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CHANGES OF CALCIUM AND MAGNESIUM CONTENT IN BIOMASS OF GOAT'S RUE (*Galega orientalis* Lam.) DURING VEGETATION

ZMIANY ZAWARTOŚCI WAPNIA I MAGNEZU W BIOMASIE RUTWICY WSCHODNIEJ (*Galega orientalis* Lam.) PODCZAS WEGETACJI

Abstract: Calcium and magnesium are among macroelements, both in plant and animal feeding. Green forage, especially that obtained from legume plants, is their main source in ruminants' feed. The optimum content of those elements in fodder positively affects its quality. The aim of this study was to trace changes in calcium and magnesium content in fodder galega (*Galega orientalis*) during the vegetation period, depending on the year of cultivation and development phase. The results are based on two field experiments, conducted for the third and seventh year. Samples were taken during the harvest from 1 m² of the field during the following development phases: budding, start of blossoming, full bloom, end of blossoming and full ripeness. Subsequently, the samples were dried and ground. Calcium and magnesium were determined by the ICP-AES method following dry mineralisation.

Statistical calculations have revealed significant variation in calcium and magnesium content in fodder galega (*Galega orientalis*), depending on the year of cultivation and development phase. The average calcium content in dry matter of the test plant was equal to 15.57 g \cdot kg⁻¹, while that of magnesium was 2.54 g \cdot kg⁻¹. The largest amounts of calcium and magnesium were found in the leaves of the test plant in the third year of cultivation. Considering different development phases of fodder galega, it can be concluded that the highest level of calcium was determined at the end of the blossoming phase and the highest level of magnesium – during the full ripeness phase. The average Ca : Mg ratio was equal to 6.12 : 1.

Keywords: fodder galega (Galega orientalis Lam.), calcium, magnesium, year of cultivation, development phase, biomass, leaves, stem, Ca : Mg ratio

Fodder galega (*Galega orientalis* Lam.) is a perennial plant of the legume family. It originates in Caucasus, and it is currently grown in Estonia, Latvia, Finland, France,

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Canada and Poland. It has a high ability to reduce molecular nitrogen $(379.7 \text{ kgN} \cdot \text{ha}^{-1})$ on average) [1], owing to inoculating soil or seeds with *Rhizobium galegae* bacteria [2–5]. This makes its cultivation for fodder feasible and highly profitable. It can be used as fodder for animals in agriculture and animal feeding as green forage, hay, dried material, silage and protein concentrate [6-8]. Demand for calcium and magnesium from dicotyledonous plants is higher than from monospermous ones. Of the cultivated plants, legumes contain particularly high concentrations of calcium and magnesium. In overlimed soils, antagonism between Mg2+ and Ca2+ ions may occur. A high concentration of Mg²⁺ ions in soil may have a harmful effect on plants as the Ca/Mg balance is disturbed. To ensure proper absorption of calcium from fodders, it is necessary to maintain the proper balance between cations and anions [9]. Calcium action in animal bodies is closely linked with magnesium, which is its antagonist. There have been few reports in the foreign and Polish literature on the chemical composition of the plant [10–14]. It is important to monitor plants intended for fodder for calcium and magnesium content because any deficit or excess negatively affects the fodder quality and animal health status.

The aim of this study was to trace changes in calcium and magnesium content in the biomass of fodder galega (*Galega orientalis*) and in its leaves and stems, depending on the year of cultivation and development phase.

Materials and methods

Field experiments were conducted on soil formed from clayey sand, with pH in 1 mol KCl \cdot dm⁻³ – 6.6. The soil contained 11.5 g \cdot kg⁻¹ of organic carbon and 0.1 g \cdot kg⁻¹ of total nitrogen. The content of available phosphorus and potassium (determined by the Egner-Riehm method) was referred to as high (80 mg \cdot kg⁻¹ P and 140 mg \cdot kg⁻¹ K) and that of magnesium (determined by the Schachtschabel method) as medium (50 mg \cdot kg⁻¹). The total Ca content in soil on which galega was cultivated for the third year was 7.88 g \cdot kg⁻¹, while in the seventh year it was 5.61 g \cdot kg⁻¹. Magnesium content in the surface layer of the soil was $0.84 \text{ g} \cdot \text{kg}^{-1}$ under galega grown for the third year and $0.68 \text{ g} \cdot \text{kg}^{-1}$ in the seventh year of cultivation. Fodder galega was sown in May 1997 and 2001 at the depth of 2–3 cm in the amount of 24 kg \cdot ha⁻¹ in 12–15 cm rows. Scarified seeds were sown into soil infected with a strain of Rhizobium galegae bacteria. Weed control procedures were performed during the vegetation period and proper humidity level was maintained (sprinkling). Samples in the following development phases: budding, start of blossoming, full bloom, end of blossoming and full ripeness were taken from 1 m² of the field during the harvest of green mass of fodder galega in 2003 from both fields (3rd and 7th year of the experiment). The samples were dried, in some of them leaves were separated from stems and ground. Calcium and magnesium were determined in the prepared material by the ICP-AES method following dry mineralisation [15]. The results were analysed statistically with an analysis of variance and significant differences were calculated with Tukey's test at the level of significance of p = 0.05.

Results and discussion

Weather data for the 2003 vegetation period is shown in Table 1.

Table 1

Rainfall and air temperatures in the vegetation in the years 2003.
Reported by the measurement centre in Siedlce

May	June	July	August	September	Sum or mean						
Total monthly rainfall [mm]											
37.2	26.6	26.1	24.3	118.9							
	Multiyear monthly rainfall [mm]										
50.0	75.0	80.0	68.0	47.3	320.3						
	Means monthly air temperature [°C]										
15.6	18.4	20.0	18.4	13.5	18.5						
	Multiyear temperature mean [°C]										
12.6	16.6	17.7	26.9	12.7	17.3						

Weather conditions during that season were not favourable for field crops. A low rainfall level is particularly noteworthy. It was nearly three times lower than the multiyear average. It significantly reduced the yield and changed the calcium and magnesium content in the development phases of fodder galega (*Galega orientalis* Lam.) and changed the content of the macroelements under study in the analysed plant parts.

The average calcium content in the dry matter of fodder galega was equal to $15.57 \text{ g} \cdot \text{kg}^{-1}$ (Table 2) and it was significantly varied by the analysed factors and their combinations.

Table 2

Vara fariting						
Year of cultivation (A)	Budding	Begin of flowering	Full flowering	End of flowering	Full ripeness	Mean
Third	10.97	15.27	17.53	23.09	23.10	17.99
Seventh	10.28	9.44	10.42	18.10	17.56	13.16
Mean	10.62	12.35	13.98	20.59	20.33	15.57

The content of calcium $[g \cdot kg^{-1} \text{ d.m.}]$ in biomass of goat's rue

 $LSD_{0.05}$ for: years (A) – 0.61; development stage (B) – 1.38; interaction (AxB) – 1.36; interaction (BxA) – 1.94.

Significantly, the highest calcium content in dry matter of the test plant biomass was found in the third year of the experiment (17.99 g \cdot kg⁻¹). Considering the effect of development phase on the Ca level in dry matter of the plant, it is clear that the highest amount of calcium was accumulated in the full ripeness phase (23.10 g \cdot kg⁻¹). Statistical analysis has shown significant differences in calcium content between consecutive development phases and the full ripeness phase. These analyses results

were confirmed by a study by Symanowicz and Kalembasa [16], who examined the effect of phosphorus-potassium fertilisation on the yield and macroelement content in biomass of fodder galega. An increase in calcium uptake by the test plant was also affected by the weather conditions, soil pH and calcium content in soil. The results lay within the acceptable range for fodders [9].

Potential for the production of protein concentrates from leaves of fodder galega encouraged the authors to examine those parts of the plant. The analysed factors and their combinations significantly differentiated the total content of calcium in leaves of fodder galega (Table 3). A significantly higher calcium content in dry matter of leaves of the test plant was found in the third year of the experiment (26.29 g \cdot kg⁻¹). Chemical analyses of leaves of the test plant in consecutive development phases showed a significant increase in calcium content. In the ripeness phase, it was more than twice as high as in the budding phase. The results for the budding phase and start of blossoming phase were confirmed by a study conducted by Ignaczak [7] and Symanowicz and Kalembasa [14].

Table 3

The content of calcium	[g · kg	⁻¹ d.m.] in leaves of goat's rue
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Year of cultivation							
(A)	Budding	Begin of flowering	Full flowering	End of flowering	Full ripeness	Mean	
Third Seventh	16.43 13.46	19.43 14.72	26.42 19.26	34.37 30.68	34.82 31.94	26.29 22.01	
Mean	14.94	17.07	22.84	32.53	33.38	24.15	

 $LSD_{0.05}$ for: years (A) - 0.39; development stage (B) - 0.88; interaction (AxB) - 0.87; interaction (BxA) - 1.24.

Statistical analysis has shown significant differences in the calcium content in stems (Table 4).

Table 4

X C 10 0	Development stage (B)								
Year of cultivation (A)	Budding	Begin of Full flowering flowerin		End of flowering	Full ripeness	Mean			
Third	6.76	7.45	8.09	12.31	11.88	9.30			
Seventh	5.59	3.89	4.82	6.92	6.88	5.62			
Mean	6.17	5.67	6.45	9.62	9.38	7.46			

The content of calcium $[g \cdot kg^{-1} \text{ d.m.}]$ in stems of goat's rue

 $LSD_{0.05}$ for: years (A) - 0.20; development stage (B) - 0.45; interaction (AxB) - 0.64; interaction (BxA) - 0.45.

The average content of calcium in dry matter of stems of the test plant was equal to 7.46 g \cdot kg⁻¹ and it was much higher than the findings of other studies by Symanowicz

and Kalembasa [14]. Differences in calcium content in dry matter of stems in the third and seventh year of the experiment were highly significant (9.30 and 5.62 g \cdot kg⁻¹). There was significant differentiation between development phases in calcium content. Significantly, the highest level of Ca was found in stems at the end of the blossoming phase (9.62 g \cdot kg⁻¹ of d.m.), with the level decreasing insignificantly (9.38 g \cdot kg⁻¹ d.m.) during the next development phase (full ripeness).

The average magnesium content in dry matter of fodder galega was equal to 2.54 g \cdot kg⁻¹ (Table 5) and it was significantly varied by the analysed factors and their combinations. Significantly, the highest magnesium content in dry matter of the test plant biomass was found in the third year of the experiment (2.74 g \cdot kg⁻¹). Considering the effect of development phase on the Mg level in dry matter of the plant, it was found that the highest amount of magnesium was accumulated in the full ripeness phase (2.87 g \cdot kg⁻¹). Statistical analysis has shown significant differences in magnesium content between consecutive development phases except for the relationship between the budding phase, start of blossoming as well as the full bloom and end of blossoming phases. The results are similar to the findings of a study of the effect of inoculation of seeds of fodder galega (Galega orientalis Lam.) on macroelement content [14]. Ignaczak [7] examined the quality of yield of green forage obtained from the first cut (harvested in spring during the budding phase) and determined the magnesium content to be equal to 2.6 g · kg⁻¹ d.m. According to Anke [17], Falkowski et al [18], Kabata-Pendias and Pendias [19], magnesium content of about 2 g \cdot kg⁻¹ is sufficient to cover the nutritional needs of animals. Patorczyk-Pytlik [20] showed the sward of grassland to be usually of low value as fodder in terms of its mineral composition. According to the findings of that study, 59 % samples of pasture sward did not meet the feeding standards for dairy cattle in terms of magnesium content. Conversely, a study by Ignaczak [21], comparing the traditional and extensive system of using fodder galega, gave entirely different results. When the plant was used traditionally as green forage (3 cuts), the magnesium content ranged $1.07-2.01 \text{ g} \cdot \text{kg}^{-1}$ d.m., while in the third year of fallowing, it ranged 4.6–11.3 g \cdot kg⁻¹ d.m. A large extent of magnesium release from soil and incorporation into circulation in the cultivating of perennial plants (including fodder galega) has been indicated by the findings of studies of Raig et al [22] and Zarczynski et al [23].

Table 5

Development stage (B) Year of cultivation Mean Begin of Full End of Full (A) Budding flowering flowering flowering ripeness Third 2.18 2.45 3.01 2.93 3.15 2.74 Seventh 2.40 2.09 2.26 2.36 2.60 2.34 2.29 2.27 Mean 2.63 2.64 2.87 2.54

The content of magnesium $[g \cdot kg^{-1} \text{ d.m.}]$ in biomass of goat's rue

 $LSD_{0.05}$ for: years (A) - 0.07; development stage (B) - 0.16; interaction (AxB) - 0.22; interaction (BxA) - 0.15.

The analysed factors significantly differentiated the total content of magnesium in leaves of fodder galega (Table 6). The average magnesium content in dry matter of leaves was equal to $3.58 \text{ g} \cdot \text{kg}^{-1}$. A significantly higher magnesium content in leaves of the test plant was found in the third year of the experiment (3.67 g \cdot kg⁻¹). The dry matter of fodder galega harvested at the consecutive development phases was found to contain significantly more magnesium during the full bloom phase (4.12 g \cdot kg⁻¹ of d.m.). The results have confirmed the findings of earlier studies by the authors [14].

Table 6

The content of magnesium $[g \cdot kg^{-1} \text{ d.m.}]$ in leaves of goat's rue

T C III II							
Year of cultivation (A)	Budding	Begin of Full flowering flowering		End of flowering	Full ripeness	Mean	
Third Seventh	2.83 2.70	2.87 3.17	4.35 3.90	4.24 3.94	4.05 3.77	367 3.49	
Mean	2.77	3.02	4.12	4.09	3.91	3.58	

 $LSD_{0.05}$ for: years (A) - 0.08; development stage (B) - 0.17; interaction (AxB) - 0.24; interaction (BxA) - 0.17.

The average content of magnesium in dry matter of stems of the test plant was equal to 1.42 g \cdot kg⁻¹ (Table 7) and it confirmed the findings of an earlier study by Symanowicz and Kalembasa [14]. Statistical calculations have revealed significant variation in magnesium content in stems of fodder galega, depending on the year of cultivation and development phase. The differences in magnesium content in dry matter of stems in the third and seventh year of the experiment were highly significant (1.61 g \cdot kg⁻¹ and 1.24 g \cdot kg⁻¹). There is significant differentiation between each development phase with reference to magnesium content. Significantly, the highest Mg content was found in stems during the budding phase (1.63 g \cdot kg⁻¹ d.m.).

Table 7

The content of magnesium $[g \cdot kg^{-1} \text{ d.m.}]$ in stems of goat's rue

X C Ivi vi						
Year of cultivation (A)	Budding	Begin of flowering	Full flowering	End of flowering		
Third Seventh	1.46 1.81	1.58 1.00	1.54 0.90	1.91 1.29	1.59 1.18	1.61 1.24
Mean	1.63	1.29	1.22	1.60	1.38	1.42

 $LSD_{0.05}$ for: years (A) - 0.05; development stage (B) - 0.11; interaction (AxB) - 0.15; interaction (BxA) - 0.11.

The average calcium/magnesium ratio in the test plant was about 6.12 : 1 (Table 8) and it lay within the range acceptable for fodders, which ranges between 3 : 1 and 6.5 : 1 [9, 24]. The Ca/Mg ratio slightly exceeded the acceptable range in the third year of the experiment. Considering individual development phases of fodder galega, it must be said that the calcium and magnesium content, determined during the budding phase, set the Ca : Mg ratio at the optimum level of 4.64 : 1. This is advantageous, as fodder galega should be harvested during the budding phase for direct feeding or for dried material. A much lower Ca : Mg ratio (2.7 : 1) was found by Ignaczak [7], who examined the quality of green forage from the first cut harvested in spring at the budding phase. A wide range of Ca : Mg ratio, calculated for fodder galega harvested during the blossoming phase, and full ripeness phase indicates the absence of a possibility of using such fodder in animal feeding [9].

Table 8

Budding	Begin of flowering	Full flowering	End of flowering	Full ripeness	Mean
5.03	6.23	5.82	7.88	7.33	6.56
4.28	4.52 5.44	5.31	7.80	7.08	5.62 6.12
	5.03 4.28	BuddingBegin of flowering5.036.234.284.52	BuddingBegin of floweringFull flowering5.036.235.824.284.524.62	Budding flowering flowering flowering 5.03 6.23 5.82 7.88 4.28 4.52 4.62 7.67	BuddingBegin of floweringFull floweringEnd of floweringFull ripeness5.036.235.827.887.334.284.524.627.676.75

The values of molar ratio Ca : Mg in biomass of goat's rue

Table 9 shows the correlation coefficients between the content of calcium and magnesium in the plant biomass, in its leaves and stems of Galega orientalis Lam. in consecutive years of study. Calcium content determined in biomass of the entire plant in the consecutive development phases in the third year of the experiment was significantly positively correlated with the calcium content in leaves and stems, as determined in the third year, with calcium content in the entire plant and in leaves in the seventh year of the experiment as well as with the magnesium content in leaves in the seventh year of the experiment. The correlation coefficients indicate a significant positive correlation between the calcium content in leaves in the third year of the experiment and the calcium content in stems, as determined in the third year, with calcium content in the entire plant biomass and in leaves in the seventh year of the experiment as well as with the magnesium content in leaves in the seventh year of the experiment. Calcium content determined in stems in the third year of the experiment was significantly correlated with the calcium content in the entire plant and in leaves in the seventh year of the experiment. Calcium content in the biomass of the test plant in the seventh year was positively correlated with the calcium content in leaves and stems. Total magnesium content determined in galega biomass in the third year of the experiment was significantly correlated with the magnesium content in the leaves in the third and the seventh year of the experiment. A significant correlation of magnesium content in leaves was observed in the third and seventh year of the experiment.

Casvii Mgtiii Mgtiii Mgsiii Mgtvii Mgtvii Mgtvii Mgsvii						1.00	0.49 1.00	0.53 0.93* 1.00	0.53 0.43 0.51 1.00	0.86 0.41 0.36 0.04 1.00	0.42 0.95* 0.95* 0.61 0.20 1.00	0.35 -0.61 -0.48 -0.16 0.40 -0.66 1.00	* – significant at $\alpha = 0.05$; Ca _{1 III} – the content of calcium in total biomass in third year of cultivation; Ca _{1 III} – the content of calcium in leaves in third year of cultivation; Ca _{s III} – the content of calcium in leaves in third year of cultivation; Ca _{1 VII} – the content of calcium in stems in stems in third year of cultivation; Ca _{1 VII} – the content of calcium in total in seventh year of cultivation; Ca _{1 VII} – the content of calcium in total in seventh year of cultivation; Ca _{1 VII} – the content of calcium in leaves in seventh year of cultivation; Mg _{1 III} – the content of magnesium in total biomass in third year of cultivation; Mg _{1 III} – the content of magnesium third year of cultivation; Mg _{1 VII} – the content of magnesium in total biomass in third year of cultivation; Mg _{1 VII} – the content of magnesium in third year of cultivation; Mg _{1 VII} – the content of magnesium in third year of cultivation; Mg _{1 VII} – the content of magnesium in third year of cultivation; Mg _{1 VII} – the content of magnesium in the cultivation; Mg _{1 VII} – the content of magnesium in teal in seventh year of cultivation; Mg _{1 VII} – the content of magnesium in stems in third year of cultivation; Mg _{1 VII} – the content of magnesium in stems in third year of cultivation; Mg _{1 VII} – the content of magnesium in stems in third year of cultivation; Mg _{1 VII} – the content of magnesium in stems in seventh year of cultivation; Mg _{1 VII} – the content of magnesium in stems in seventh year of cultivation.
Ca _{t VII} Ca _{1 VII}				1.00	0.97* 1.00	0.92* 0.84	0.67 0.82	0.62 0.75	0.72 0.68	0.69 0.65	0.62 0.77	0.03 -0.19	* – significant at $\alpha = 0.05$; Ca _{1 III} – the content of calcium in total biomass in third year c cultivation; Ca _{1 VII} – th cultivation; Ca _{1 VII} – th content of calcium in stems in third year of cultivation; Ca _{1 VII} – th content of calcium in leaves in seventh year of cultivation; Ma _{1 VII} – the content of calcium in in total biomass in third year of cultivation; Mg _{1 VII} – the content of calcium in leaves in third year of cultivation; Mg _{1 VII} – the content of calcium in third year of cultivation; Mg _{1 VII} – the content of magnesium in leaves in third year of cultivation; Mg _{1 VII} – the content of magnesium in total in seventh year of cultivation; Mg _{8 VII} – the content of magnesium in total in seventh year of cultivation.
Ca s III			1.00	0.98^{*}	0.99*	0.83	0.76	0.70	0.77	0.58	0.74	-0.16	the content o alcium in stem th year of cult vation; Mg 1 m e content of n of magnesium
Ca I III		1.00	0.96^{*}	0.91^{*}	0.98^{*}	0.75	0.91^{*}	0.86	0.68	0.55	0.88^{*}	-0.35	05; Ca t III – content of ci ves in sevent year of culti Mg t vII – th he content c
CatIII	1.00	0.98^{*}	0.96*	0.89^{*}	0.96^{*}	0.67	0.89^{*}	0.80	0.72	0.44	0.88^{*}	-0.44	unt at $\alpha = 0$. Ca $_{s \text{ III}}$ – the alcium in lea intrimit asson third asson third Ng $_{s \text{ VII}}$ – t
Content	Ca _{t III}	Ca IIII	Ca _{s III}	Ca_{tVII}	$Ca_{1 \rm VII}$	$Ca{\rm \ s\ vII}$	${\rm Mg}_{\rm tIII}$	Mg _{1III}	Mg_{sIII}	Mg_{tVII}	$Mg_{1\rm VII}$	Mg_{sVII}	* - significant at $\alpha = 0.05$; C cultivation; Ca s _{in} - the conte content of calcium in leaves in in total biomass in third year of third year of cultivation; Mg s _{vin} - the co

Table 9

696

Conclusions

1. The highest content of Ca and Mg was found in leaves of fodder galega (*Galega orientalis* Lam.). The content of calcium and magnesium in stems was 2–3 times lower than in leaves.

2. Significantly, the highest amounts of the elements under analysis were found in the plant in the third year of cultivation.

3. The highest content of calcium was found during the full ripeness phase and the highest content of magnesium was found during the full bloom phase.

4. A high content of calcium in biomass of fodder galega (*Galega orientalis* Lam.) set the Ca : Mg ratio at a high level.

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ZMIANY ZAWARTOŚCI WAPNIA I MAGNEZU W BIOMASIE RUTWICY WSCHODNIEJ (*Galega orientalis* Lam.) PODCZAS WEGETACJI

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Abstrakt: Wapń i magnez należą do podstawowych składników mineralnych organizmów zwierzęcych. Są one niezbędne do funkcjonowania całego organizmu i metabolizmu komórek. Podstawowym źródłem Ca i Mg w żywieniu zwierząt przeżuwających są zielonki, szczególnie roślin bobowatych. Optymalna zawartość tych składników w paszy korzystnie wpływa na jej jakość. Celem przeprowadzonych badań było prześledzenie zmian w zawartości wapnia i magnezu w biomasie rutwicy wschodniej w zależności od roku uprawy i fazy rozwojowej. Wyniki badań uzyskano na podstawie dwóch doświadczeń polowych prowadzonych trzeci i siódmy rok. Podczas zbioru pobrano próbki z 1 m² w następujących fazach rozwojowych: pąkowanie, początek kwitnienia, pełnia kwitnienia, koniec kwitnienia i dojrzałość pełna. Następnie próbki te wysuszono i rozdrobniono. Wapń i magnez oznaczono metodą ICP-AES, po mineralizacji "na sucho".

Obliczenia statystyczne wykazały istotne zróżnicowanie w zawartości wapnia i magnezu w biomasie rutwicy wschodniej (*Galega orientalis* Lam.) w zależności od roku uprawy i fazy rozwojowej. Średnia zawartość wapnia w suchej masie rośliny testowej wynosiła 15,57 g \cdot kg⁻¹ a magnezu 2,54 g \cdot kg⁻¹. Największe ilości wapnia i magnezu oznaczono w liściach rośliny testowej w trzecim roku uprawy. Rozpatrując poszczególne fazy rozwojowe rutwicy wschodniej, należy stwierdzić, że w fazie koniec kwitnienia oznaczono najwięcej wapnia, natomiast magnezu w fazie dojrzałości pełnej. Średni stosunek Ca : Mg ukształtował się na poziomie 6,12 : 1.

Słowa kluczowe: rutwica wschodnia (*Galega orientalis* Lam.), wapń, magnez, rok uprawy, faza rozwojowa, biomasa, liście, łodyga, stosunek Ca : Mg