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ORIGINAL PAPER

EFFECT OF THE FERTILIZATION OF MEADOW SWARD WITH AMINO ACIDS OBTAINED FROM ENZYMATIC HYDROLYSIS ON SILAGE QUALITY*

Adam Radkowski[1], Iwona Radkowska[2], Jan Bocianowski[3]

[1] Department of Agroecology and Crop Production
University of Agriculture in Kraków, Kraków, Poland
[2] Department of Cattle Breeding
National Research Institute of Animal Production, Balice, Poland
[3] Department of Mathematical and Statistical Methods
Poznań University of Life Sciences, Poznań, Poland

Abstract

The quality of meadow sward grown for silage suffers because of the increasingly frequent droughts occurring in recent years in Poland. To alleviate the drought stress, a number of preparations known as plant growth stimulants or biostimulants are increasingly used for cultivation alongside basic mineral fertilization. The aim of the study was to determine the effect of fertilizing meadow sward with an amino acid formula on silage quality. The experiment, conducted in 2016-2018, was set up on a privately owned farm situated in the Kraków District, Małopolskie Voivodeship, Poland. A field trial was established on a permanent meadow in a randomized block design with four replications. The experimental plots were 10 m^2 in area. The soil under the experimental meadow was degraded Chernozem formed from loess, categorized as quality class I. The treatment consisted of spraying with an amino acid preparation, which was applied in two doses: 1 and 2 dm^3 ha^{-1}. The prepared plant material was analysed for basic chemical composition using the standard method. Comparison of the mean values showed that silages from the plot fertilized with an amino acid formula had a favourable content of crude protein, crude fat, water soluble carbohydrates and lactic acid, the highest concentration of which occurred when 2 dm^3 ha^{-1} of the amino acid formula was applied. Out of all the silages, the highest score was awarded to the silage from meadow sward in the plot fertilized with 2 dm^3 ha^{-1} of the amino acid formula. Good results were also obtained for plants originating from the plot fertilized with 1 dm^3 ha^{-1} of the amino acid formula. In summary, it is concluded that the supplemental amino acid formula had a favourable effect on the development of aerial biomass. The silages made from it were characterized by high quality owing to low butyric acid

Adam Radkowski, Assistant Professor, Department of Agroecology and Crop Production, University of Agriculture in Kraków, Al. Mickiewicza 21, 31-120 Kraków, Poland, e-mail: adam.radkowski@urk.edu.pl

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and ammonia concentrations. The higher the concentration of the investigated stimulator used for spraying, the higher its effectiveness.

Keywords: left-handed amino acids, enzymatic hydrolysis, meadow sward, organic nutrient content, organic acids.

INTRODUCTION

Amino acids are organic chemicals that are the building blocks of proteins (Liu et al. 2008); in addition, amino acids are involved in many cellular processes, thus influencing a number of physiological processes, such as plant growth and development, intracellular pH regulation, generation of metabolic energy, enhancement of plant resistance to biotic and abiotic stresses (Fagard et al. 2014, Galili et al. 2014, Häusler et al. 2014, Pratelli, Pilot 2014). Amino acids stimulate growth and are natural nutrient carriers, easily recognized by plants (Liu, Bush 2006). Research into plant metabolism has shown that amino acid catabolism may be important not only during normal ageing, but also for stress tolerance of plants (Zagorchev et al. 2013).

In crop production, amino acids may represent an alternative source of nitrogen, but it has been debated whether amino acids applied to soil are taken up by plants as NO^{3-}, NH^{4+} or in the organic form as simple amino acids (Näsholm et al. 2000, Owen, Jones 2001).

There is now growing awareness that amino acids can be taken up by crop plants (Chapin et al. 1993, Warren, Adams 2006), and studies using 15N have shown that amino acids are an easily accessible form of nitrogen for higher plants (Öhlund, Näsholm 2004, Warren 2006).

Applicability of amino acids in modern farming has been implicated by many researchers (Kamar, Omar 1987, Abdel-Mawgoud et al. 2011). Studies carried out mostly on crops have demonstrated that foliar application of amino acids can increase crop growth and yield, with beneficial effects on plants' chemical composition. Positive effects of amino acid fertilization on crop yield and quality have been reported for cucumber (Kamar, Omar 1987), garlic (El-Shabasi et al. 2005), potato (Awad et al. 2007) and green bean (Abdel-Mawgoud et al. 2011).

According to Sarojnee et al. (2009), amino acids can enhance crop yields by increasing fertilizer assimilation, nutrient and water uptake, photosynthetic rate and dry matter partitioning. Amino acids are also used to alleviate the negative impacts of certain environmental stresses, e.g. salinity (Neeraja et al. 2005, Tantawy et al. 2009). It has been shown that amino acids, such as glutamate, cysteine, phenylalanine and glycine, may weaken oxidative stress in plants directly or indirectly (Ashraf, Foolad 2007, Gill, Tuteja 2010), which is why their application to seeds or leaves might be an option to reduce consequences of oxidative stress.

The nutritive value of meadow sward silage and its suitability as a ruminant feed largely depend on the quality of material intended for ensiling (Purwin et al. 2006). Silage intake and utilization by animals is closely correlated to its quality and thus determines production efficiency (Kirkland et al. 2005).

Forage quality is one of key factors determining profitability of dairy farms, which is why monitoring the quality of bulk forage, particularly silages, is an important aspect of feeding programmes. Silages should be rich in nutrients. Grass silage is treated as protein-rich forage. Meadow plants harvested at early developmental stages contain readily digestible organic matter and high amounts of water-soluble carbohydrates. High levels of these parameters ensure high energy value. An important parameter of silage quality is the content of raw ash. Its high share indicates silage contamination with soil. An increased amount of raw ash always reduces the total protein content and energy value of the silage. Other factors determining the silage quality include the content of crude fibre and shares of cell wall fractions (neutral detergent fibre – NDF, acid detergent fibre – ADF, acid detergent lignin ADL), which can be determined with detergent methods (Brzóska, Śliwiński 2011).

Over the last years, Poland has experienced falling amounts of precipitation during the plant growing season, which is detrimental to the quality and chemical composition of plants, including meadow sward. As studies have shown, the application of amino acids has positive effects on alleviating adverse environmental conditions (Radkowski, Radkowska 2018). Therefore, the objective of this experiment was to determine effects of the fertilization with an amino acid formula on quality parameters of silage made from meadow sward.

MATERIAL AND METHODS

Study site and soil analysis

The experiment was performed on a family-run farm located in the Kraków District, Małopolskie Voivodeship, Poland. A field trial was set up on permanent meadow in a randomized block design with four replications. The size of an experimental plot was 10 m^2. The soil under the experimental meadow was degraded Chernozem formed from loess, categorized as quality class I. The content of available forms of phosphorus (12.6 mg 100 g^{-1} soil), potassium (16.2 mg 100 g^{-1} soil) and magnesium (8.1 mg 100 g^{-1} soil) in the soil was average.

Before the trial, the dominant grasses in natural sward were: perennial ryegrass (*Lolium perenne*), meadow fescue (*Festuca pratensis*) and orchard grass (*Dactylis glomerata*) – Table 1. Leguminous plants were represented

Table 1

Floristic composition of meadow sward prior to the trial

Grasses	**80%**
Perennial ryegrass (*Lolium perenne* L.)	22%
Meadow fescue (*Festuca pratensis Huds.*)	16%
Orchard grass (*Dactylis glomerata* L.)	14%
Smooth-stalked meadow-grass (*Poa pratensis* L.)	12%
Red fescue (*Festuca rubra* L.)	7%
Timothy (*Phleum pratense* L.)	6%
Rough stalked meadow-grass (*Poa trivialis* L.)	3%
Leguminous plants	**12%**
Red clover (*Trifolium pratense* L.)	8%
White clover (*Trifolium repens* L.)	4%
Dicotyledonous plants	**8%**
Ribwort plantain (*Plantago lanceolata* L.)	2%
Great plantain (*Plantago maior* L.)	1%
Chickweed (*Stellaria media* (L.) Vill.)	+
Red dead-nettle (*Lamium purpureum* L.)	1%
Yarrow (*Achillea millefolium* L.)	1%
Dandelion (*Taraxacum officinale* F. H. Wigg.)	1%
Hedge bedstraw (*Galium mollugo* L.)	+
Shepherd's-purse (*Capsella bursa pastoris*)	1%

by red clover (*Trifolium pratense*) and white clover (*Trifolium repens* L.) at 12%. The sward herbage also contained other dicotyledons, 8 species of which formed 8% of the sward composition.

Weather conditions

Total annual precipitation during the study period (2016-2018) was 626.7, 553.5 and 441.6 mm, respectively (Table 2). The average total precipitation during the plant growing period (April to September) was 321.6 mm in 2016, 412.0 mm in 2017 and 335.6 mm in 2018. The average annual temperature in the years 2016–2018 was 6.9, 6.2 and 7.0°C, including 12.1, 12.4 and 14.5°C in the April to September period, respectively.

Materials and experimental designs

The experiment used three plots: a control plot (no formula applied) and experimental plots sprayed with the amino acid preparation at two doses (1 and 2 dm^3 ha^{-1}) for each regrowth. The first foliar spray was applied five days after the plant growth commenced (for the first cut), and the second

Table 2

Rainfall and average air temperature at the Plant Breeding Station in Polanowice in the years 2016-2018

Month/Year	**2016**	**2017**	**2018**
	monthly rainfall (mm)		
January	23.0	0.0	32.0
February	94.0	0.0	0.0
March	42.5	13.5	35.0
April	67.0	103.3	9.0
May	38.3	72.0	46.6
June	35.8	31.5	67.5
July	80.6	51.4	118.5
August	82.0	2.3	60.0
September	18.0	151.5	34.0
October	99.1	59.5	23.5
November	33.5	41.0	0.0
December	13.0	27.5	15.5
Total	626.7	553.5	441.6
Total April-September	321.6	412.0	335.6
	Average monthly air temperature (°C)		
January	-1.3	-7.2	-1.0
February	3.0	-6.4	-5.0
March	2.0	3.9	-1.6
April	5.3	4.2	9.3
May	10.9	10.1	13.8
June	15.5	15.8	16.4
July	17.0	16.7	18.2
August	14.0	16.8	17.8
September	10.1	10.8	11.5
October	5.5	6.7	5.8
November	1.3	1.9	0.2
December	-0.6	0.9	-1.6
Average	6.9	6.2	7.0
Average April-September	12.1	12.4	14.5

and third doses were sprayed five or six days after harvest (for the second and third cut).

Spray solutions were prepared by dissolving appropriate amounts of the amino acid formula in such an amount of water as to yield a working liquid volume of 300 dm^3 ha^{-1}.

The AGRO-SORB® Folium amino acid formula is a growth stimulant with 18 biologically active free amino acids (L-alpha) obtained by enzymatic hydrolysis. The formula has a high content of biologically active free amino acids (at least 9.3% m m^{-1} – minimum of 100 g 1000 ml^{-1}), namely: aspartic acid 0.450%, serine 0.321%, glutamic acid 1.814%, glycine 2.743%, histidine 0.208%, arginine 0.131%, threonine 0.323%, alanine 0.524%, proline 0.347%, cysteine 0.435%, tyrosine 0.174%, valine 0.551%, methionine 0.349%, lysine 0.661%, isoleucine 0.308%, leucine 0.180%, phenylalanine 0.218%, tryptophan 0.05% (data confirmed by a chemical analysis). The amino acids join together in the order specified by the DNA code. As a result, each plant is able to produce the proteins it needs and to transport them to deficient sites so as to enhance the metabolic process. The experimental fertilizer is manufactured by Biopharmacotech Sp. z o.o. Sp. K. from Częstochowa, Poland. Throughout the study, basic mineral fertilization was also used: 80 kg N ha^{-1} for the first cut, and 60 kg N ha^{-1} in the form of 34% N ammonium nitrate each for the second and third cut. Phosphorus was applied once in spring (35.2 kg P ha^{-1}) in the form of superphosphate enriched with 17.6% P, and the first and third cuts were fertilized with potassium (49.8 kg K ha^{-1} each as 49.8% K potassium salt).

The green crop was cut at the heading stage of dominant grass species for the first cut, and after seven weeks of growth up to 6-7 cm for the second and third cut. Then, the harvested green matter was dried for one day. At the next stage, the wilted material was cut with a straw cutter into 2 cm chaff. Chaffed and wilted green crop was compacted in 5 L plastic containers (three repetitions) and tightly sealed. Compaction of the silaged plant material reached 200 kg of dry matter per 1 m^3.

The containers with silage were stored indoors at about 15°C for four months. Then, they were opened and samples were collected for chemical analyses. The content of basic components was determined by the Weende method (AOAC, 1995), pH was measured with a pH-meter, and the level of ammonia was evaluated with the Conway method (Conway 1988). The lactic acid content was determined on a Varian 3400 CX gas chromatograph, coupled with an FID (flame ionization) detector, a DB-FFAP column (JW Scientific) 30 cm in length, 0.53 mm in diameter, argon as a carrier gas, dispenser temp. set at 200°C, temp. detector set at 240°C, and the temp. in the column set at 60-210°C. The content of acetic and butyric acids was determined using an LCP 5020 liquid chromatograph (INGOS), composed of a 8 × 250 mm steel column with an OSTION LG-KS 0800 H + packing from Tessek, and the mobile phase was 5 mM H_2SO_4.

Statistical analysis

Normality of the distribution of the studied factors (dry matter, ash, crude protein, ether extract, crude fibre, NDF, ADF, (ADL), water soluble carbohydrates, pH, N-NH_3, lactic acid, acetic acid and butyric acid) was tested using the Shapiro-Wilk's normality test (Shapiro, Wilk 1965). Multivariate analysis of variance (MANOVA) was performed on the basis of the following model using the MANOVA procedure in GenStat ver. 18: Y=XT+E, where: Y is an (n×p)-dimensional matrix of observations, n is the number of all observations, p is the number of traits (in this study p=14), X is ($n \cdot k$)-dimensional matrix of design, k is the number of combinations (in this study k=9, control, A1 and A2 in the three cuts), T is a (k×p)-dimensional matrix of unknown effects, E – is an (n×p)-dimensional matrix of residuals. Next, two-way analyses of variance (ANOVA) were performed in order to verify the zero hypothesis of the lack of effects induced by cuts and doses as well as a cut dose interaction in terms of values of the observed factors, independently for each trait. The arithmetical means and standard deviations of factors were calculated. Moreover, Fisher's least significant differences (LSDs) were also estimated at the significance level α=0.05. Variation of the observed traits was presented in the form of a boxplot. The relationships between observed between the traits were assessed on the basis of the Pearson's correlation. The results were also analysed using multivariate methods. The canonical variate analysis was applied in order to make a multitrait assessment of the similarity of the tested combinations of cuts and doses in a lower number of dimensions with the least possible loss of information (Rencher 1992). This makes it possible to illustrate variation in the combinations of cuts and doses in terms of all the observed traits in a graphic form. The Mahalanobis distance was suggested as a measure of "polytrait" of the similarity between the cuts doses combinations (Seidler-Łożykowska, Bocianowski 2012), whose significance was verified by means of critical value D_α called "the least significant distance" (Camussi et al. 1985). Mahalanobis distances were calculated for combinations of cuts and doses (Mahalanobis 1936). All the analyses were conducted using the GenStat ver. 18 statistical software package.

RESULTS AND DISCUSSION

The results of the MANOVA indicated that the cuts (Wilk's λ=0.00007, $F_{28,10}$=42.27, P<0.0001) and doses (Wilk's λ=0.001285, $F_{28,10}$=9.61, P<0.0001) were significantly different with regard to all of the 14 quantitative traits. The multivariate cuts doses interaction was not statistically significant (Wilk's λ=0.014, $F_{56,22}$=0.77, P=0.784). The ANOVA results indicated significant effects of a dose on crude protein, ether extract, crude fibre, water soluble carbohydrates, NH_3-N, acetic acid and butyric acid; however, effects of cuts on ash, crude protein, ether extract, crude fibre, NDF, ADF, ADL and water soluble carbohydrates were not statistically significant (Table 3). The cut dose interaction was statistically significant only for water soluble carbohydrates (Table 3).

Table 3

Mean squares from two-way analysis of variance for observed traits

Source of variation	Dose	Cut	Dose Cut	Residual
Degrees of freedom	2	2	4	18
Dry matter	497	87.5	34.5	167.6
Ash	10.7	537.9*	132.5	143.5
Crude protein	268.1***	3868***	3.88	22.02
Ether extract	154.9***	228.3***	25.61	10.92
Crude fibre	557.4*	5763***	34.5	107.8
NDF	3553	10447*	449	1919
ADF	4709	6071*	642	1414
ADL	201.7	390.0*	53.11	99.98
Water soluble carbohydrates	3866***	4635***	303.2*	89.18
pH	0.918	0.495	0.028	0.289
NH_3-N	1.201**	0.061	0.058	0.155
Lactic acid	671.6	2242	125.7	664.3
Acetic acid	326.6***	9.63	4.91	20.37
Butyric acid	15.45***	3.26	0.325	2.365
* P<0.05, ** P<0.01, *** P<0.001				

The results of determination of the basic chemical composition of the silages are presented in Figure 1. Silages made from meadow sward from the experimental plots compared to those from the control plot showed no significant differences (p>0.05) in the dry matter content with regard to a fertilizer treatment (Table 3, Figure 1). The crude protein content of the silages varied from 93.1 to 144.1 g kg^{-1} of dry matter, depending on a fertilizer treatment and harvested cut (Figure 1). Foliar application of the amino acids at a higher dose (2 dm^3 ha^{-1}) had a beneficial effect (p≤0.05) on the crude protein content at each harvest date (Figure 1). In turn, silages from the control plot had the lowest concentration of crude protein (Figure 1). The CP increase compared to the control plot at different cuts averaged 9.7% for the plot with the higher dose of the amino acid formula. Evaluation of the crude fibre content in absolute dry matter showed a statistically significant (p≤0.05) increase in the content of this component compared to the control plot (by 38.6% on average) in favour of the plot with the higher amino acid dose (Figure 1). The crude fibre content in the analysed silages varied from 16.0 to 34.2 g kg^{-1} of dry matter depending on a fertilizer variant and harvested cut, and the average values for silages from experimental plots were 24.0 g kg^{-1} of dry matter in response to the lower dose, and 30.7 g kg^{-1} of dry matter when the higher dose of the amino acid formula had been applied.

In the silages made from meadow sward fertilized with the higher amino acid formula dose, there a significant decrease in the crude fibre content only in the third cut, where it decreased by an average of 7% compared to the

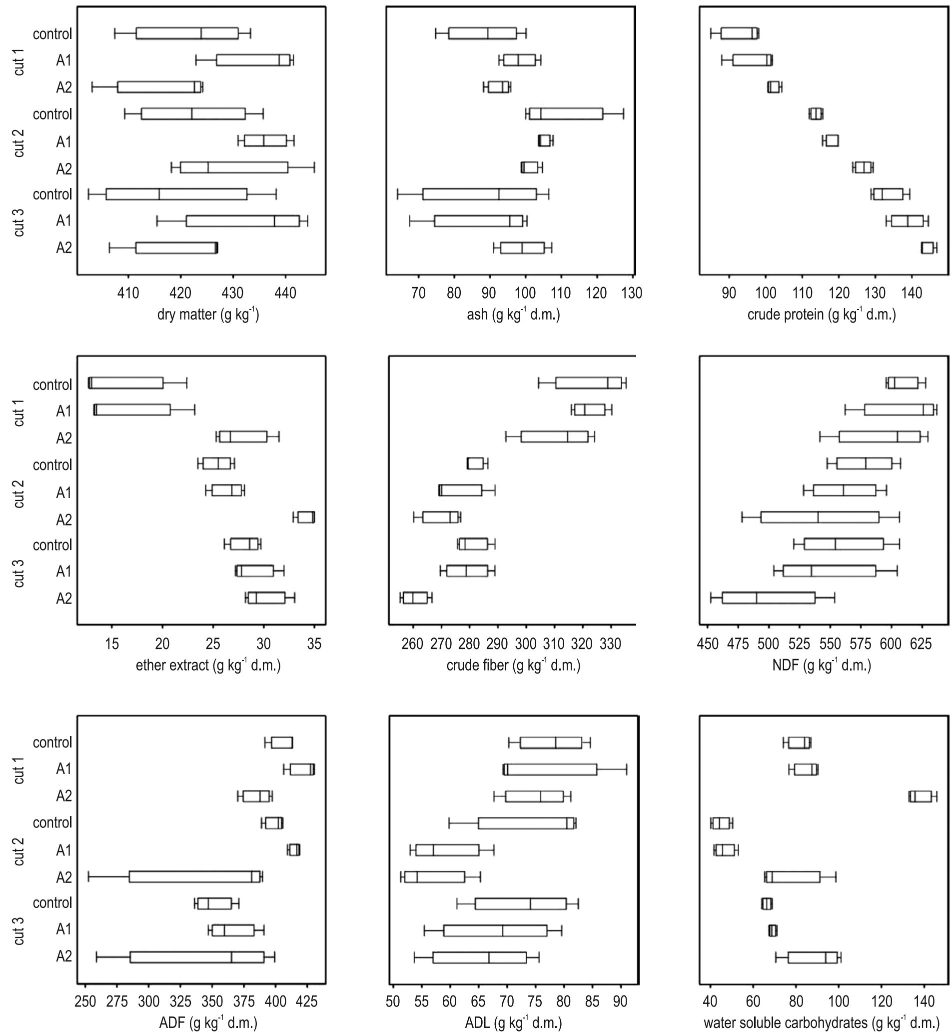

Fig. 1. Boxplots of the values of dry matter, ash, crude protein, ether extract, crude fibre, NDF, ADF, ADL and water soluble carbohydrates for three cuts and three doses

content from the control plot (Figure 1). No significant differences were found between the treatments with regard to fibre fractions ADF, ADL and NDF, although the NDF and ADL content decreased with the decreasing dose of the amino acid formula (Figure 1).

A higher concentration of water-soluble carbohydrates was obtained for silages from the sward fertilized with the higher amino acid formula dose (Figure 1). No significant differences were observed between the fertilizer treatments in the pH of the silages obtained. This value was in the 5.15-6.14 range and decreased with an increasing dose of the amino acid formula (Figure 2).

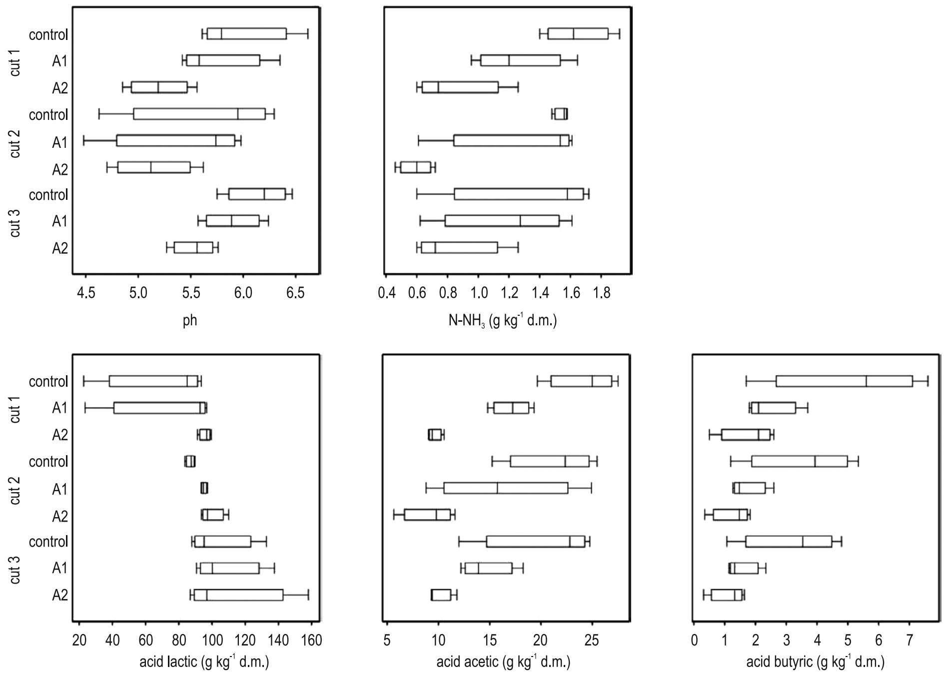

Fig. 2. Boxplots of the values of pH, $N\text{-}NH_3$, acid lactic, acid acetic and acid butyric for three cuts and three doses

The content of ammonia, which is an indicator of protein degradation during ensiling, showed certain differences (Figure 2). The amino acid formula caused a significant ($p \leq 0.05$) reduction in the amino acid content of all the silages.

The lactic acid content in different silages was quite varied and depended on amino acid fertilization and time of meadow sward harvest (Figure 2). The lactic acid content increased under the higher amino acid formula dose. An analysis of the acetic acid content showed that it was the lowest in the silage made from meadow sward fertilized with 2 dm^3 ha^{-1} of the amino acid formula. The highest amount of acetic acid and butyric acid occurred in the silage from the control plot (Figure 2). The silages made from meadow sward fertilized with 1 dm^3 ha^{-1} of the amino acid formula had an intermediate content of these acids compared with the other silages (Figure 2).

The correlation analysis indicated statistically significant correlation coefficients for 26 out of 91 coefficients (Table 4). 14 out of 26 significantly correlated pairs of traits were characterized by positive correlation coefficients.

Individual traits differ in importance and contribute different shares to the joint multivariate variation. A study on multivariate variation also includes identification of the most important traits in the multivariate variation of cuts x doses combinations. An analysis of canonical variables

Table 4

Correlation coefficients between observed traits

Trait	Dry matter	Ash	Crude protein	Ether extract	Crude fibre	NDF	ADF	ADL	Water soluble carbo-hydrates	pH	NH_3-N	Lactic acid	Acetic acid	Butyric acid
Dry matter	1													
Ash	0.01	1												
Crude protein	0.04	0.03	1											
Ether extract	0	0.11	0.63***	1										
Crude fibre	0.06	-0.2	-0.82***	-0.67***	1									
NDF	0.07	-0.2	-0.62***	-0.57**	0.62***	1								
ADF	0.22	0.06	-0.56**	-0.48*	0.55**	0.65***	1							
ADL	0.24	-0.3	-0.35	-0.51**	0.57**	0.41*	0.22	1						
Water soluble carbohydrates	-0.2	-0.2	-0.25	-0.01	0.39*	0.1	0.01	0.17	1					
pH	0.21	-0.2	0.09	-0.33	0.21	0.22	0.11	0.36	-0.11	1				
NH_3-N	-0.2	-0.2	-0.33	-0.48*	0.32	0.21	0.40*	0.21	-0.28	0.17	1			
Lactic acid	0.23	0	0.50**	0.62***	-0.32	-0.33	-0.2	-0.1	-0.02	0.09	-0.32	1		
Acetic acid	0.1	-0.1	-0.28	-0.51**	0.35	0.33	0.33	0.3	-0.45*	0.47*	0.60**	-0.3	1	
Butyric acid	-0.3	-0.2	-0.51**	-0.38*	0.39*	0.36	0.21	0.25	-0.21	0.04	0.46*	-0.1	0.3	1

* $P<0.05$; ** $P<0.01$; *** $P<0.001$

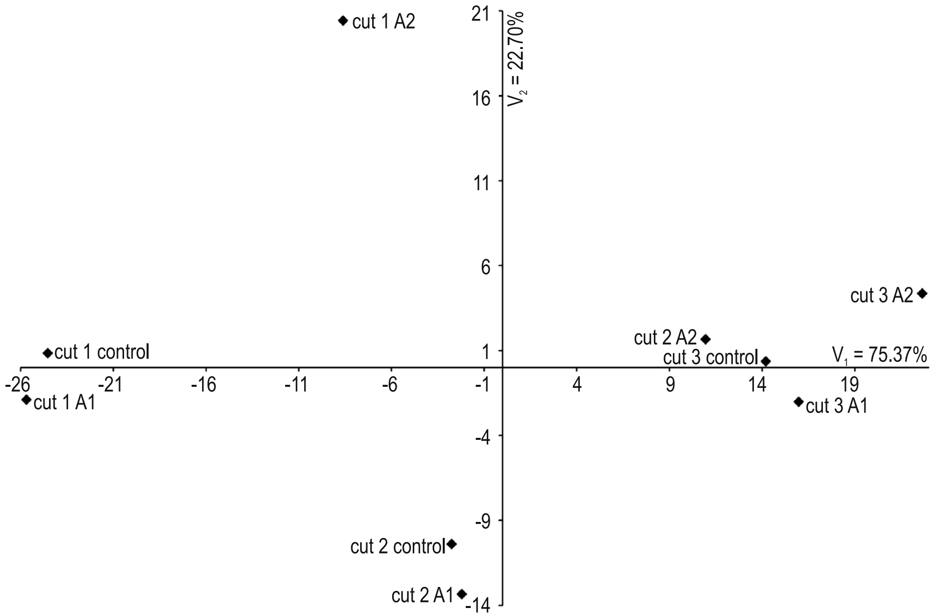

Fig. 3. Distribution of nine cuts-by-doses combinations in the space of the first two canonical variables

is a statistical tool that makes it possible to solve the problem of multivariate relationships (NIEMANN et al. 2018, PIESIK et al. 2018, WROŃSKA--PILAREK et al. 2018, RYBIŃSKI et al. 2019). Figure 3 shows the variability of the nine cuts x doses combinations in terms of the first two canonical variables. In the graph, the coordinates of the point for particular combinations are the values for the first and second canonical variables, respectively. The first two canonical variables accounted for 98.07% of the total multivariate variability between the individual combinations. The most significant positive, linear relationship between the first canonical variables was found for crude protein, ether extract and lactic acid, while being negative for crude fibre, NDF and ADF (Table 5). The second canonical variable was significantly positively correlated with water soluble carbohydrates (Table 5). The greatest variation in terms of all the 14 traits together (measured by the Mahalanobis distances) was found for A1 in cut 1 and A2 in cut 3 (the Mahalanobis distance between them equalled 48.8). The greatest similarity was found for the control in cut 3 and A1 in cut 3 (4.67) – Table 6.

The yield and productivity of crops can be increased by improving and regulating their growth conditions and influencing the rate of photosynthesis (DRECCER et al. 2000, SHARMA-NATU, GHILDIYAL 2005). One possible solution is the application of growth stimulants in the form of amino acids. Foliar fertilization at later stages of plant development stimulates regeneration of the root system and activates photosynthesis, thus increasing the productivity of plants. The application of amino acids has an advantage of limiting

Table 5

Correlation coefficients between the first two canonical variables and original traits

Trait	First canonical variable	Second canonical variable
Dry matter	-0.128	-0.61
Ash	-0.016	-0.508
Crude protein	0.979***	-0.123
Ether extract	0.872**	0.162
Crude fibre	-0.913***	0.291
NDF	-0.923***	0.045
ADF	-0.861**	-0.343
ADL	-0.599	0.27
Water soluble carbohydrates	-0.181	0.966***
pH	-0.101	-0.27
NH_3-N	-0.514	-0.507
Lactic acid	0.961***	0.15
Acetic acid	-0.48	-0.527
Butyric acid	-0.604	-0.211
Percentage of explained multivariate variability	75.37%	22.70%
** P<0.01, *** P<0.001		

Table 6

Mahalanobis distances between cuts-by-doses combinations on the basis of all 14 observed traits

Combination	Cut 1 control	Cut 1 A1	Cut 1 A2	Cut 2 control	Cut 2 A1	Cut 2 A2	Cut 3 control	Cut 3 A1	Cut 3 A2
Cut 1 control	0								
Cut 1 A1	5.44	0							
Cut 1 A2	25.64	28.23	0						
Cut 2 control	24.77	24.93	31.53	0					
Cut 2 A1	27.06	26.22	34.44	5.84	0				
Cut 2 A2	36.04	36.93	27.26	19.07	20.05	0			
Cut 3 control	38.76	40.26	30.73	20.33	22.24	7.51	0		
Cut 3 A1	40.74	41.7	33.42	20.97	21.7	7.43	4.67	0	
Cut 3 A2	47.41	48.8	35.24	29.57	30.7	12.87	10.08	9.58	0
$D_{0.05}$=25.73									

mineral fertilization and improving its efficiency. Research has shown that amino acids may positively affect development of the root system and activate the growth of aerial parts of plants (NIKIFOROVA et al. 2006). These authors suggest that nutrient uptake by roots depends on the amount of aspartic and glutamic acids in plants. Research has shown that amino acid fertilization stimulates respiration and photosynthesis as well as water flow through plants. Furthermore, amino acids accelerate protein synthesis and increase the content of ascorbic acid. All of these modifications have a beneficial effect on plant growth and yield (Alaru et al. 2003, Meijer 2003).

Preparations that contain amino acids as stimulants are mainly used in growing fruits and vegetables. Research shows that amino acids also have positive effects on the growth of meadow sward plants (Ahmed et al. 2011, Sadak et al. 2015, Kandil et al. 2016, Radkowski et al. 2018). The high fertilizer efficiency of amino acids stems from the fact that they are a source of easily available nitrogen for plant cells, which is more readily absorbed than inorganic nitrogen. In addition, amino acids play a key role in plant metabolism, including the absorption of proteins, which are among the most important compounds conducive to the formation of cells, which increases amounts of fresh and dry matter. Higher crop yields after an application of amino acids have been reported by many authors (Saeed et al. 2005, El-Zohiri, Asfour 2009, Ahmed et al. 2011, Tarraf et al. 2015). Studies have shown that fertilization with amino acids causes a significant increase in soybean yields (Saeed et al. 2005), considerably higher plant height, stem diameter, and fresh and dry matter of roselle leaves (Ahmed et al. 2011), and increased plant height and dry matter yield in potato (El-Zohiri, Asfour 2009). Similar results have been noticed in cultivation of herbs, where an application of amino acids increased the number of leaves, number of branches, plant height, and fresh and dry weights of fenugreek (*Trigonella foenum-graecum* L.) (Tarraf et al. 2015) and chamomile (Gamal El-Din, Abd El-Wahed 2005). This may be due to the fact that amino acids play a major role in metabolism of plants and assimilation of proteins, which are needed for producing new cells, thus increasing fresh and dry matter yields (Kandil et al. 2016).

Our study demonstrated that the amino acid fertilization of meadow sward had an effect on the crude protein and crude fat content of silage. For the third cut, a significant decrease in the crude fibre content and a higher concentration of water soluble carbohydrates were observed. Higher protein content of the plants fertilized with amino acids was also reported by Zewail (2014) and Pooryousef, Alizadeh (2014). The use of Aminol-forte caused crude protein to increase by 22% compared to 17% in control. Goss (1973) indicated that amino acids can serve as a source of carbon and energy. When carbohydrates become deficient in the plant, amino acids are deaminated, releasing the ammonia and the organic acid from which the amino acid was originally formed.

Analysis of the silage for chemical composition showed that it is a good feed value. The minimum crude protein content in feed to ensure normal digestion in the gastrointestinal tract of cattle should be 150-170 g kg d.m. (Brzóska 2008). The crude protein content in the analysed silages varied and was slightly below the optimum, especially in the plot without amino acid fertilization. Grassland feeds used in ruminant nutrition should contain around 200-250 g kg^{-1} d.m. of crude fibre and should not exceed 280 g kg^{-1} d.m. (Brzóska 2008). In our study, we found no significant differences between different treatments, but NDF and ADL concentrations tended to decrease under the influence of amino acid fertilization. The use of the amino acid stimulant had a beneficial effect on the crude fat content of the silages, and in the experimental treatments these values fell within the optimum range of 20-50 g kg^{-1} d.m. for roughages (Brzóska, Śliwiński 2011).

An important determinant of the quality of silages and their suitability in diets for animals is the content of organic acids (Podkówka, Potkański 1993). Good silage should have a pH of around 4.2. The lower the pH, the higher the acidity and thus the higher the content of lactic acid and acetic acid in the silage. A pH value of 5.0 and above is indicative of the content of butyric acid (Nowak, Šařec 2001). The presence of butyric acid reduces the quality of silage, and amounts over 10 g kg^{-1} d.m. disqualify the use of silage as a feed. In our study, silage pH was slightly above the optimum level. The use of the amino acid preparation contributed to a lower pH value of silage, which decreased with an increasing dose of the amino acid formula.

The yield and quality of the ensiling material from permanent meadows are significantly influenced by weather conditions during plant growth. Because recent years have witnessed an increasing incidence of droughts in our climate, which considerably lower the yield of crops, means and methods to offset the negative effects of drought stress in plants are being searched for. Research has shown that amino acids are particularly effective when used under stress conditions for plants. The mitigating action of amino acids may result from an increased content of photosynthetic pigments and from the number of leaves in plants following an application of the amino acid stimulant. The findings of Sadak et al. (2015) showed that the application of amino acids increased the content of carbohydrates and polysaccharides in stressed plants. These results are in agreement with other results obtained for different plant species (Abdel Aziz et al. 2010). There is a positive correlation between photosynthesis rates and nitrogen content in leaves. A high rate of photosynthesis owing to a high nitrogen supply results in higher biomass production (Neuberg et al. 2010).

CONCLUSIONS

Silages made from plants fertilized with the amino acid preparation contained more total protein, crude fat, water-soluble carbohydrates and lactic acid. The highest levels of these components were found in the silages where the amino acid preparation was applied at 2 dm^3 ha^{-1}.

Meadow plants fertilized with the amino acid preparation make good raw material for silages. The resulting silages are of high quality owing to a low content of butyric acid and ammonia. The higher the concentration of the investigated amino acid preparation used for spraying, the higher its effectiveness. The amino acid preparation positively affected fermentation and the quality of silages, and may be recommended for fertilizing meadow sward intended for silages.

Conflict of interest

The authors declare no conflict of interest.

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