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ORIGINAL PAPER

TOTAL PROTEIN AND MACROELEMENT CONTENT IN SELECTED PSAMMOPHILOUS GRASSLAND SPECIES UNDER FREE-RANGE SHEEP GRAZING IN KÓZKI NATURE RESERVE*

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Abstract

Psammophilous grasslands are among the most valuable but also severely threatened habitats protected within the Natura 2000 network and, like most grassy ecosystems, they are also subject to secondary succession. Grazing has turned out to be an adequate conservation measure in inland sand ecosystems. Sheep are small ruminants which ingest plant species ignored by other livestock species; they also eat some undesirable plants containing toxic alkaloids and glycosides. The study objective is to assess the macroelement content of plants ingested by sheep of the breed called Swiniarka. Regarded as being of low value, these plants occur in the sward of psammophilous grasslands in Kózki Nature Reserve. The Braun-Blanquet method was used to study the vegetation cover and identify the type of a plant community. The total protein, phosphorus, potassium, magnesium and sulphur content in the aerial biomass of selected plant species (Armeria maritima, Calamagrostis epigejos, Carex praecox, Corynephorus canescens, Cynoglossum officinale, Dianthus deltoides, Juniperus communis, Koeleria glauca, Sedum acre and *Thymus pulegioides*) was determined. The analysed plant species occurred in the floristically rich psammophilous grasslands of the Vicio lathyroidis-Potentillion argenteae alliance, Spergulo-Corynephoretum association and in a community with the predominance of Calamagrostis epigejos. The biomass of psammophilous grasslands was characterised by a very low content of protein and phosphorus, potassium, magnesium and sulphur, in most cases below the optimum

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levels that would satisfy the nutritional requirements of animals. The biomass of *Cynoglossum* officinale had the significantly highest content of protein, phosphorus and magnesium, which could have been the reason why this species, despite its poisonous properties, was ingested by the Świniarka sheep grazing under the poor conditions of psammophilous grasslands. Species typical of psammophilous grasslands, such as *Corynephorus canescens* or *Koeleria glauca*, had the lowest nutritional value. *C. canescens* and *Sedum acre* were not ingested by sheep.

Keywords: macroelements, psammophilous grassland, Świniarka breed.

INTRODUCTION

Polish agricultural areas are a patchwork of fields, orchards, grasslands and pastures with flora and fauna characteristic for specific habitats. The landscape typical of pastures has been created by the grazing of ruminants over the ages. The existence of some habitats is inextricably linked with livestock farming. Psammophilous grasslands constitute such a habitat and the most valuable ones are protected within the Natura 2000 network. Housing and recreational developments as well as sand exploitation, trampling and collection of medicinal plants are the major threats to psammophilous grasslands (TRABA, ROGUT 2013). Like most grassy ecosystems, psammophilous grasslands are also subject to secondary succession, i.e. encroachment of shrubs and trees. Sheep grazing has turned out to be an adequate conservation tool in inland sand ecosystems (Stroh et al. 2002, Hellström et al. 2003, FAUST et al. 2011, WARDA et al. 2011, KULIK et al. 2013). Sheep are small ruminants which ingest plant species ignored by other livestock species. Sheep can also eat some undesirable plants containing toxic alkaloids and glycosides, resulting in an increased level of liver enzymes (LIPIEC et al. 2015). The chemical composition of fodder is one of the most important factors determining the nutritional well-being of sheep under the free-range grazing system. Total protein, whose share in the sward ranges from 80 to 300 g kg^{-1} , plays an important role as it influences the proper digestion in the digestive tract of ruminants. Potassium (optimum 17 g kg⁻¹), phosphorus (3 g kg⁻¹), magnesium and sulphur (2 g kg^{-1}) are important macroelements from the perspective of the nutritional needs of animals. The content of these elements depends on many factors, mainly soil conditions, type of management or growth phase of plants (BORAWSKA-JARMUŁOWICZ 2003, KRZYWIECKI, KOZŁOWSKI 2003, BARYLA et al. 2009, KULIK 2009).

The study objective has been to assess the macroelement content in plants ingested and not ingested by sheep of the Świniarka breed. Regarded as being of low value, these plants occur in the sward of psammophilous grasslands in Kózki Nature Reserve. The studies provide an answer as to whether the species not ingested by the Świniarka sheep have a poorer chemical composition than ingested ones. Both groups of species occur in similar habitats all over Europe.

MATERIAL AND METHODS

Study area

The studies were conducted in the growing season of 2012, in Kózki Nature Reserve located in Podlaski Przełom Bugu Landscape Park near Binduga in Mazowieckie Province. The 82.1 ha Kózki Nature Reserve is located on the Bug River. In the southern part of the reserve (separated from the northern part by an old riverbed of the Bug), the soils are of alluvial origin, formed from sediments transported by the river. Poorly developed alluvial soils composed of loose sand are prevalent. In the study part, sheep of the native Świniarka breed, weighing 35-40 kg, grazed over an area of approx. 13.7 ha, as part of Package 7 of the agri-environmental scheme "Preservation of endangered genetic animal resources in agriculture". Being a typical primitive, multi-purpose breed with modest nutritional requirements, Świniarka sheep are characterised by good health and high resilience to disease. They cope very well in the difficult conditions of psammophilous grasslands (GRUSZECKI, LIPECKA 2007, KULIK et al. 2013, LIPIEC et al. 2015).

Field study

The Braun-Blanquet method was used to study the vegetation and identify the type of a plant community. The studies encompassed the dominant plant communities occurring in the grazing area. The adopted nomenclature of communities was according to MATUSZKIEWICZ (2008). The plant communities and the area occupied by them were mapped with a GPS receiver. In the third decade of June 2012, the aerial biomass of selected plant species (Armeria maritima, Calamagrostis epigejos, Carex praecox, Corynephorus canescens, Cynoglossum officinale, Dianthus deltoides, Juniperus communis, Koeleria glauca, Sedum acre and Thymus pulegioides) in three replications was cut in homogeneous phytocoenoses within the pasture. Species were collected from an area of 25 m² where phytosociological relevés were prepared.

Chemical analyses

The content of total protein, P, K, Mg and S was determined at the Institute of Agrophysics, PAS, in Lublin. Protein was determined using the Kjeldahl method and calculated by multiplying the total nitrogen content by a coefficient of 6.25. Phosphorus, magnesium, potassium and sulphur were determined by ICP OES according with the method described by OLESZEK et al. (2014). The tested samples were subjected to digestion in a mixture of 6 ml 65% HNO₃ and 1 ml 30% H_2O_2 , using a microwave mineralizer. Next, the liquid was converted to an aerosol using a nebulizer and then it was sprayed into the center of the plasma. The particles within the aerosol were dried, atomized, ionized, excited and relaxed. The amount of light emitted during relaxation is proportional to the concentration of the corresponding element in the solution. Next, the results were expressed in g kg⁻¹ of dry matter of the samples subjected to digestion. Nitrogen content for the analysed plant species is expressed in ecological indicator values according to ELLENBERG et al. (1992). Furthermore, based on the values above and the species composition of the plant communities distinguished (Table 1), the mean nitrogen content (trophism) index was calculated for them.

Table 1

Species	N _s	Phytosociological status according to MATUSZKIEWICZ (2008)	Community of species occurence	N _c
Armeria maritima	4	Ch.All. Vicio lathyroidis-Potentillion argenteae, Ch.Ass. Diantho-Armerietum elongate	All. Vicio lathyroidis- -Potentillion argenteae	2.848
Calamagrostis epigejos	6	Ch.Cl. Epilobietea angustifolii, Ch.Ass. Calamagrostietum epigeji; D.Ass. Corynephoro-Silenetum tataricae	Ass. Corynephoro- -Silenetum tataricae	5.868
Carex praecox	4	Ch.All. Cirsio-Brachypodion pinnati, Ch.Ass. Thalictro-Salvietum pratensis; D.All. Calluno-Arctostaphylion, D.All. Cnidion dubii, D.Ass. Violo- -Cnidietum dubii, D.Ass. Corynephoro- -Silenetum tataricae	Ass. Corynephoro- -Silenetum tataricae	5.868
Corynephorus canescens	2	Ch.Cl. Koelerio glaucae-Corynephoretea canescentis	Ass. Spergulo- -Corynephoretum	2.656
Cynoglossum officinale	7	ChSubAll. Onopordenion acanthii	community with Carex praecox bordering the patches of O. Onopordetalia acanthii communi- ties	4.820
Dianthus deltoides	2	Ch.All. Vicio lathyroidis-Potentillion argenteae, Ch.Ass. Diantho-Armerietum elongate	All. Vicio lathyroidis- -Potentillion argenteae	2.848
Juniperus communis	-	D.All. Dicrano-Pinion	Ass. Spergulo- -Corynephoretum	2.656
Koeleria glauca	1	Ch.All. Koelerion glaucae	All. Vicio lathyroidis- -Potentillion argenteae	2.848
Sedum acre	1	Ch.Cl. Koelerio glaucae-Corynephoretea canescentis	Ass. Corynephorion canescentis murawa szczotli- chowa	2.550
Thymus pulegioides	1	D.All. Vicio lathyroidis-Potentillion argenteae	All. Vicio lathyroidis- -Potentillion argenteae	2.848

Plant species and communities of their occurrence

N - nitrogen content value, ecological indicator acc. to Ellenberg et al. (1992);

 $\rm N_{_s}-nitrogen$ content for species; $\rm N_{_c}-nitrogen$ content for plant community

Data analysis

The chemical analysis results were subjected to the ANOVA analysis complemented by the Tukey test (p < 0.05). Using the pragmaTax program, cluster analysis was carried out for differences between the content of total protein and macroelements in the selected species.

RESULTS AND DISCUSSION

Phytosociological affiliation

Within the 13.7 ha grazing ground of the Świniarka sheep, there were floristically rich psammophilous grasslands of the Vicio lathyroidis-Potentillion argenteae alliance (4.8 ha), from which Armeria maritima, Dianthus deltoides, Thymus pulegioides and Koeleria glauca were collected, loose Spergulo-Corynephoretum grasslands (2.2 ha) featuring Corynephorus canescens, Juniperus communis and Sedum acre, dense grasslands of the poor form of the Corynephoro-Silenetum tataricae community (0.95 ha) with the predominance of *Calamagrostis epigejos* and a share of *Carex praecox* as well as small patches of ruderal vegetation adjoining grasslands with a share of Cynoglossum officinale (Table 1). C. officinale and C. epigejos showed the highest level of the trophism index (7 and 6, respectively). The former species was predominant in the association and, consequently, this community also showed the highest nitrogen content (5.868). The lowest indices were found for Koeleria glauca, Sedum acre and Thymus pulegioides (Table 1). This was not reflected in the index values for communities where these species occurred because they had a small percentage share. The bulk of the area was covered by poor grasslands with a low production potential. The remainder of the pasture (5.75 ha) consisted of vegetation of the Molinio-Arrhenatheretea class that was the best nutritional base for the sheep (WARDA et al. 2011, KULIK et al. 2013, 2014). In relation to such a patchwork character of the vegetation, it should be noted that in the threatened sand ecosystems, sheep grazing is an appropriate way to ensure vegetation dynamics owing to intermediate disturbances and prevention of grass encroachment or ruderalisation (Stroh et al. 2002).

Chemical composition

The biomass of the analysed plants showed varying protein and macroelement content. The significantly largest amount of protein was found in the biomass of *Cynoglossum officinale* (104.1 g kg⁻¹) while the lowest one was in *Sedum acre* (30.00 g kg⁻¹) and *Koeleria glauca* (30.60 g kg⁻¹) – Figure 1. Howeveer, *Cynoglossum officinale* did not occur often. Besides, it is a toxic species because it contains tumorigenic pyrrolizidine alkaloids (FU et al. 2002). Livestock and human poisoning upon consuming plants containing pyrrolizi-



Fig. 1. Mean total protein content in plant biomass: Arm mar – Armeria maritima, Cal epi – Calamagrostis epigejos, Car pra – Carex praecox, Cor can – Corynephorus canescens, Cyn off – Cynoglossum officinale, Dia del – Dianthus deltoids, Jun com – Juniperus communis, Koe gla – Koeleria glauca, Sed acr – Sedum acre, Thy pul – Thymus pulegioides

dine alkaloids has been reported in many countries. The poisoned animal species include horses, cattle, sheep and goats (DE LANUX-VAN GORDER 2000). In comparison with the analysed species, *Calamagrostis epigejos* had a high protein content (73.15 g kg⁻¹) and was abundant in patches of a degraded Corynephoro-Silenetum tataricae association. Sheep grazing reduced the cover-abundance of this expansive plant and enhanced the habitat-specific phytodiversity (Süss et al. 2004, EICHBERG et al. 2007, Süss, SCHWABE 2007, KULIK et al. 2013). The sheep willingly ingested this species but only in the early development phase before the formation of inflorescences (WARDA et al. 2011). However, compared with a typical grassland-pasture sward, the tested species had a very low protein content and therefore did not constitute valuable fodder. The general protein content in such biomass ranges from 80 to more than 300 g kg⁻¹, depending on fertilisation, sward species composition, habitat conditions, harvesting time, development phase and kind of use (Bo-RAWSKA-JARMUŁOWICZ 2003, KRZYWIECKI, KOZŁOWSKI 2003, BARYŁA et al. 2009, Kulik 2009).

The significantly highest phosphorus content was found in the biomass of *Cynoglossum officinale* (5.365 g kg⁻¹), while the smallest one was in *Koeleria glauca* (1.750 g kg⁻¹) – Figure 2. Phosphorus plays an important role in photosynthesis, respiration, metabolism of lipids and in nitrogen transformations. According to FALKOWSKI et al. (2000), about 3 g kg⁻¹ DM in animal fodder is the optimum content from the perspective of their nutritional needs, while GAWEL (2011) suggests a wider range of optimum values, i.e. from 2.8 to 3.6 g kg⁻¹. Most of the analysed species are characterised by a lower phosphorus content than the quoted authors recommend. The phosphorus content in biomass is lower in dry habitats or in periods of drought (FALKOWSKI et al. 2000, KULIK 2009).



Fig. 2. Mean phosphorus content in plant biomass. Explanations: cf. Figure 1

Potassium is another very important mineral that has a positive impact on photosynthesis and water management in plants. The optimum content in fodder is about 17 g kg⁻¹ DM (FALKOWSKI et al. 2000, GAWEL 2011). The biomass of *Dianthus deltoides* is characterised by the significantly highest potassium content (16.71 g kg⁻¹), while the lowest (2.184 g kg⁻¹) occurs in *Armeria maritima* (Figure 3). All values are lower than the optimum content. It should be noted that grazing leads to higher potassium content in soil and plant biomass, especially in places covered by animal faeces (WARDA 1994, KULIK 2009). Thus, the potassium content in vegetation without grazing can be lower.





Fig. 3. Mean potassium content in plant biomass. Explanations: cf. Figure 1

Magnesium, another element analysed, affects photosynthesis, phosphorus management and the formation of protein compounds in plants. About 2 g kg⁻¹ DM in fodder is the optimum content considering the animals' nutritional needs (FALKOWSKI et al. 2000). The biomass of typical psammophilous grassland grasses such as *Koeleria glauca* (0.727 g kg⁻¹) and *Corynephorus canescens* (0.790 g kg⁻¹) as well as *Calamagrostis epigejos* (1.060 g kg⁻¹) showed the significantly lowest magnesium content (Figure 4). The values for





Fig. 4. Mean magnesium content in plant biomass. Explanations: cf. Figure 1

these grasses were lower than the optimum content in fodder. The significantly highest values were found in the biomass of *Thymus pullegioides* $(3.633 \text{ g kg}^{-1})$ and *Cynoglossum officinale* $(3.463 \text{ g kg}^{-1})$. Dicotyledons usually accumulate bigger amounts of this element in comparison to grasses. Moreover, the biomass of plants on mineral soils had a smaller magnesium content (KULIK 2009, KULIK et al. 2014).

The significantly highest sulphur content occurred in the biomass of Armeria maritima (1.862 g kg⁻¹) and Calamagrostis epigejos (1.741 g kg⁻¹), while the lowest one was in Juniperus communis (0.814 g kg⁻¹), Thymus pullegioides (0.839 g kg⁻¹), Koeleria glauca (0.883 g kg⁻¹) and Corynephorus canescens (0.898 g kg⁻¹) – Figure 5. Sulphur is an essential element for all plants; its deficiency can cause lower yields and usually occurs in light soils. The optimum sulphur content in fodder is about 2 g kg⁻¹, while the critical value which can inhibit the development of grasses is 1 g kg⁻¹. Furthermore, sulphur content decreases with the development of plants (FALKOWSKI et al. 2000). It should be noted that plant biomass was collected in the third decade of June. All the tested plants were characterised by a very small sulphur content, which fell below the optimum level or even reached the critical level (Figure 5), which confirmed low productivity of these plant communities (WARDA et al. 2011, KULIK et al. 2013).

When analysing the chemical composition of all species versus the nutritional preferences of sheep, it has to be noted that the species that were ignored by the grazing animals (*Corynephorus canescens* and *Sedum acre*) belong to the



••••• the optimum content in fodder acc. to FALKOWSKI et al. (2000)

Fig. 5. Mean sulphur content in plant biomass. Explanations: cf. Figure 1

group of nutrient-poor species (Figure 6). *Corynephorus canescens* is characterised by the significantly lowest content of protein, phosphorus, magnesium and sulphur, while *Sedum acre* is poor in protein, phosphorus and sulphur (Figures 2-5). These are typical species with a xeromorphic structure. Therefore, taking into account the avoidance of *Corynephorus canescens* by sheep and the loose structure of grasslands with the predominance of this species, lower numbers of sheep should be planned in such habitats so as to prevent their degradation.





Moreover, the behaviour of the grazing animals on pasture depends on the time in the growing season and weather conditions, particularly precipitation (CHRUPEK et al. 2006). This highlights the importance of maintaining a vegetation patchwork providing sheep with fodder of varied quality.

CONCLUSIONS

The analysed plant species occurred in the floristically rich psammophilous grasslands of the *Vicio lathyroidis-Potentillion argenteae* alliance, *Spergulo-Corynephoretum* association and community with the predominance of *Calamagrostis epigejos* but, at the same time, these communities had poor chemical composition, which was confirmed by the analyses of the biomass of the particular species and the Ellenberg's indicator values.

The biomass of psammophilous grasslands was characterised by a very low content of protein and phosphorus, potassium, magnesium and sulphur, in most cases below the optimum levels from the perspective of the nutritional requirements of animals.

The biomass of *Cynoglossum officinale* had the significantly highest content of protein, phosphorus and magnesium, which could have been the reason why this species, despite its poisonous properties, was ingested by the Świniarka sheep under the poor conditions of psammophilous grasslands.

Species typical of psammophilous grasslands, such as *Corynephorus canescens* or *Koeleria glauca*, had the lowest nutritional value. *C. canescens* and *Sedum acre* were not ingested by sheep.

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