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EFFECTIVENESS OF FERTILIZATION OF *DACTYLIS GLOMERATA* AND *FESTULOLIUM BRAUNII* WITH NITROGEN AND THE BIOSTIMULANT KELPAK SL

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ABSTRACT

The objective of the present work was to determine the influence of nitrogen fertilization and Kelpak SL application on the yield and nitrogen content of *Dactylis glomerata* and *Festulolium braunii*, and to assess the effectiveness of both the treatments. A field experiment was arranged as a randomised sub-block design (split-split-plot) with three replicates. The following factors were examined: a) nitrogen rate: 50, 100 and 150 kg·ha^{-1} and a control (0 kg N·ha^{-1}); b) biostimulant with the trade name Kelpak SL applied at 2 dm^{3}.ha^{-1} and a control – no biostimulant; c) pure stands of grass species grown in monoculture; - orchard grass (*Dactylis glomerata)*, cv. Amila, - Braun's festulolium *(Festulolium braunii)*, cv. Felopa. Increasing nitrogen rates significantly enhanced yields of grasses, nitrogen content and uptake by plants, but decreased agricultural effectiveness and physiological effectiveness. An application of the biostimulant Kelpak favourably affected the experimental characteristics. Physiological effectiveness was the only trait to decline following Kelpak application. The grasses tested in the experiment differed as to the parameters analysed in the study. The performance of *Dactylis glomerata* was better as far as yield, nitrogen uptake, and indicator of agricultural effectiveness were concerned.

Key words: seaweed extract, grass, nitrogen uptake, content of nitrogen, crop yields, *Ecklonia maxima.*

INTRODUCTION

Fertilization, nitrogen application in particular, is a factor which determines the level and quality of grassland yield. Nitrogen plays a major role in increasing plant production and its deficiency is believed to be the most frequent factor restricting plant growth (Zotarelli et al., 2008). According to many authors (Kruczek and Szulc, 2000; Małecka and Blecharczyk, 2005; Ciepiela et al., 2012), effectiveness of nitrogen fertilization is displayed in the quantitative and qualitative yield characteristics, as well as physiological effectiveness (plant's ability to convert nitrogen supplied with fertilizers to usable yield) and agricultural effectiveness (yield increase per unit of N applied with fertilizers).

Seaweed extracts find a wide application range in agriculture worldwide and interest in these products is due to the pro-ecological tendencies in crop growing and an urge to take care of natural environment and food safety. *Ecklonia maxima* is a seaweed which is used to manufacture a product applied in crop plant fertilization. The plant is a brown alga, also called 'sea bamboo', which is sourced from the coastline of South Africa where it dominates offshore kelp forests. When stranded on the shore, it pollutes the environment. *Ecklonia maxima* fronds are used to produce an extract which contains the phytohormones: auxins and cytokinins. Some reports (Stirk and Van Staden 1997) indicate that cytokinins are more important from the point of view of the stimulation of the growth and development of crops fertilized with this extract. Moreover, the extract contains other compounds, e.g.: polysaccharides (not to be found in terrestrial plants), amino acids, polyphenols, macroelements and microelements, which have a great influence on life processes in crop plants (Connan et al., 2006; Craigie, 2011; Haider et. al., 2012; Khan et al., 2009; Norrie and Keathley, 2006; Oyoo et. al., 2010). The effect of seaweed application depends on many factors, including crop plant species (Zodape, 2001; Ferreira and Lourens, 2002).

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Scientific literature lacks works on the effectiveness of natural biostimulants applied to fodder grasses, which was an incentive to conduct the research presented in this paper. The objective of the present work was to determine the influence of nitrogen fertilization and Kelpak SL application on the yield and nitrogen content of *Dactylis glomerata* and *Festulolium braunii*, and to assess the effectiveness of both treatments.

MATERIAL AND METHODS

A field experiment was arranged as a randomised sub-block design (split-split-plot) with three replicates at the Siedlce Experimental Unit of the Siedlce University of Natural Sciences and Humanities (Poland) in late April, 2009. The plot area was 10 m^2. The study was conducted in 2009-2012. The soil of the experimental site represents average soils, Hortic Anthrosol (WRB). Prior to the experiment set-up the characteristics of the soil were as follows: neutral pH (pH in 1n KCl=7.2), high humus content (3.78%), high available phosphorus and magnesium contents (P_2O_5 – 900 $mg \cdot kg^{-1}$, Mg – 84 $mg \cdot kg^{-1}$). Total nitrogen content and available potassium content in the soil averaged, respectively, 1.8 $g \cdot kg^{-1}$ d.m. and 190 mg $K_2O \cdot kg^{-1}$. Soil chemical analysis was carried out at an accredited laboratory of the Chemical and Agricultural Research Laboratory in Warsaw (Poland). Available phosphorus and potassium in the soil were extracted by means of the Egner-Riehm method (Staugaitis and Rutkauskiene, 2010) and available magnesium – using the Schachtschabel method (Staugaitis and Rutkauskiene, 2010). Phosphorus was determined by the colorimetric method, total nitrogen by the Kjeldahl method and potassium and magnesium by the atomic absorption spectrophotometry AAS.

The following factors were examined:

- nitrogen rate: 50, 100 and 150 $kg \cdot ha^{-1}$ and a control (0 kg $N \cdot ha^{-1}$);
- biostimulant with the trade name Kelpak SL applied at 2 $dm^3 \cdot ha^{-1}$ and a control – no biostimulant;
- pure stands of grass species grown in monoculture;
- orchard grass (*Dactylis glomerata*), cv. Amila;
- Braun's festulolium *(Festulolium-braunii*), cv. Felopa.

The growth stimulant applied in the experiment is an extract from the fastest growing seaweed (kelp) *Ecklonia maxima* harvested the coast of South Africa. The extract contains, among others, the natural plant hormones auxins (11 $mg \cdot l^{-1}$) and cytokinins (0.03 $mg \cdot l^{-1}$). The commercial name of the stimulant is Kelpak SL and it is manufactured by Kelp Products (Pty) Ltd P.O. Box 325, Simon's Town, the Republic of South Africa.

The growth regulator was not applied in the year when the experiment was established (2009). The season was an introductory period when three weed-control cuttings were made. After the second cutting, mineral fertilization was applied to all the plots at the rates of 30 $kg.ha^{-1}$ N (ammonium nitrate) and 30 $kg \cdot ha^{-1}$ K_2O (potash salt). Phosphorus was not applied as the soil was rich in available forms of this element. Three cuts were harvested in each study year (2010-2012). Ammonium nitrate was applied three times per year. The total nitrogen (50, 100 and 150 $kg \cdot ha^{-1}$) amount was split into three equal rates which were applied before each cutting. Phosphorus and potassium needs of the grass were calculated taking into account the expected dry matter yields, the appropriate mineral (from the ruminant nutrition standpoint) contents of hay and soil P and K availability. Moreover, to determine phosphorus and potassium application rates, coefficients given by Fotyma and Mercik (1995) were used to convert the amounts of the nutrients taken up by grass and yields into the rates of phosphorus and potassium fertilizers. P and K fertilization was applied to all the plots. Phosphorus was applied once as triple superphosphate at a rate of 40 $kg \cdot ha^{-1}$ P_2O_5 in the spring. The amount of potassium (160 $kg \cdot ha^{-1}$ K_2O) was split into three equal rates and applied prior to each cutting as 60% potash salt. The seaweed extract was sprayed as an aqueous solution; the rate was 2 dm^3 of biostimulant per hectare diluted in water to 400 dm^3. The spraying was performed before each cutting: the first application was three weeks before the first cutting, the second two weeks after the first harvest, and the last three weeks after the second harvest. Plots with no biostimulant were water sprayed (400 $dm^{3} \cdot ha^{-1}$).

During harvest of each cut, green matter from each plot (its whole area, that is 10 m^2) was weighed to determine its yield and 0.5 kg green matter samples of grasses were taken to obtain the drying-up coefficient and carry out chemical analyses. The samples were left to dry in a ventilated room. Airy dry matter was weighed (to determine airy dry matter yield per plot) and then it was shredded and ground. The obtained material was subjected to chemical analysis to determine absolutely dry matter, and N-total. Absolutely dry matter was determined by the gravimetric method; air-dry matter samples were dried in a drying oven at 105° C till constant weight was achieved. Total nitrogen was determined by the Kjeldahl method.

Grass dry matter yield was calculated according to the formula:

$$Y_{DM} = Y_{ADM} + Z_{ADM},$$

where: Y_{DM} – dry matter yield;
Y_{ADM} – airy dry matter yield;
Z_{ADM} – absolutely dry matter content in air-dry matter.

Effectiveness indicators were calculated according to the following formulae (Fotyma and Mercik, 1995):

$$P = Y \times Z_N,$$

$$Er = (Y_N - Y_0)/N,$$

$$Ef = (Y_N - Y_0)/(P_N - P_0),$$

where: P – nitrogen uptake with plant yield;
Er – agricultural effectiveness;
Ef – physiological effectiveness;
Y – plant yield;
Z_N – nitrogen content;
Y_N – yield in plots where N was applied;
Y_0 – control yield (no nitrogen and biostimulant);
N – nitrogen rate applied to unit Y_N;
P_N – nitrogen uptake with plant yield in unit Y_N;
P_0 – nitrogen uptake with plant yield in control (no nitrogen and biostimulant).

All the indicators were calculated based on the total yield of three cuts, means across three cuts, total nitrogen content and overall nitrogen rate (applied per year).

Statistical analysis of results was conducted using the program STATISTICA, StatSoft, Inc. (2011), version 10 (www.statsoft.com). Significance of differences between means describing the experimental factors was checked by the Tukey's test as the significance level of $\alpha \leq 0.05$.

Weather conditions differed during the study period (Table 1). Average air temperatures and precipitation sums in all the growing seasons were higher than the long-term means and the precipitation was very unevenly distributed. In 2010 and 2011 rainfall was, respectively, by 115.3 and 80.5 mm higher than the long-term means. It is worth noticing that in July 2010 the precipitation was 4.5 times higher than the long-term mean for July, and it constituted 48% rainfall of the whole growing season. By contrast, high rainfall shortages were recorded in April 2010 and September 2011.

RESULTS AND DISCUSSION

Plant yield, which is an outcome of the effect of many various factors, is the major criterion of assessing plant fertilization effectiveness. The results obtained indicated that yields harvested in individual study years differed significantly for all characteristics examined.

The greatest yields were produced by grasses in the first study year (13.2 $t \cdot ha^{-1}$) and the lowest in the third study year (8.0 $t \cdot ha^{-1}$). Grass yield quantity depends on not only the amount and quality of elements supplied with fertilizers but also on weather conditions. Thus, it can be assumed that such low yields in the third study year were due to low atmospheric precipitation and its very uneven distribution (Table 1).

Table 1. Meteorological condition in growing season 2010-2012 by meteorological station in Siedlce

Years	Means air temperatures (°C)						Mean daily air temperature in growing season (°C)
	IV	V	VI	VII	VIII	IX	
2010	8.9	14.0	17.4	21.6	19.8	11.8	15.6
2011	9.8	13.4	18.1	18.2	18.1	14.4	15.3
2012	9.0	14.5	16.4	20.4	18.0	14.2	15.4
Means of many years (2002- 2012)	7.7	10.0	16.1	19.3	18.0	13.0	14.0
Years	Monthly precipitations (mm)						Sum of precipitation in growing season (mm)
	IV	V	VI	VII	VIII	IX	
2010	10.7	93.2	62.6	77.0	106.3	109.9	459.7
2011	38.1	55.6	44.3	204.2	55.4	26.6	424.2
2012	40.3	59.7	118.7	41.4	64.1	30.8	355.0
Means of many years (2002- 2012)	52.3	50.0	68.2	45.7	66.8	60.7	343.7

Reported by the meteorological station in Siedlce.

It should also be noted that *Dactylis glomerata* was more stable in terms of yielding in the first two study years (2010-2011). Yielding stability of this species has also been reported in other scientific papers (Ciepiela, 2004; Łyszczarz et al., 2003). Moreover, the grasses examined represent species which rapidly develop after sowing so they can be used to establish short-lived grasslands in ploughed fields. These species produce the best yields in the first full-use year whereas in the subsequent years the yields tend to decline (Falkowski et al., 1994). Analysis of results averaged across three years demonstrated that all the factors examined in the experiment significantly affected yields of the grasses (Table 2). Increasing nitrogen rates contributed to increased plant yields in all the study years, regardless of grass species or biostimulant application. Yields of grasses harvested in plots fertilized with 150 kg·N ha^{-1} increased by 76%, on average, compared with yields in non-fertilized plots.

Kelpak application significantly increased yields of the grasses (by 11.2%, on average), regardless of the remaining factors. Similar results were obtained by Ciepiela et al. (2013). In their study, grass yield increase exceeded 10% following an application of *Ecklonia maxima* extract. However, detailed analysis of the results, which included interaction of all the experimental factors, demonstrated that, in each study year, the biostimulant Kelpak SL insignificantly affected the dry matter yield of *Dactylis glomerata* fertilized with 50 and 100 kg N·ha^{-1}. Similar response was observed for *Festulolium braunii* in non-fertilized plots and plots where 50 kg N·ha^{-1} had been applied in 2011 and 2012. Also, the effect of the biostimulant on individual grassy species was different in successive study years. Nevertheless, when dry matter yields, averaged across the study years, were taken into account, the biostimulant Kelpak SL was found to significantly increase *Festulolium braunii* yields in both non-N fertilized plots and plots where 100 and 150 kg N·ha^{-1} had been applied. *Dactylis glomerata* yield increased significantly following an application of biostimulant only in the control (0 kg N·ha^{-1}) and in plots fertilized with 150 kg N·ha^{-1}. Many authors reported a beneficial effect of seaweed extract on yield performance of various plant species (Kocira et al., 2013; Haider et al., 2012, Khan et al., 2009; Kumar and Sahoo, 2011). However, this inference cannot be extended to include all species, because the impact of natural biostimulants is to a large degree species - or even cultivar - dependent (Sultana et al., 2005). The results obtained in the study described here demonstrated that the effect of seaweed extract, at the background of different nitrogen rates, resulted is a significant yield increase in the first two study years, no significant differences being found in the third study year.

Table 2. Dry matter yield ($t \cdot ha^{-1}$) of *Dactylis glomerata* and *Festulolium braunii*
(Total of three cuts)

Year	Dose of N ($kg \cdot ha^{-1}$)	*Dactylis glomerata*		*Festulolium braunii*		Mean		*Dactylis glomerata*	*Festulolium braunii*	Mean
		Treatment		Treatment		Treatment				
		I	II	I	II	I	II	Mean		
2010	0	9.5 a	10.6 b	9.8 a	10.9 b	8.6 a	10.3 b	10.2 A	8.7 A	9.4 A
	50	12.6 a	13.3 a	11.7 a	13.4 b	11.8 a	12.5 a	13.1 B	11.3 B	12.2 B
	100	14.5 a	14.8 a	14.0 a	15.9 b	13.9 a	14.4 a	14.6 C	13.7 C	14.2 C
	150	16.0 a	19.0 b	15.9 a	17.2 b	16.0 a	18.1 b	17.5 D	16.7 D	17.0 D
2011	0	7.8 a	9.9 b	4.2 a	5.0 a	6.0 a	7.4 b	8.9 A	4.6 A	6.7 A
	50	11.2 a	11.5 a	5.8 a	5.9 a	8.5 a	8.7 a	11.4 B	5.6 B	8.6 B
	100	12.5 a	13.3 a	7.6 a	9.2 b	10.0 a	11.3 b	12.9 C	8.4 C	10.7 C
	150	14.0 a	16.5 b	11.0 a	11.9 b	12.5 a	14.2 b	15.2 D	11.5 D	13.3 D
2012	0	7.0 a	7.6 a	5.4 a	6.0 a	6.2 a	6.8 a	7.3 A	5.7 A	6.5 A
	50	8.0 a	8.4 a	6.4 a	6.9 a	7.2 a	7.6 a	8.2 B	6.6 B	7.4 B
	100	8.7 a	9.1 a	7.2 a	8.1 b	8.0 a	8.6 a	8.9 B	7.7 C	8.3 C
	150	9.8 a	11.0 b	8.8 a	9.6 a	9.3 a	10.3 a	10.4 C	9.2 D	9.8 D
Mean	0	8.1 a	9.5 b	5.7 a	6.9 b	6.9 a	8.2 b	8.8 A	6.3 A	7.6 A
	50	10.7 a	11.1 a	7.7 a	8.2 a	9.2 a	9.6 a	10.9 B	7.9 B	9.4 B
	100	11.9 a	12.4 a	9.4 a	10.4 b	10.7 a	11.4 b	12.1 C	9.9 C	11.0 C
	150	13.3 a	15.5 b	11.9 a	12.9 b	12.6 a	14.2 b	14.4 D	12.4 D	13.4 D
2010	Mean	13.2 a	14.5 b	12.0 a	13.2 b	12.6 a	13.8 b	13.9 A	12.6 A	13.2 A
2011		11.4 a	12.8 b	7.2 a	8.0 a	9.3 a	10.4 b	12.1 B	7.6 B	9.8 B
2012		8.4 a	9.0 a	7.0 a	7.6 a	7.7 a	8.3 a	8.7 C	7.3 B	8.0 C
Mean		11.0 a	12.1 b	8.7 a	9.6 b	9.8 a	10.9 b	11.6 a	9.2 b	10.4

I – without biostimulant (control); II – with biostimulant Kelpak SL (2 $dm^3 \cdot ha^{-1}$).
The values in the lines for the individual factors and their interaction indicated with different small letters differ significantly.
The values in columns for individual factors and their interaction indicated with different capital letters differ significantly.

Dactylis glomerata produced the best yields when treated with the biostimulant and fertilised with nitrogen at the rate of 150 $kg \cdot ha^{-1}$. Yield of both tested species were significantly higher following an application of the biostimulant.

The average total nitrogen content in the grasses (Table 3) amounted to 21.1 $g \cdot kg^{-1}$. It should be pointed out that although nitrogen (and thus protein) content of both the species was high, it did not exceed the optimum N concentration in cattle feed (Gaweł, 2011). Nitrogen fertilization in each study year significantly increased nitrogen content in the plants tested compared with control. In his study, Czapla (2000) demonstrated a negative association between high nitrogen rates and nitrogen content of the sward. Worldwide, it is claimed in literature that seaweed extract application to agricultural crops increases nitrogen concentration in the plants (Mancuso et al., 2006; Abdel-Mawgoud et al., 2010; Godlewska and Ciepiela, 2013; Pramanick et al., 2014), which has also been confirmed in the present study. An application of Kelpak significantly increased nitrogen content in both grassy species, regardless of the remaining factors, the increase approximating 10%. An application of the biostimulant to both species significantly increased plant content of nitrogen, more N being accumulated by *Festulolium braunii.*

Table 3. The content of total nitrogen (g kg^{-1} d.m.) in *Dactylis glomerata* and *Festulolium braunii* (mean from three cuts)

Year	Dose of N (kg.ha^{-1})	*Dactylis glomerata* Treatment I	*Dactylis glomerata* Treatment II	*Festulolium braunii* Treatment I	*Festulolium braunii* Treatment II	Mean Treatment I	Mean Treatment II	Mean *Dactylis glomerata*	Mean *Festulolium braunii*	Mean
2010	0	16.1 a	18.2 b	18.4 a	20.3 b	17.2 a	19.2 b	17.2 A	19.3 A	18.2A
	50	18.9 a	20.3 b	20.3 a	22.0 b	19.6 a	21.2 b	19.6 B	21.1 B	20.4 B
	100	20.3 a	22.1 b	21.7 a	23.6 b	21.0 a	22.9 b	21.2 C	22.7 C	21.9 C
	150	22.1 a	24.8 b	24.1 a	26.6 b	23.1 a	25.7 b	23.4 D	25.4 D	24.4 D
2011	0	15.5 a	17.6 b	17.5 a	19.2 b	16.5 a	18.4 b	16.6 A	18.3 A	17.5 A
	50	17.9 a	19.7 b	19.9 a	22.3 b	18.9 a	21.0 b	18.8 B	21.1 B	20.0 B
	100	19.5 a	21.8 b	21.6 a	23.7 b	20.6 a	22.8 b	20.7 C	22.6 C	21.7 C
	150	21.8 a	24.4 b	24.3 a	26.1 b	23.1 a	25.3 b	23.1 D	25.2 D	24.2 D
2012	0	16.2 a	18.0 b	18.5 a	20.2 b	17.3 a	19.1 b	17.1 A	19.4 A	18.2 A
	50	18.4 a	20.4 b	20.2 a	23.0 b	19.3 a	21.7 b	19.4 B	21.6 B	20.5 B
	100	19.8 a	21.7 b	22.4 a	24.3 b	21.1 a	23.0 b	20.8 C	23.3 C	22.1 C
	150	21.8 a	24.8 b	24.6 a	26.6 b	23.2 a	25.7 b	23.3 D	25.6 D	24.4 D
Mean	0	15.9 a	18.0 b	18.1 a	19.9 b	17.0 a	18.9 b	16.9 A	19.0 A	18.0 A
	50	18.4 a	20.1 b	20.1 a	22.4 b	19.3 a	21.3 b	19.3 B	21.3 B	20.3 B
	100	19.9 a	21.9 b	21.9 a	23.9 b	20.9 a	22.9 b	20.9 C	22.9 C	21.9 C
	150	21.9 a	24.7 b	24.3 a	26.4 b	23.1 a	25.6 b	23.3 D	25.4 D	24.3 D
2010	Mean	19.3 a	21.4 b	21.1 a	23.1 b	20.2 a	22.3 b	20.4 A	22.1 A	21.2 A
2011		18.7 a	20.9 b	20.8 a	22.8 b	19.8 a	21.9 b	19.8 A	21.8 A	20.8 A
2012		19.0 a	21.2 b	21.4 a	23.5 b	20.2 a	22.4 b	20.1 A	22.5 A	21.3 A
Mean		19.0 a	21.2 b	21.1 a	23.6 b	20.1 a	22.2 b	20.1 a	22.1 b	21.1

Explanation see Table 2.

Also, significant interactions between the factors studied were found. In all the study years, an application of the biostimulant significantly increased total nitrogen content in both grassy species fertilized with each nitrogen rate.

Nitrogen uptake with plant yield varied and depended on nitrogen rate and biostimulant application (Table 4). When N rate was 150 kg ha^{-1}, the uptake increased by 140% compared with the control plants.

Moreover, increasing nitrogen rates were found to significantly increase nitrogen uptake with plant yield. Also Ciepiela et al. (2008) demonstrated that nitrogen uptake with yield of grasses significantly depended on nitrogen rates. Biostimulant application increased nitrogen uptake as well (by 21.9%, on average). Unfortunately, lack of literature on the subject makes it impossible to discuss the finding. An interaction of biostimulant and nitrogen fertilization was observed in plots fertilized with the following nitrogen rates: 0, 100 and 150 kg.ha^{-1} (mean across years). Nitrogen uptake by plants cultivated in these plots increased by 31.6, 17.6 and 24.8%, respectively. In contrast, no significant differences in nitrogen uptake between the grasses tested were found.

The highest nitrogen uptake by plants occurred in the first study year, which was due to the best yield performance of the grasses and the most favorable weather conditions compared with the remaining study years.

Agricultural effectiveness is a measure of efficacy of crop fertilization with nitrogen, expressed in terms of yield increase per 1 kg N supplied with fertilizers (Ciepiela et al., 2012 after Fotyma, 1997). In the present study, the value of the aforementioned indicator ranged between 17 and 76 kg d.m. kg^{-1} N (Table 5). It was the lowest in the third study year for plots fertilized with 100 kg N.ha^{-1} and no biostimulant applied, and the highest in the first study year for biostimulant-treated units fertilized with 50 kg N.ha^{-1}. The results indicate that agricultural effectiveness of nitrogen fertilizer application to grasses declines as the rate of this nutrient increases, regardless of the study year and biostimulant application. This finding has been confirmed in studies by other authors (Ciepiela et al., 2012; Kitczak, 1997).

Table 4. Total nitrogen uptake ($kg \cdot ha^{-1}$) with the yield *Dactylis glomerata* and *Festulolium braunii* (mean from three cuts)

Year	Dose of N ($kg \cdot ha^{-1}$)	*Dactylis glomerata*		*Festulolium braunii*		Mean		*Dactylis glomerata*	*Festulolium braunii*	Mean
		Treatment		Treatment		Treatment				
		I	II	I	II	I	II	Mean		
2010	0	153 a	198 b	140 a	199 b	147 a	198 b	176 A	169 A	173 A
	50	243 a	271 b	220 a	258 b	231 a	264 b	257 B	239 B	248 B
	100	294 a	326 b	291 a	331 b	293 a	329 b	310 C	311 C	311 C
	150	353 a	473 b	384 a	459 b	368 a	466 b	413 D	421 D	417 D
2011	0	121 a	175 b	73 a	95 a	97 a	135 b	148 A	84 A	116 A
	50	201 a	227 b	116 a	131 b	159 a	179 a	214 B	124 B	169 B
	100	243 a	291 b	164 a	219 b	204 a	255 b	267 C	192 C	229 C
	150	305 a	403 b	269 a	311 b	287 a	357 b	353 D	290 D	322 D
2012	0	114 a	137 b	100 a	121 a	107 a	129 b	126 A	111 A	118 A
	50	147 a	171 b	129 a	157 b	138 a	164 b	159 B	143 B	151 B
	100	172 a	199 b	162 a	196 b	167 a	197 b	185 B	179 C	182 B
	150	214 a	274 b	215 a	255 b	215 a	265 b	244 C	236 D	240 C
Mean	0	129 a	170 b	105 a	138 a	117 a	154 b	150 A	121 A	136 A
	50	197 a	223 a	155 a	182 a	176 a	203 a	210 B	169 B	189 B
	100	237 a	272 a	206 a	249 b	221 a	260 b	254 B	227 C	241 C
	150	291 a	383 b	289 a	342 b	290 a	362 b	337 C	316 D	326 D
2010	Mean	261 a	317 b	259 a	312 b	260 a	314 b	289 A	285 A	287 A
2011		218 a	274 b	156 a	189 a	187 a	231 b	246 B	172 B	209 B
2012		162 a	195 a	152 a	183 a	157 a	189 a	178 C	167 BC	172 BC
Mean		213 a	262 b	189 a	228 b	201 a	245 b	238 a	208 a	223

Explanation see Table 2

Table 5. Agricultural of efficiency fertilization of *Dactylis glomerata* and *Festulolium braunii* with nitrogen and the biostimulant Kelpak (kg d.m. $\cdot kg^{-1}$ N)

Year	Dose of N ($kg \cdot ha^{-1}$)	*Dactylis glomerata*		*Festulolium braunii*		Mean	
		Treatment		Treatment		Treatment	
		I	II	I	II	I	II
2010	50	A 62.0 a	A 76.0 b	A 38.0 a	A 72.0 b	A 50.0 a	A 74.0 b
	100	B 50.0 a	B 53.0 a	A 42.0 a	B 61.0 b	AB 46.0 a	B 57.0 b
	150	C 43.3 a	C 63.3 b	A 40.7 a	C 49.3 b	B 42.0 a	B 56.3 b
2011	50	A 68.0 a	A 74.0 b	A 32.0 a	A 34.0 a	A 50.0 a	A 54.0 a
	100	B 47.0 a	B 55.0 b	A 34.0 a	B 50.0 b	B 40.5 a	A 52.5 b
	150	C 41.3 a	B 58.0 b	B 45.3 a	B 51.3 b	B 43.3 a	A 54.7 b
2012	50	A 20.0 a	A 28.0 b	A 20.0 a	A 30.0 b	A 20.0 a	A 29.0 b
	100	A 17.0 a	B 21.0 a	A 18.0 a	A 27.0 b	A 17.5 a	A 24.0 b
	150	A 18.7 a	AC 26.7 b	A 22.7 a	A 28.0 a	A 20.7 a	A 27.3 b
Mean	50	A 50.0 a	A 59.3 b	A 30.0 a	A 45.3 b	A 40.0 a	A 52.3 b
	100	B 38.0 a	B 43.0 a	AB 31.3 a	A 46.0 b	A 34.7 a	B 44.5 b
	150	BC 34.4 a	C 49.3 b	B 36.2 a	A 42.9 b	A 35.3 a	BC 46.1 b
Mean		40.8 a	50.6 b	32.5 a	44.7 b	36.7 a	47.6 b

Explanation see Table 2.

Seaweed extract application contributed to a significant increase in agricultural effectiveness of nitrogen fertilization. The value of the indicator increased by 29.7% following sprying with the biostimulant. However, it is not possible to relate this finding to literature data due to lock of such information.

Statistical analysis including interaction of the experimental factors revealed that the biostimulant did not significantly affect the agricultural effectiveness of *Dactylis glomerata* fertilized with 100 kg N·ha^{-1} in 2010 and 2012. In case of *Festulolium braunii*, its agricultural effectiveness did not differ following an application of Kelpak SL in plots fertilized with 50 kg N·ha^{-1} in 2011 and 150 kg N·ha^{-1} in 2012.

Physiological effectiveness (increase in yield per unit of nitrogen taken up by plants from soil and fertilizers) is a measure of the ability of plants to convert the nitrogen to usable yield, and it indicates how efficient the processes of nitrogen management in the plant are (Kruczek, 2000). The value of physiological effectiveness was lower than agricultural effectiveness, which does not agree with findings reported by Ciepiela et al. (2009). The disagreement was probably due to the fact that too much nitrogen was taken up by plants, which were unable to convert it into yield. Detailed analysis of the values obtained, taking into consideration nitrogen rates, study years, biostimulant and plant species, demonstrated that physiological effectiveness declined as nitrogen rates increased (Table 6). However, for the majority of fertilization combinations, there was no significant effect of nitrogen rates on values of the indicator discussed here. A significant decline in physiological effectiveness following an application of Kelpak was also observed. Regardless of the remaining experimental factors, seaweed extract application contributed to a decline in the value of the indicator, by 10%, on average. It can be explained by the fact that, when the same nitrogen rate had been applied, biostimulant-treated grasses took up much more nitrogen compared with non-treated plants and their ability to convert this nitrogen taken up from the soil and fertilizers might have been insufficient. Probably, the nitrogen was accumulated in plants as non-protein nitrogen compounds as well as free amino acids. Lack of literature on the subject pertaining to the issue of physiological effectiveness makes it impossible to further discuss this finding.

Table 6. Physiological efficiency of fertilization in *Dactylis glomerata* and *Festulolium braunii* with nitrogen and the biostimulant Kelpak (kg d.m. ·kg^{-1} N uptake by plants)

Year	Dose of N (kg·ha^{-1})	*Dactylis glomerata*		*Festulolium braunii*		Mean	
		Treatment		Treatment		Treatment	
		I	II	I	II	I	II
2010	50	A 34.4 a	A 32.2 a	A 23.8 a	A 30.5 b	A 29.1 a	A 31.4 a
	100	A 35.5 a	A 30.6 b	B 27.8 a	A 31.9 b	A 31.6 a	A 31.3 a
	150	A 32.5 a	A 29.7 a	AB 25.0 a	B 23.2 a	A 28.8 a	A 26.4 a
2011	50	A 42.5 a	A 34.9 b	A 37.2 a	A 29.3 b	A 39.9 a	A 32.1 b
	100	B 38.5 a	AB32.4 b	A 37.4 a	B 34.2 b	A 37.9 a	A 33.3 b
	150	C 33.7 a	B 30.9 a	A 34.7 a	B 32.4 a	B 34.2 a	A 31.6 b
2012	50	A 30.3 a	A 24.6 b	A 34.5 a	A 26.3 b	A 32.4 a	A 25.4 b
	100	A 29.3 a	A 24.7 b	B 29.0 a	A 28.1 a	B 29.2 a	A 26.4 b
	150	A 28.0 a	A 25.0 b	B 29.6 a	A 27.1 a	B 28.8 a	A 26.0 b
Mean	50	A 35.7 a	A 30.6 b	A 31.8 a	A 28.7 b	A 33.8 a	A 29.6 b
	100	A 34.4 a	A 29.2 b	A 31.4 a	AB 31.4 a	AB 32.9 a	A 30.3 b
	150	B 31.4 a	A 28.5 a	A 29.8 a	AC 27.5 a	B 30.6 a	A 28.0 b
Mean		33.9 a	29.4 b	31.0 a	29.2 a	32.4 a	29.3 b

Explanation see Table 2.

When an interaction of all the factors was taken into account, no effect of Kelpak was found. In the first and second study year, the biostimulant did not affect the physiological effectiveness of *Dactylis glomerata* fertilized with the N rate of 50 and 150 $kg^{.}ha^{-1}$ in 2010 and the N rate of 150 $kg.ha^{-1}$ in 2011. The response of *Festulolium braunii* to Kelpak was similar. In all the study years, an application of the biostimulant significantly reduced *Festulolium braunii* physiological effectiveness in plots fertilized with 150 kg $N^{.}$ ha^{-1}.

CONCLUSION

Increasing nitrogen rates significantly enhanced yields of grasses, nitrogen content and uptake by plants, but decreased agricultural effectiveness and physiological effectiveness. An application of the biostimulant Kelpak favourably affected the experimental characteristics. Physiological effectiveness was the only trait to decline following Kelpak application. The grasses tested in the experiment differed as to the parameters analysed in the study. The performance of *Dactylis glomerata* was better as far as yield, nitrogen uptake, and indicator of agricultural effectiveness were concerned. A significant interaction of all the factors studied in the experiment was confirmed.

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