65$	$1.77 ba \pm 0.11$	$1.30a\pm0.07$	$0.16ba \pm 0.013$	
AA30+Mg10	$2.96a\pm0.14$	$24.13a \pm 1.26$	$1.96a \pm 0.14$	$1.47a \pm 0.11$	$0.20a \pm 0.018$	
SD	0.30	1.41	0.20	0.14	0.03	
V(%)	11.22	6.41	12.45	11.49	24.39	

Mineral content (g kg¹ d.m.) depending on the applied fertilization (means for three harvests from three years of the study), mean±SEM

a, b, c – values in columns marked with different letters differ significantly ($P \le 0.05$) according to the Duncan's multiple range test, SEM – standard error of the mean

lative variation found for sodium. A significant increase in the content of all the analyzed ions was found for the combined use of magnesium sulfate and L-ascorbic acid in the higher doses. Ascorbic acid fertilization also increased their content in the variants where no Mg fertilization was applied, which is indicative of the effect of ascorbic acid on the ion absorption capacity. Measurements of the chlorophyll content of leaves showed significant differences according to the date of measurement and foliar fertilization (Figure 3a, b). Average SPAD readings during the three years were higher for plots where magnesium sulfate was applied together with L-ascorbic acid compared to plots with foliar application of L-ascorbic acid alone. SPAD readings were also observed to vary according to the date of measurement, with the highest values found for the first regrowth of timothy. Leaf greenness index of the studied plants was in the 31.9-52.6 range depending on the date of measurement. The application of magnesium sulfate with L-ascorbic acid caused slight improvement in the post-winter condition of the plants, their spring regrowth and sward density before and after the winter. In turn, foliar fertilization with magnesium sulfate additionally improved the regrowth after the grass was cut (Table 4).

DISCUSSION

In the present experiment, a synergistic effect of magnesium and ascorbic acid on yielding, spring regrowth and forage quality of timothy was observed. In our study, foliar application of magnesium and ascorbic acid cau-



Fig. 3a. Leaf greenness index (SPAD) of timothy grass cv. Egida depending on fertilization with ascorbic acid (means for three years of the study); b, c – values within cuts marked with different letters differ significantly ($P \le 0.05$) according

p, c – values within cuts marked with different letters differ significantly ($P \le 0.05$) according to the Duncan's multiple range test



 $\begin{array}{ll} {\rm MgSO_4\cdot 7H_2O\ 10\ (kg\ ha^{-1})\ (constant)\ +\ ascorbic\ acid\ (g\ ha^{-1})} \\ {\rm Fig.\ 3b.\ Leaf\ greenness\ index\ (SPAD)\ of\ timothy\ grass\ cv.\ Egida\ depending\ on\ fertilization\ with\ ascorbic\ acid\ and\ magnesium\ sulfate\ (means\ for\ three\ years\ of\ the\ study);} \\ a,\ b-{\rm values\ within\ cuts\ marked\ with\ different\ letters\ differ\ significantly\ (P\leq 0.05)\ according\ to\ the\ Duncan's\ multiple\ range\ test.} \end{array}$

	density	spring before winter		$8.2b \pm 0.49$	$8.2b\pm0.57$	$8.3b \pm 0.42$	$8.4b \pm 0.34$	$8.6ab \pm 0.43$	$8.6ab \pm 0.52$	$8.8a \pm 0.62$	$8.8a \pm 0.44$	0.25
	Sward			$7.7b \pm 0.54$	$7.8b \pm 0.39$	$8.0b\pm0.48$	$8.4b \pm 0.34$	$8.4b\pm0.26$	$8.6a \pm 0.43$	$8.6a\pm0.52$	$8.7a \pm 0.61$	0.39
		after the cuts	III	$7.8b \pm 0.46$	$7.9b \pm 0.46$	$8.0b \pm 0.46$	$8.0b \pm 0.46$	$8.1ab \pm 0.46$	$8.2a \pm 0.46$	$8.2a \pm 0.46$	$8.3a \pm 0.46$	0.17
	of plants		Π	$6.7b \pm 0.34$	$6.8b\pm0.55$	$6.9b \pm 0.40$	$7.0b \pm 0.32$	$7.2ab \pm 0.41$	$7.3a \pm 0.49$	$7.3a \pm 0.57$	$7.4a \pm 0.42$	0.26
	Regrowth		Ι	$8.1b\pm0.36$	$8.0b \pm 0.40$	$8.2b \pm 0.49$	$8.2b\pm0.74$	$8.3ab \pm 0.58$	$8.4a \pm 0.76$	$8.4a \pm 0.67$	$8.5a\pm0.77$	0.17
			spring	$8.0b \pm 0.48$	$8.0b\pm0.56$	$8.2b \pm 0.41$	$8.2b\pm0.36$	$8.3b \pm 0.42$	$8.5a \pm 0.51$	$8.6a \pm 0.60$	$8.6a \pm 0.43$	0.24
	Post-winter condition of plants		$8.2b\pm0.36$	$8.2b \pm 0.46$	$8.3b \pm 0.41$	$8.3b \pm 0.45$	$8.4ab \pm 0.54$	$8.4ab \pm 0.64$	$8.6a\pm0.55$	$8.6a \pm 0.40$	0.16	
		Treatment		AA0	AA10	AA20	AA30	Mg10	AA10+Mg10	AA20+Mg10	AA30+Mg10	SD

 1° – poorest condition, 9° – optimum condition,

a, b - values in columns marked with different letters differ significantly ($P \leq 0.05$) according to the Duncan's multiple range test, SEM – standard error of the mean

2.92

4.69

2.09

3.68

2.04

2.95

1.89

V(%)

Table 4

Agricultural and performance traits of timothy grass depending on the fertilization on a 9-point scale (means for three years of the study)

sed a slight increase in dry matter yield in the entire growing season. The application of magnesium sulfate with L-ascorbic acid resulted in a slight improvement in spring regrowth and sward density before and after the winter. In turn, foliar fertilization with magnesium sulfate additionally improved the regrowth after harvest. The ascorbic acid content increased in plants under stress conditions, especially under parallel magnesium deficiency (CAKMAK, MARSCHNER 1992, TEWARI et al. 2005). Also magnesium deficiency *per se* induced antioxidative response of plants, including ascorbic acid biosynthesis.

In our study, the combined fertilization with magnesium and ascorbic acid increased the chlorophyll content of leaves of tymothy. The SPAD values were higher for the plots fertilized with magnesium sulfate and L-ascorbic acid compared to the plots with foliar application of L-ascorbic acid alone. Furthermore, SPAD readings were observed to vary according to the date of measurement, with the highest values noted for the first regrowth. The leaf greenness index (SPAD) of the plants studied was in the 31.9-52.6 range depending on the date of measurement. In a study by CIECKO et al. (2012), foliar application of magnesium had a decisive effect on the synthesis of chlorophyll in leaves of table potatoes and on the yield increase. OLSZEWSKA (2005) demonstrated that magnesium deficiency significantly reduced foliar photosynthesis of orchard-grass and perennial ryegrass. The most pronounced consequences of magnesium deficiency were found for cv. Argona ryegrass, in which the rate of photosynthesis decreased by 30-50%. Similar findings were reported by GABORCIK, ZMETAKOVA (2001), who found magnesium deficiency to reduce significantly the chlorophyll content of ryegrass leaves. Chlorophyll concentration decreased by 3 to 7%, and the difference between the control and the magnesium deficient plots increased with after each cut. According to CAKMAKA, KIRKBY (2008), magnesium deficiency may damage chloroplasts, which produce less of the green pigment, and this affects both plant productivity and the quality of crops. This assumption is supported by DHIRAJ, KUMAR (2012), who concluded that foliar fertilization had a significant effect on the qualitative and qualitative characteristics of mulberry yields. SABO et al. (2002) showed that winter wheat genotypes are able to absorb 80-130 kg ha⁻¹ magnesium from soil, which significantly increases dry matter accumulation and crop productivity.

In our study, the beneficial effects of ascorbate/magnesium nutrition were the strongest in the second and third cut in 2011 (increases in the relative numbers of 15 and 35%, respectively) and in the first cut in 2013 (43%). It is possible that the yielding of both the second and third cut in 2011 was affected by very dry May and June, while in 2013 the first cut was influenced by the low precipitation in April. Also CIECKO et al. (2012) observed a particularly stimulating effect of magnesium application on yielding and chlorophyll content in potato in years with adverse weather conditions.

In our study, the control plants had less basic nutrients except fibre and its fractions. The fat content did not increase significantly after the application of magnesium and ascorbic acid. These results contradict the data published by UPADHYAY and PATRA (2011), who showed that 200 mg pot⁻¹ Mg per 10 kg soil significantly increased the shoot length, shoot weight and fat content in chamomile. In our study, the highest crude protein values were observed in the treatment where magnesium sulfate was applied together with the highest concentration of L-ascorbic acid. The combination of magnesium sulfate and L-ascorbic acid (30 g ha⁻¹) applied there contributed to a significant increase in the amount of accumulated macronutrients (P, K, Ca, Mg, Na). Positive influence of magnesium on nitrogen absorption, which results in an increase of the protein content, was reported by FERNANDES, ROSSIELLO (1995), who showed that magnesium had a significant effect on enzymatic processes responsible in plants for the uptake and utilization of ions from soil. EL-ZANATY et al. (2012) found magnesium sulfate to have a marked effect on the concentration of nutrients by increasing N, Ca, Mg, Na, Fe and Cu concentrations in wheat shoot tissues.

CONCLUSIONS

Foliar application of the tested substances increased the dry matter yield and the content of organic and mineral components in timothy grass. Best effects in terms of yielding and forage quality were obtained after the combined application of magnesium sulfate and ascorbic acid (30 g ha⁻¹).

ACKNOWLEDGMENTS

The authors thank the Intermag LLC from Olkusz for their assistance in conducting this experiment. In particular, we are grateful to Mr. Piotr Lubaszka, Director of Sales Companies Intermag.

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