

Nutritional value and fermentation characteristics of silage made from hybrid *Rumex patientia* L. × *Rumex tianschanicus* A.Los (Rumex OK 2) in different months during the year

Michal Rolinec^{1*}, Dzhamal Rakhmetov², Daniel Bíro¹, Miroslav Juráček¹,
Milan Šimko¹, Branislav Gálik¹, Ondrej Hanušovský¹

¹Slovak University of Agriculture in Nitra, Faculty of Agrobiological and Food Resources,
Department of Animal Nutrition, Slovakia

²M. M. Gryshko National Botanical Garden of Ukraine National Academy of Sciences in Kyiv, Ukraine

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The aim of this study was to determine the nutritive value and fermentation parameters of silage made from hybrid *Rumex patientia* L. × *Rumex tianschanicus* A.Los (Rumex OK 2). Silages were made in months September, October and November of the year 2017 and next in April and May of the year 2018. In each month two variants were analysed, one without additives and second with an addition of 1% of dried molasses to wilted Rumex OK 2 plants. After 5 weeks fermentation in hermetic sealed plastic bags at temperature 20 °C the concentration of nutrients and parameters of fermentation were analysed in average samples. The content of dry matter in all silages were low and ranged from 7.1 to 18.8%. Content of crude protein was highest in autumn months, when was from 289 to 339 g.kg⁻¹ DM, which is much more compared to alfalfa silages. Crude fiber was in spring months from 295 to 422 g.kg⁻¹ DM and in autumn months from 126 to 166 g.kg⁻¹ DM. Development of fiber components was similar to development of crude fiber content. The concentration of crude protein and neutral detergent fiber in Rumex OK 2 silages from autumn months meet the criteria for first class legume silage. Only silages from October and November had the content of lactic acid more than 10 g.kg⁻¹ of original matter. Addition of dried molasses increased ($P < 0.05$) concentration of lactic acid and decreased ($P > 0.05$) concentration of acetic acid in silages from September, October, April and May. All Rumex OK 2 silages did not contain butyric acid. Silage pH value appertain to its dry matter concentration was relative high, which make impossible the good overall assessment. However, according to concentration of crude protein, neutral detergent fiber and proteolysis can be Rumex OK 2 silages from autumn months considered as a nutritional valuable feed. On the other hand Rumex OK 2 silage from September contains high concentration of oxalic acid, which can be potentially hazardous for animals.

Keywords: Rumex OK 2, silage, dried molasses, nutrients, fiber complex, fermentation process

1 Introduction

Production of conserved feeds is very important both for yearlong producing of feed rations and for creating of sufficient amount of substrate reserves for biogas stations for whole year (Juráček et al., 2010; Bíro et al., 2014). Advantages of feed conservation are mainly in option of crop harvesting in time of ideal nutritional value and in possibilities of long-term storage of nutrients and energy. Some members of *Rumex* L. family were and in nowadays still are consider as aggressive plants which force cultural grasses and legumes out from their surroundings and cause problems at herbage conservation due to their low dry matter concentration

(Klimeš, 1993). At M. M. Gryshko National Botanical Garden of Ukraine (Kyiv) was from the 1980s hybridized new crop with name Rumex K-1, which contains relative high amount of crude protein, but had not stabile production (Rakhmetov, 2018). Hybridization continues and in year 2001 was in Ukraine registered hybrid Rumex OK 2 (*Rumex patientia* L. × *Rumex tianschanicus* A.Los), which contains high amount of crude protein similar as Rumex K-1 and its production was stabilized. Rumex OK 2 can be used as a feed crop, vegetables, health plant and also as a technical plant (Usták, 2007). Bazhay-Zhezherun and Rakhmetov (2014) published that hybrid Rumex OK 2 can be used also as a food or as a medical plant. The

*Corresponding Author: Michal Rolinec, Slovak University of Agriculture in Nitra, Faculty of Agrobiological and Food Resources, Department of Animal Nutrition, Tr. Andreja Hlinku 2, Nitra, Slovakia, e-mail: michal.rolinec@uniag.sk

relative high concentration of crude protein in *Rumex* OK 2, mainly in young plants, was determined previously in experiment of Hejduk and Doležal (2004). Content of crude protein of *Rumex* OK 2 plant in months March and April was determined on values 261 and 245 g.kg⁻¹ DM respectively and after harvesting in June the crude protein concentration of *Rumex* OK 2 second crop was on values from 302 to 314 g.kg⁻¹ DM. The disadvantage was the very low concentration of dry matter in March and September and the very low production of biomass during these months – only in form of leaves. Very low concentration of dry matter was determined also by Derrick et al. (1993); Bockholt and Kannewurf (2001); Hejduk and Doležal (2004 and 2008). Despite the low concentration of dry matter, the chance of silage production with such high concentration of crude protein need to be verified. For effective utilisation of conserved feed either for animals or in bioenergy industry is necessary to know the concentration of nutrients, eventually other parameters such as the parameters of fermentative process of silages. Therefore the aim of this study was to determine the nutritional value and fermentative process parameters of silages made from hybrid *Rumex* OK 2 produced in different months without or with an addition of dried molasses.

2 Material and methods

This experiment was realized with silage made from hybrid *Rumex patientia* L. × *Rumex tianschanicus* A.Los (*Rumex* OK 2). Sample of *Rumex* OK 2 for silage production were collected in September, October and November of the year 2017 and in April and May of the year 2018. *Rumex* OK 2 from autumn months of the year 2017 which was used for silage samples preparation was grown after first harvesting of dried *Rumex* OK 2 plants in July 2017. Samples collecting was realised always around the 20th day of the month. After collecting, whole plants were wilted for three days in indoor condition, by open windows and without external heating. After wilting, *Rumex* OK 2 plants were cut to the theoretical length of cut 1.5 cm and ensiled (hermetic sealed in plastic bags). Average weight of silage samples was 1,347 ± 151 g. In each month, a two variants were ensiled and analysed. First variant was silage from wilted *Rumex* OK 2 without addition of any additives, this variant was marked as control (C). Second variant was silage of wilted *Rumex* OK 2 with an addition of 1.0% of dried molasses, this variant was marked with abbreviation (M). During fermentation process, which last for five weeks, plastic bags with silage samples were stored in room without light and at 20 °C. For measuring of the content of basic nutrients and fermentative process parameters a three samples of each variant were analysed. After five weeks, silage samples were prepared for basic nutrients and fermentation

process parameters determination. After predrying and homogenisation of samples the concentration of basic nutrients were detected according to Regulation no. 2145/2004-100. Concentration of dry matter (DM) was determined by the gravimetric method, crude protein (CP) by the Kjeldahl's method, crude fat (CFa): extraction by light petroleum, crude fiber (CFi): gravimetrically as the residue remaining after extraction in acid and alkali reagent, acid detergent fiber (ADF): gravimetrically as the residue remaining after extraction in acid detergent solution, neutral detergent fiber (NDF): gravimetrically as the residue remaining after extraction in neutral detergent solution, acid detergent lignin (ADL): gravimetrically as the residue remaining upon ignition after 72% H₂SO₄ treatment, ash: ashing with the use of a muffle furnace by 550 °C and starch by polarimetry. Cellulose (CEL) and hemicellulose (HE) were calculated, CEL = ADF - ADL; HE = NDF - ADF. Content of nitrogen free extract (NFE) and organic matter (OM) were also calculated NFE = dry matter - crude protein - crude fiber - crude fat - ash; OM = dry matter - ash (Juráček et al., 2011). The content of fermentative organic acids (lactic acid, acetic acid, formic acid and butyric acid) and oxalic acid was determined by analyser EA 100 (Villa Labeco, SVK) using the method of ionic electrophoresis. The content of alcohols was determined by Conway microdiffusion method, the content of water extract acidity (AWE) was determined by alkalimetric titration to pH 8.5 and active acidity (pH) was determined by the electrometric method. Proteolysis was calculated as a percent of N-NH₃ out of total N. Results were statistically analyzed in statistic program IBM SPSS v. 20.0. Descriptive statistic and the effect of sampling time on nutrient content was calculated using ANOVA. Differences of nutrient mean values between silage variant in that month were tested by independent *t*-test. *P* < 0.05 was considered as significant.

3 Results and discussion

It is general known that herbage plant of *Rumex* L. family contains at the start of growth very low concentrations of dry matter (Derrick et al., 1993; Bockholt and Kannewurf, 2001; Hejduk and Doležal, 2004 and 2008). Hejduk and Doležal (2008) determined slower decrease of water content also during wilting of *Rumex obtusifolius*. This statement was confirmed with results of this study (Table 1). The *Rumex* OK 2 silage, mainly from young plants in autumn months, contains high concentration of crude protein. The decrease of crude protein, crude fat, starch and ash concentration in *Rumex* OK 2 silage was detected from September to November as well as from April to May. The aging effect of plant causes the increase of crude fiber and its components (except hemicellulose) in *Rumex* OK 2 silage from April to May (Table 1 and 2). Interesting was the decrease of crude fiber and its

Table 1 Nutritional characteristic of silage from Rumex OK 2 made in different months

	Sep 2017		Oct 2017		Nov 2017		April 2018		May 2018		SEM	P
	C	M	C	M	C	M	C	M	C	M		
DM (%)	7.1*	7.4	18.2	17.2	18.0	18.5	12.1*	13.2	18.3	18.8	0.71	***
CP (g.kg ⁻¹ DM)	339*	319	304*	292	305*	289	175*	162	102	110	14.0	***
CFa (g.kg ⁻¹ DM)	32.4*	28.4	22.5*	19.1	17.4	16.9	11.8*	9.4	8.4	8.6	1.28	***
CFi (g.kg ⁻¹ DM)	160	164	158*	166	126*	138	299	295	422	406	17.2	***
NFE (g.kg ⁻¹ DM)	309*	337	369	372	420*	426	462	486	398	415	9.41	***
S (g.kg ⁻¹ DM)	35.0*	25.5	29.8	28.0	14.9	4.7	53.7*	41.7	27.7	21.8	2.85	***
OM (g.kg ⁻¹ DM)	841	848	854	850	868	869	908	903	930*	940	5.57	***
Ash (g.kg ⁻¹ DM)	159	152	146	150	132	131	92.2	96.9	70.1*	59.1	5.57	***

C – control (silage without additives); M – molasses (silage with addition of 1% of dried molasses); DM% – concentration of dry matter in%; CP – crude protein; CFa – crude fat; CFi – crude fiber; NFE – nitrogen free extract; S – starch; OM – organic matter; SEM – standard error of the mean; * – means between C and M silage in that month were significantly different at $P < 0.05$; *** – effect of sampling time on nutrient content was significant at $P < 0.001$

Table 2 Fiber characteristic of silage from Rumex OK 2 made in different months

	Sep 2017		Oct 2017		Nov 2017		April 2018		May 2018		SEM	P
	C	M	C	M	C	M	C	M	C	M		
ADF (g.kg ⁻¹ DM)	174	191	212	209	158	161	350	353	504	506	20.9	***
NDF (g.kg ⁻¹ DM)	222*	247	229*	243	172*	187	416	420	566	565	23.1	***
ADL (g.kg ⁻¹ DM)	22.4	21.5	47.7*	38.4	26.8	27.0	61.4	57.9	127*	136	6.40	***
CEL (g.kg ⁻¹ DM)	151*	169	164	171	131	142	288	295	377	371	14.8	***
HE (g.kg ⁻¹ DM)	48.7	56.6	29.4	33.8	14.3*	26.0	66.0	67.6	61.1	58.7	3.07	***

C – control (silage without additives); M – molasses (silage with addition of 1% of dried molasses); ADF – acid detergent fiber; NDF – neutral detergent fiber; ADL – acid detergent lignin; CEL – cellulose; HE – hemicellulose; DM – dry matter; SEM – standard error of the mean; * – means between C and M silage in that month were significantly different at $P < 0.05$; *** – effect of sampling time on nutrient content was significant at $P < 0.001$

components concentration between months October to November. This decrease can be caused by higher count of leaves and lower count of caulicles in silage from November compared to silage from October. Bockholt and Kannewurf (2001) determined concentration of fiber in *Rumex obtusifolius* leaves on value from 120 to 150 g.kg⁻¹ DM and concentration of fiber in caulicles on value of 130 to 380 g.kg⁻¹ DM. First class legume silage contains at once more than 225 g.kg⁻¹ DM of crude protein and less than 375 g.kg⁻¹ DM of neutral detergent fiber (Mitrík, 2014). According the concentration of crude protein and neutral detergent fiber, all Rumex OK 2 silages from autumn months meet the criteria for first class legume silages.

Because of low dry matter concentration after wilting, dissatisfy results of fermentation process were expected. Due to concentration of crude protein and nitrogen free extract in silages of Rumex OK 2, the fermentation process quality of this silages were evaluated according to criteria for legume silages. Škultéty (2014) published that

legume silage classified in first or in second qualitative class have to have concentration of lactic acid on value at least 10 g.kg⁻¹ original matter or more. This criteria met only Rumex OK 2 silages from October and November, silages from other months had concentration of lactic acid lower than 10 g.kg⁻¹ original matter. Concentration of acetic acid of Rumex OK 2 silages in November, April and May met according to Mitrík (2014) the requirement for first qualitative class of legume silage. Dry matter concentration and its apparent pH value in Rumex OK 2 silages was in September and May inadequate and in other months was high. Weissbach (1998) declared for cell contents of fresh *Rumex obtusifolius* plants the pH value from 4.5 to 4.8. In general the pH value of all Rumex OK 2 silages was compared to required criteria on high value, which has negative effect on classification of this silages. On the other hand, the value of proteolysis was in all Rumex OK 2 silages adequate for first class legume silage (Mitrík, 2014). Also Weissbach (1998) determined for silage of *Rumex obtusifolius* low value of proteolysis. There is rule, that maize or alfalfa silages with low

Table 3 Results of fermentation process of silage from Rumex OK 2 made in different months

	Sep 2017		Oct 2017		Nov 2017		April 2018		May 2018		SEM	P
	C	M	C	M	C	M	C	M	C	M		
LA (g.kg ⁻¹ DM)	32.6*	53.4	99.1*	133	58.2	70.1	28.5*	38.4	6.3*	13.2	6.65	***
AA (g.kg ⁻¹ DM)	49.9	39.8	36.7	32.6	n.d.	15.4	7.0	4.9	8.0	6.1	3.00	***
FA (g.kg ⁻¹ DM)	15.4	15.0	11.8*	12.9	n.d.	n.d.	n.d.	n.d.	2.7	n.d.	0.92	***
OA (g.kg ⁻¹ DM)	137*	121	52.1	54.4	50.5	48.6	48.3	42.9	30.8	30.3	0.58	***
Alco. (g.kg ⁻¹ DM)	32.0*	44.9	13.6	11.5	15.2	10.9	36.9*	16.5	15.1*	24.2	2.09	***
pH	4.9	4.7	5.0	4.7	4.5	4.5	4.3	4.5	4.9	4.9	0.05	***
AWE	500*	573	1009*	1152	878	934	469	431	285	330	47.1	***
DP (%)	4.2	4.0	3.7	3.8	2.1	2.3	10.1	10.6	7.4*	6.3	0.51	***

C – control (silage without additives); M – molasses (silage with addition of 1% of dried molasses); LA – lactic acid; AA – acetic acid; FA – formic acid; OA – oxalic acid; Alco. – alcohols; AWE – water extract acidity (mg KOH pre 100 g of silage); DP – degree of proteolysis in% (N-NH₃ out of total N); n.d. – not detected; * – means between C and M silage in that month were significantly different at $P < 0.05$; SEM – standard error of the mean; *** – effect of sampling time on fermentation process parameter was significant at $P < 0.001$

concentration of dry matter contains butyric acid. Rumex OK 2 silages did not contain any butyric acid. This fact was also determined previously by Hejduk and Doležal (2008).

Rumex L. family is characteristic with content of oxalic acid, which concentration more than 100 g.kg⁻¹ DM can be considered as potentially hazardous as published Kalač and Míka (1997). Higher intake of oxalate by cattle can causes hypocalcaemia, tetanic contraction or death (Tůmová et al., 2010). In this experiment the concentration of oxalic acid in Rumex OK 2 silages in months October, November April, and May was in interval from 30.3 to 54.4 g.kg⁻¹ DM, whereas in September it was in interval from 121 to 137 g.kg⁻¹ DM. Upon the concentration of oxalic acid in Rumex OK 2 silage from September, it is impossible to feed this silage as single feed to animals. In general, detected concentration of oxalic acid in Rumex OK 2 silage samples (except Rumex OK 2 silage from September) is similar to that in *Rumex L.* herbage published by Hejduk and Doležal (2004). According to Hatcher et al. (1997) concentration of anti-nutritional substances, mainly oxalates and nitrates in dock plants was increased by the fungus *Uromyces rumicis* (Schum.) Wint. and by fertilisation. On the other hand, fertilisation is important factor of plant production (Hric et al. 2018; Kovár et al. 2017). In disagreement with statement of Hatcher et al. (1997), that young leaves of dock plant contains the lowest content of oxalates, result of this study was, that concentration of oxalic acid was highest in silage from younger plants (April and September) and decreased with the time (Table 3). Derrick et al. (1993) published, that the content of anti-quality substances in the dock leaves is a natural protection of the plant against the herbivores.

Additionally the significance of difference of mean values between control (C) Rumex OK 2 silage without additives and (M) Rumex OK 2 silage with addition of 1% of dried molasses was calculated. Addition of dried molasses significantly ($P < 0.05$) decreased concentration of crude protein in Rumex OK 2 silage in September, October, November and April (Table 1). Rumex OK 2 silage with addition of molasses in September, October, April and May ($P < 0.05$) and in November ($P > 0.05$) had higher concentration of lactic acid. Except silage of November in all Rumex OK 2 silages was after addition of dried molasses lower concentration of acetic acid ($P > 0.05$) (Table 3). In silage used for feeding of ruminants is increase of lactic acid and low concentration of acetic acid desirable. All significant differences between C and M variants in particular months are marked in tables with asterisk (Table 1, 2 and 3). Significant effect ($P < 0.001$) of sampling time on nutrient concentration was detected for all determined nutrients and fermentation parameters (Table 1; 2 and 3).

Comparison of the average nutrient content from all analysed Rumex OK 2 silage samples without regard to variant to *Rumex obtusifolius* silage published by Hejduk and Doležal (2008) and to average of alfalfa silage published by Juráček et al. (2016) is shown in Table 4. According to concentration of crude protein and crude fiber can be silage from Rumex OK 2 considered as a nutritional valuable feed. By common value feeds exists correlation of crude protein and fiber concentration with NEL. However this statement is not consistent by all plants, mainly by herbs. According to Scehovic (2002), herb rich on crude protein and with low crude fiber concentration could have low degree of digestibility, which is connected with NEL concentration and additional this plant can be worse consumed by animals. The consumption rate of

Table 4 Average nutritional value of silage from Rumex OK 2 compared to *Rumex obtusifolius* L. silage and to alfalfa silage

	Rumex OK 2 silage*	<i>Rumex obtusifolius</i> L. silage**	Alfalfa silage***
DM (%)	14.9	17.3	37.3
CP (g.kg ⁻¹ DM)	240	197	174
CFa (g.kg ⁻¹ DM)	17.5	u.	28.2
CFi (g.kg ⁻¹ DM)	233	214	292
NFE (g.kg ⁻¹ DM)	399	429	393
Ash (g.kg ⁻¹ DM)	119	127	117
ADF (g.kg ⁻¹ DM)	282	u.	364
NDF (g.kg ⁻¹ DM)	327	u.	418
LA (g.kg ⁻¹ DM)	53.3	12.92	82.0
AA (g.kg ⁻¹ DM)	22.3	3.6	29.8
BA (g.kg ⁻¹ DM)	+	+	8.4
Alcohols. (g.kg ⁻¹ DM)	22.1	2.5	u.
pH	4.7	4.3	4.7
DP (%)	5.4	8.7	13.5

* average of analyse of all Rumex OK 2 silage samples without regard to variant; ** results published by Hejduk and Doležal (2008); *** results published by Juráček et al. (2016); DM% – concentration of dry matter in%; CP – crude protein; CFa – crude fat; CFi – crude fiber; NFE – nitrogen free extract; ADF – acid detergent fiber; NDF – neutral detergent fiber; LA – lactic acid; AA – acetic acid; BA – butyric acid; DP – degree of proteolysis in% (N-NH₃ out of total N); + silage from *Rumex* L. is characteristic with absence of butyric acid; u. – unlisted

Rumex OK 2 silage by animals need to be determined in further experiments.

4 Conclusions

The nutrient content and quality of fermentation process are important factors that determine the quality of fermented feeds. Analysed silages from hybrid Rumex OK 2 (*Rumex patientia* L. × *Rumex tianschanicus* A.Los) mainly in autumn months had high concentration of crude protein from 289 to 339 g.kg⁻¹ DM, which is higher than in alfalfa silages. Concentration of nitrogen free extract and crude fibre in Rumex OK 2 silage was comparable to that of alfalfa silage. Despite the low content of dry matter in wilted Rumex OK 2 plants in silages the butyric acid was not detected. Addition of dried molasses before ensiling increased content of lactic acid and decreased content of acetic acid in silages. Only Rumex OK 2 silages from October and November reached the minimal required content of lactic acid (10 g.kg⁻¹ of original matter). Because high concentration of oxalic acid is Rumex OK 2 silage from September not usable for feeding as single feed. According to concentration of nutrients in analysed samples the autumn Rumex OK 2 silages had better nutritional value than silages made in spring months. Problematic appears the low concentration of dry matter by all silage samples and by samples from September also the high concentration of oxalic acid. Increase of dry matter together with decrease of oxalic acid can be

realized by addition of grassland herbage which has higher content of dry matter and does not contain oxalic acid. Further research about nutritional quality of silage made from Rumex OK 2 together with grassland herbage and its intake by animals is needed.

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